

PROSPECTIVE

Therapeutic Insights from Genomic Studies of Head and Neck Squamous Cell Carcinomas

Peter S. Hammerman^{1,2}, D. Neil Hayes³, and Jennifer R. Grandis^{4,5}

Summary: Large and comprehensive genomic surveys of head and neck squamous cell carcinomas (HNSCC) are now greatly increasing our understanding of the diversity of this disease and the key genomic changes that drive these tumors. The results from these studies are beginning to inform the introduction of novel therapies for patients with HNSCCs. Here, we review some of the key findings from recent genomic studies of head and neck cancers, including the most comprehensive study to date from The Cancer Genome Atlas Network. *Cancer Discov*; 5(3); 239-44. ©2015 AACR.

INTRODUCTION

Head and neck squamous cell carcinomas (HNSCC) are the fifth most common malignancy worldwide and comprise a diverse set of cancers arising in the upper aerodigestive tract mucosa (1). Unlike many other epithelial cancers, the majority of HNSCCs present at a locally advanced stage with cervical lymph node metastases. More than 90% of patients are treated with curative intent using a combination of surgery, radiotherapy, and chemotherapy (2). To date, treatment approaches have been dictated by the anatomic site of the primary tumor, with oral cavity cancers treated primarily with surgical resection and pharyngeal and laryngeal tumors with chemoradiation (3). Although over one half of patients are cured with initial therapy, these treatments are highly morbid, and therapeutic options for individuals who relapse following initial treatment are limited (4). In the absence of information regarding the biologic underpinnings of individual tumors, predictive biomarkers have been lacking to guide therapy in both the initial treatment setting and in the treatment of relapsed/refractory disease. Establishing robust therapeutic biomarkers in HNSCCs has been challenging for several reasons, including the heterogeneity of these tumors that display diversity in terms of their anatomy, clinical characteristics, and in their association with conventional risk factors, such as tobacco and alcohol exposure, as well as with infection with the oncogenic human papillomavirus (HPV) and Epstein-Barr Virus (EBV; refs. 2, 5). Recent large-

scale genomic profiling studies, notably that of The Cancer Genome Atlas (TCGA), have shed light on the molecular underpinnings of the diversity of HNSCCs. We highlight some of the key insights from this and other studies and the implications of these findings on our understanding of HNSCCs and therapeutic approaches.

THE GENOMIC LANDSCAPE OF NON-HPV-DRIVEN HNSCCs

Despite advances in surgical approaches, radiotherapy, and chemotherapies, treatment outcomes for patients with HNSCCs associated with the traditional risk factors of tobacco use, alcohol exposure, or both, remain disappointing as compared with patients with HPV-driven disease (5, 6). This clinical challenge has stimulated genomic studies focusing on this high-risk group of patients with the goal of identifying molecular aberrations that could be targeted to improve clinical outcomes. However, at this time, there are no agents in clinical use in HNSCCs that show enhanced activity associated with a genetic biomarker. Although *EGFR* is overexpressed in HNSCC and has been shown to be associated with reduced survival, EGFR-directed therapies have not been especially efficacious (7, 8). Cetuximab, a monoclonal antibody directed against EGFR, is the only FDA-approved molecularly targeted agent for HNSCC, but response rates to this agent given as monotherapy are approximately 10%, and it remains unclear how to predict the subset of patients most likely to respond to cetuximab or other EGFR-directed therapies, despite a large number of studies addressing this topic (7, 9, 10).

Next-generation sequencing (NGS) studies of non-HPV-driven HNSCCs, including the TCGA project that characterized nearly 250 of these individuals (TCGA Network, Nature, in press), have demonstrated a complex landscape of alterations in gene expression, DNA copy number, somatic mutations, gene rearrangements, and gene promoter methylation (11-15). These tumors are characterized by the near-universal loss of *TP53* and *CKDN2A/RB1* by truncating mutation, deletion, and/or alternative splicing. A summary of somatic alterations in genes regulating a number of key cellular pathways in HPV-negative and HPV-positive HNSCCs is presented in Table 1.

¹Department of Medical Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts. ²Cancer Program, Broad Institute of Harvard and MIT, Cambridge, Massachusetts. ³Department of Hematology/Oncology, University of North Carolina Lineberger Cancer Center, Chapel Hill, North Carolina. ⁴Department of Otolaryngology, University of Pittsburgh and University of Pittsburgh Cancer Institute, Pittsburgh, Pennsylvania. ⁵Department of Pharmacology and Chemical Biology, University of Pittsburgh, Pittsburgh, Pennsylvania.

Corresponding Author: Jennifer R. Grandis, University of Pittsburgh School of Medicine, W912 Biomedical Science Tower, Suite 500, 200 Lothrop Street, Pittsburgh, PA 15213. Phone: 412-383-5405; Fax: 412-647-2080; E-mail: grandisjr@upmc.edu

doi: 10.1158/2159-8290.CD-14-1205

©2015 American Association for Cancer Research.

Downloaded from http://aacrjournals.org/cancerdiscov/article-pdf/5/3/239/1837393/239.pdf by guest on 11 December 2023

Table 1. Comparison of the common genomic pathway alterations and of specific genomic alterations by functional category in HPV-negative versus HPV-positive HNSCCs

Genetic pathways and alterations	HPV-	HPV+
RTK Amplification	>20% with <i>ERBB</i> family, <i>FGFR</i> , <i>INSR</i>	Rare
RTK mutations/fusions	Rare	<i>FGFR2/3</i> mutations in >10%, <i>FGFR3-TACC3</i> fusions
<i>H/K/NRAS</i> , <i>NF1</i>	5%-10%, <i>HRAS</i> may be most common	5%-10%, <i>NF1</i> loss may be more common
<i>PIK3CA</i> amplification/mutation	Common ~30%	Very common >50%
<i>TP53</i>	Genomic loss in nearly all cases	HPV-driven loss
Cell-cycle deregulation	Loss of <i>CDKN2A/RB1</i> by multiple mechanisms in nearly all cases, <i>CCND1/CDK4/CDK6</i> amplification common (30%)	HPV-driven <i>CDKN2A</i> loss, <i>E2F1</i> amplification (20%)
Oxidative stress regulation	Common activation of <i>NFE2L2/KEAP1/CUL3</i> (25%)	Rare
Differentiation	Common loss of <i>NOTCH1/FAT1/AJUBA</i> , <i>TP63</i> gain	<i>NOTCH1</i> loss less common, <i>TP63</i> gain more common
Immune evasion	Uncommon <i>HLA</i> mutations, <10%	<i>HLA</i> , <i>B2M</i> mutations and <i>TRAF3</i> loss

Abbreviation: RTK, receptor tyrosine kinase.

Of note, HPV-negative HNSCCs most closely resemble lung squamous cell carcinomas in terms of their spectra of genomic alterations and contain statistically enriched mutations and copy-number alterations in genes regulating many of the same pathways, in addition to widespread loss of both *TP53* and *CDKN2A/RB1* (16). These include regulation of squamous differentiation (*NOTCH1*, *RIPK4*, *IRF6*, and *TP63*), oxidative stress (*NFE2L2* and *KEAP1*), WNT signaling (*AJUBA* and *FAT1*), immune evasion (*HLA-A*, *B2M*, and *TGFBR2*), and chromatin remodeling (*MLL2* and *NSD1*; TCGA Network, Nature, in press; ref. 15). Although 20% of HPV-negative HNSCCs in the TCGA cohort displayed amplifications of receptor tyrosine kinase (RTK) genes, such as *EGFR*, *ERBB2*, *MET*, and *FGFR1*, there were no recurrent mutations or fusions in RTK genes that have been associated with dramatic responses to small-molecule kinase inhibitors in other tumor types, such as lung adenocarcinoma. One possible exception is oncogenic exon 14 skipping in *MET*, which was reported in two HNSCC cases by TCGA and is found in 4% to 5% of lung adenocarcinoma, and which may be associated with sensitivity to *MET* small-molecule inhibitors.

Mutually exclusive mutations in *RAS* family genes, notably *RHOA*, *KRAS*, and *HRAS*, are present but infrequently found in HNSCC (6% of TCGA cases), though recurrent mutations in *RHOA* at amino acid position 40 are worth noting; however, the biologic significance of these *RHOA* mutations is unclear. Amplification of chromosome 3q, a region containing the *TP63*, *SOX2*, and *PIK3CA* genes, is seen in the majority of both HPV-negative and HPV-positive HNSCCs, and *PIK3CA* mutations are commonly found in both HPV-negative and HPV-positive disease, in agreement with prior studies (11, 14, 15, 17).

HPV-negative HNSCCs arise from a number of anatomic sites, including the larynx, oral cavity, and oropharynx, and generally occur in the setting of heavy alcohol and/or tobacco exposure, or less commonly in patients without these well-established risk factors. The TCGA cohort did not identify any

mutated genes specific to an anatomic site, though the numbers of cases in each of these categories was insufficient to comprehensively address this question. It should be noted that a prior report suggested that *TERT* promoter mutations are enriched in tongue cancers (18). In contrast with lung cancers in which many targetable genomic alterations have been identified specifically in patients who lack exogenous carcinogen exposure in the form of tobacco, two small studies of HNSCCs arising in HPV-negative individuals with minimal tobacco or alcohol histories did not identify any recurrent kinase alterations (19, 20).

HPV-negative HNSCCs demonstrate clear evidence of molecular diversity, as suggested by expression profiling studies that clearly demonstrate diverse biologic subclasses within HPV-negative disease, including a class of tumors without *EGFR* amplification and/or overexpression, previously termed “atypical” HNSCCs, which consist of approximately 20% of HPV-negative cases and the vast majority of HPV-positive HNSCCs (21, 22). An intriguing mutational pattern identified by TCGA was a subset of HPV-negative HNSCCs originating in the oral cavity with few to no copy-number alterations statistically enriched for *HRAS*, *CASP8*, and *PIK3CA* mutations and lack of *TP53* mutation (TCGA Network, Nature, in press).

HPV-POSITIVE HNSCC

It is well accepted that the clinical features of patients with HPV-driven HNSCC are distinct from those of patients with HPV-negative disease. These include distinct sites of origin of the disease (e.g., tonsil and base of tongue), younger age, and improved relapse-free survival following initial definitive treatment. Clinically, an emphasis on protocol development for patients with HPV-driven HNSCCs has consisted largely of de-escalation of standard therapy with a lower dose of radiation, more limited resection, or deintensification of chemotherapy.

Initial NGS studies of HPV-positive patients confirmed that these individuals harbor few genomic alterations in *TP53* and/

or *CDKN2A*, presumably due to the activity of the HPV E6 and E7 viral oncoproteins (11). These studies also suggested that HPV-driven HNSCCs display less genomic complexity as compared with HPV-negative disease, though TCGA and other more recent cohorts did not confirm this finding, perhaps due to tobacco use in the HPV-positive individuals in these studies. This possibility is supported by the prevalence of both the virally associated Tp* Cp(A/C/T) substitution mutation in the HPV-positive individuals as well as CpG transversions, a mutation class typically associated with smoking. A major limitation in most studies reported to date has been the relatively small numbers of characterized HPV-positive tumors.

HPV-driven HNSCCs are distinct from HPV-negative disease in that they lack focal RTK amplifications but do display a higher rate of focal *PIK3CA* amplification and mutation. *PIK3CA* alterations have been reported as therapeutic biomarkers in this patient population based on cell line and patient-derived xenograft studies (14). HPV-associated HNSCCs also demonstrate enrichment for copy-number gains in *TRAF3* and *E2F1* and a lack of *CCND1* amplification when compared with HPV-negative disease.

HPV-driven cancers display both mutations and fusions in the *FGFR3* gene, with mutations at position 249 reported at 14% in one study of 50 cases of locoregionally advanced disease, and *FGFR3-TACC3* fusions have been reported in multiple cases by the TCGA and other groups (15, 23). These two *FGFR3* alterations have been associated with therapeutic response to FGFR small-molecule inhibitors in preclinical (24) and clinical studies (25, 26). TCGA did not detect any genes displaying statistical enrichment for mutation in HPV-positive individuals as compared with HPV-negative individuals, though *B2M* truncating mutations most closely approached significance. In addition to *FGFR3* mutation, other studies have identified the RNA helicase *DDX3X*, which is mutated in medulloblastoma and is a regulator of β -catenin, as a gene more commonly altered in HPV-positive individuals (15). HPV-positive disease shares many common altered genes and pathways with HPV-negative HNSCC (e.g., *NOTCH*, *MLLs*, *RAS*, and *WNT*; ref. 15).

In addition to the genomic context in which HPV resides, it has become increasingly clear that the virus itself plays an important role in oncogenesis beyond expression of E6 and E7 (27, 28). Studies of the interaction of HPV with the human genome in HNSCCs have shown that HPV may be present in integrated or nonintegrated forms, may be associated with the presence or absence of ongoing E6/E7 expression, and also may be present in extrachromosomal elements. Sites of HPV integration are nonrandom, occurring in gene- and microRNA-rich areas of the genome as well as in sites that are commonly associated with somatic copy number alterations. HPV integration can have a profound impact on local gene structure and function and result in high-level amplifications, gene disruptions, alternative splicing, novel gene fusions, and changes in global promoter methylation and transcription. An intriguing finding in the field of HPV integration is recurrent disruptive integration in the *RAD51* gene, perhaps facilitating further HPV integration by hindering DNA repair (29). Although the study of host genome-HPV interactions is still maturing, it is clear that the role of HPV extends beyond the production of E6 and E7 in HNSCCs.

A very important feature of the TCGA data and other cohorts is the demonstration that HPV may be detected in HNSCCs using a number of methods, including mass spectrometry and massively parallel RNA and DNA sequencing, and that these methods are far more sensitive than those currently applied in the clinic. These methods identify patients without conventional clinical features (e.g., larynx cancers and p16-negative) who appear to have HPV-driven disease. As NGS methods are increasingly applied in the clinic, it will be a challenge moving forward to further define the sensitivity and specificity of these newer methods for HPV detection and if there is any significance related to quantitative differences in the amount of HPV detected in a given tumor. Although p16 immunohistochemistry is a simple assay, these recent studies suggest that it should not be regarded as an appropriate surrogate for direct assessment of HPV status. p16 assessment alone will misclassify individuals in whom HPV is present in the absence of E6/E7 expression, and more importantly, will overlook HPV in tumors in which both HPV is present and p16 is lost by an independent mechanism, an especially relevant issue in patients with both HPV and a tobacco history. For studies moving forward, it will be critical to accurately determine HPV status by direct measurement of HPV to avoid systematic errors in patient classification and stratification.

GENOMICS TO TARGETED THERAPEUTICS

The application of targeted therapeutics in HNSCCs has been disappointing to date as compared with other cancer types. This has been due, in part, to the slower development of therapeutic biomarkers and a lack of understanding of the genomic landscape of these diseases. As noted above, cetuximab is the only targeted agent approved for HNSCC; its use in the metastatic setting is associated with a low response rate of 10% to 15%, and most studies have failed to find an association between EGFR expression and/or gene amplification with response to EGFR inhibitors, including cetuximab, in HNSCC cohorts. There are no prospectively validated biomarkers to enable the selection of patients for EGFR-directed therapy.

Before the TCGA and other recent studies, earlier reports noted the prevalence of a number of genomic alterations in HNSCCs, which are associated with therapeutic response to targeted agents in other cancer types. These include *EGFR* mutations and *ALK* and *ROS1* fusions. It is now clear that these events are extremely rare in HNSCC, if present at all, and that the therapeutic opportunities in HNSCCs more closely resemble squamous cell cancers from other tissue types. A few specific examples are discussed below.

Fibroblast Growth Factor Receptors

FGFRs have been shown to be activated by amplification, mutation, and translocation in a wide range of cancer types. In HNSCC, *FGFR1* amplifications are found in HPV-negative patients at a rate of approximately 10% and appear to be enriched in non-oro-pharynx tumors. *FGFR1* amplification has been associated with therapeutic response to FGFR tyrosine kinase inhibitors (TKI) in lung squamous cell cancers, though response rates represent only a modest improvement

as compared with chemotherapy (30–32). Multiple explanations for this disappointing result have been reported, including the presence of co-mutations activating the *RAS-MAPK* pathway, a lack of correlation between *FGFR1* amplification and expression or activation of the protein, and difficulty in standardizing assays for detection of amplification by FISH or NGS methods. Several early-phase clinical trials are ongoing or planned in patients with HNSCC with *FGFR1* amplifications who have relapsed/refractory HNSCC (e.g., NCT01962532, NCT01004224, and NCT01948297), though no public data have been reported on efficacy. Given that FGFR inhibitors appear to be well tolerated and may also be radiosensitizers, the combination of these agents in the curative treatment setting with the current standard of care may be reasonable in high-risk HPV-negative patients.

FGFR2 and *FGFR3* mutations and *FGFR3-TACC3* fusions are of particular interest, as these genetic lesions have been associated with dramatic responses to FGFR TKIs in preclinical models and in early-phase clinical settings, including a case report of a dramatic response to pazopanib in a patient with an *FGFR2*-mutated tongue cancer (24). In contrast to *FGFR1* amplification, *FGFR2/3* mutations and *FGFR3* fusions appear to occur largely in HPV-positive individuals at a prevalence of 10% to 20%, and clinical trials are currently targeting this patient population. However, given that these trials are focusing on patients with relapsed/refractory disease, they may encounter difficulty with accrual, as the number of HPV-positive patients who are candidates for such studies is small.

PI3K-AKT Pathway

PIK3CA is commonly amplified and/or mutated in patients with HNSCCs (37% of cases in the TCGA), and *PIK3CA* alterations are enriched in HPV-positive patients. If one examines the PI3K-AKT-mTOR pathway in detail, more than one half of patients with HNSCC have a somatic alteration that can activate this pathway. As such, there is tremendous interest in developing small-molecule inhibitors of components of this pathway for individuals with HNSCC, and ample preclinical data suggest that this may be an effective therapeutic strategy, though it should be noted that the activity of PI3K inhibitors as monotherapy in lung squamous cell cancers in patients with *PIK3CA* or *PTEN* mutations has been disappointing. However, initial data have been more encouraging when combining these agents with chemotherapy or other targeted agents.

Clinical concepts moving forward include both recruiting patients with relapsed/refractory disease with PI3K pathway lesions as well as using these agents in “window of opportunity” trials in the upfront setting or in combination with chemoradiotherapy (e.g., NCT01816984, NCT01195922, NCT01852292, and NCT01133678). Preclinical data have shown that inhibition of the PI3K pathway may sensitize cancer cells to radiation and that PI3K inhibitors may be most efficacious as radiosensitizers in patients with *NFE2L2* or *KEAP1* mutations, genomic events commonly seen in high-risk HPV-negative individuals (33). A clinical trial in this high-risk population is now ongoing based on these preclinical data (NCT02113878).

Cyclin-Dependent Kinases

In the TCGA study, 32% of HNSCCs displayed an amplification or mutation of *CCND1*, *CDK4*, or *CDK6*, with the

majority of these alterations found in HPV-negative patients. It should be noted that the genomic region on chromosome 11 containing the *CCND1* locus also contains other cancer-related genes such as *FADD*, and it is not clear that *CCND1* is the focus of amplification in all cases with chromosome 11q13 amplification. However, impressive early clinical data in breast cancer have suggested that CDK4/6 inhibitors may be effective in patient cohorts with high rates of *CCND1* amplification and that these agents are well tolerated as both single agents and in combination with other therapies. Clinical trials are moving forward at this time in other cancer types with frequent alterations of *CCND1/CDK4/CDK6*, and a subset of these trials will include patients with relapsed/refractory HNSCC (NCT02101034). Many HPV-negative patients with *CCND1/CDK4/CDK6* amplifications also harbor RTK amplifications, suggesting that combination strategies may be needed in this setting.

Immunotherapy

Immunotherapy approaches have garnered a great deal of excitement in the oncology community based on the early clinical success of immune checkpoint inhibitors in melanoma, renal cell carcinoma, and lung cancer. Trials of immunotherapeutic agents are ongoing in HNSCC in both the initial treatment setting with ipilimumab and for recurrent disease with agents targeting the PD1:PDL1 checkpoint and other immune effectors (e.g., NCT01860430 and NCT01935921), with promising data presented recently with pembrolizumab in relapsed/refractory disease (34). *PDL1* (*CD274*) expression is a biomarker reported by some groups to enrich for response to blockade of the PD1:PDL1 checkpoint, and it has been reported in other cancer types, including lymphoma and gastric cancer, that *PDL1* expression is associated with virally induced cancers (35). Although there are several reports on this topic in HNSCCs, with variable results, RNA sequencing data from the TCGA are consistent with higher levels of *PDL1* expression in HPV-negative individuals. However, it should be noted that these samples contain both tumor and stroma and it is still unclear what the optimal methods are for *PDL1* measurement in the context of patient stratification. In addition to variable expression of *PDL1*, HNSCCs display a wide range of somatic alterations in genes involved in antigen presentation, inflammation, and immune evasion, including *HLA-A*, *B2M*, *TGFBR2*, and *TRAF3*. Although the mechanisms governing immune evasion in HNSCCs remain poorly understood, it is likely that somatic alterations in these and other genes are likely to play a key role in immune surveillance of HNSCCs and may affect the responsiveness of cancers to specific immunotherapeutic approaches. One particular pathway of interest in this regard is PI3K, given reports in other cancer types that it may be associated with response to PD1:PDL1 checkpoint inhibitor therapy (36).

Other Targets and Strategies

A number of additional therapeutic targets have been proposed for HNSCC based on genomic discovery studies and preclinical models. HNSCCs frequently display hyperactivation of STAT3 via a variety of mechanisms, and STAT3 pathway inhibitors are currently being explored both in preclinical models and in early-phase clinical trials. HNSCCs

display frequent deregulation of pro- and antiapoptotic genes such as *CASP8*, and cell line studies have suggested that inhibitors of BCL2 family proteins may demonstrate activity against HNSCCs. HNSCCs also commonly display concurrent amplification of two or more putative “drivers,” and preclinical studies have demonstrated synergy in combining inhibitors in cell lines that demonstrate multiple activated kinase pathways, such as concurrent EGFR and FGFR pathway activation. Novel EGFR-targeting strategies with more potent anti-EGFR TKIs and antibodies are moving forward in HNSCCs, as are efforts to better define the subset of patients most likely to benefit from anti-EGFR therapy.

CONCLUSIONS AND RECOMMENDATIONS

HNSCCs are less common than other cancer types in which substantial strides in biomarker-based clinical trials have been made, and the number of clinical trials currently available for individuals with HNSCCs is approximately one half the number of trials for patients with lung or breast cancers. Furthermore, there are no genetic tests routinely incorporated into the management of HNSCC, and patient stratification is largely done based on clinical features and HPV status. With the widespread incorporation of NGS diagnostics into the routine care of patients with a wide array of cancer types, it will be important to consider how this information can be used to improve the efficacy of therapies for HNSCCs, and here we propose a few possible approaches.

First, the molecular alterations identified in the HNSCC TCGA project and other cohorts are not unique and are shared with a number of other epithelial cancer types, most notably lung squamous cell carcinoma. We suggest that clinical trials of novel agents in lung SCCs also include patients with HNSCCs given this overlap, and that “basket trial” approaches be considered across these disease types. Natural candidates for this approach include agents targeting FGFRs, PI3K/AKT, cyclin-dependent kinases (CDK), and immunotherapies. Second, HNSCC would seem to be an ideal cancer type for “window-of-opportunity” trials, given the relative accessibility of tumors to sample and the widespread use of surgical resection as a standard curative approach. In this study approach, a novel agent could be deployed before definitive therapy to assess its efficacy with biopsy samples taken before and during treatment to identify biomarkers of response and resistance. Third, given that we have now defined a number of potential targets in HNSCC, the use of these agents in the definitive setting with analysis of tissue before and after therapy should be conducted so that we might better define cohorts of patients for specific therapeutic approaches in which novel agents may be used in combination with chemoradiotherapy in the initial or adjuvant setting. Finally, it will be critical for the HNSCC community of surgeons, radiation oncologists, and medical oncologists to appreciate the value of a molecular understanding of HNSCCs to facilitate moving the field forward in the era of genomic medicine.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Grant Support

P.S. Hammerman is supported by NCI grant K08 CA163677.

Published OnlineFirst February 2, 2015.

REFERENCES

- Vokes EE, Weichselbaum RR, Lippman SM, Hong WK. Head and neck cancer. *N Engl J Med* 1993;328:184–94.
- Ang KK, Chen A, Curran WJ Jr, Garden AS, Harari PM, Murphy BA, et al. Head and neck carcinoma in the United States: first comprehensive report of the longitudinal oncology registry of head and neck carcinoma (LORHAN). *Cancer* 2012;118:5783–92.
- Edge SB, Compton CC. The American Joint Committee on Cancer: the 7th edition of the AJCC cancer staging manual and the future of TNM. *Ann Surg Oncol* 2010;17:1471–4.
- Janot F, de Raucourt D, Benhamou E, Ferron C, Dolivet G, Bensadoun RJ, et al. Randomized trial of postoperative irradiation combined with chemotherapy after salvage surgery compared with salvage surgery alone in head and neck carcinoma. *J Clin Oncol* 2008;26:5518–23.
- 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;363:24–35.
- Chaturvedi AK, Engels EA, Pfeiffer RM, Hernandez BY, Xiao W, Kim E, et al. Human papillomavirus and rising oropharyngeal cancer incidence in the United States. *J Clin Oncol* 2011;29:4294–301.
- 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;354:567–78.
- Chung CH, Ely K, McGavran L, Varella-Garcia M, Parker J, Parker N, et al. Increased epidermal growth factor receptor gene copy number is associated with poor prognosis in head and neck squamous cell carcinomas. *J Clin Oncol* 2006;24:4170–6.
- Cohen EE. Role of epidermal growth factor receptor pathway-targeted therapy in patients with recurrent and/or metastatic squamous cell carcinoma of the head and neck. *J Clin Oncol* 2006;24:2659–65.
- Vokes EE, Seiwert TY. EGFR-directed treatments in SCCHN. *Lancet Oncol* 2013;14:672–3.
- Stransky N, Egloff AM, Tward AD, Kostic AD, Cibulskis K, Sivachenko A, et al. The mutational landscape of head and neck squamous cell carcinoma. *Science* 2011;333:1157–60.
- Agrawal N, Frederick MJ, Pickering CR, Bettgowda C, Chang K, Li RJ, et al. Exome sequencing of head and neck squamous cell carcinoma reveals inactivating mutations in NOTCH1. *Science* 2011;333:1154–7.
- Pickering CR, Zhang J, Yoo SY, Bengtsson L, Moorthy S, Neskey DM, et al. Integrative genomic characterization of oral squamous cell carcinoma identifies frequent somatic drivers. *Cancer Discov* 2013;3:770–81.
- Lui VW, Hedberg ML, Li H, Vangara BS, Pendleton K, Zeng Y, et al. Frequent mutation of the PI3K pathway in head and neck cancer defines predictive biomarkers. *Cancer Discov* 2013;3:761–9.
- Seiwert TY, Zuo Z, Keck MK, Khattri A, Pedamallu CS, Stricker TP, et al. Integrative and comparative genomic analysis of HPV-positive and HPV-negative head and neck squamous cell carcinomas. *Clin Cancer Res*. 2014 Jul 23. [Epub ahead of print].
- TCGA. Comprehensive genomic characterization of squamous cell lung cancers. *Nature* 2012;489:519–25.
- Smeets SJ, Braakhuis BJ, Abbas S, Snijders PJ, Ylstra B, van de Wiel MA, et al. Genome-wide DNA copy number alterations in head and neck squamous cell carcinomas with or without oncogene-expressing human papillomavirus. *Oncogene* 2006;25:2558–64.
- Killela PJ, Reitman ZJ, Jiao Y, Bettgowda C, Agrawal N, Diaz LA Jr, et al. TERT promoter mutations occur frequently in gliomas and a subset of tumors derived from cells with low rates of self-renewal. *Proc Natl Acad Sci U S A* 2013;110:6021–6.

19. Li R, Faden DL, Fakhry C, Langelier C, Jiao Y, Wang Y, et al. Clinical, genomic and metagenomic characterization of oral tongue squamous cell carcinoma in patients who do not smoke. *Head Neck*. 2014 Jun 21. [Epub ahead of print].
20. Pickering CR, Zhang J, Neskey DM, Zhao M, Jasser SA, Wang J, et al. Squamous cell carcinoma of the oral tongue in young non-smokers is genomically similar to tumors in older smokers. *Clin Cancer Res* 2014;20:3842-8.
21. Walter V, Yin X, Wilkerson MD, Cabanski CR, Zhao N, Du Y, et al. Molecular subtypes in head and neck cancer exhibit distinct patterns of chromosomal gain and loss of canonical cancer genes. *PLoS ONE* 2013;8:e56823.
22. Chung CH, Parker JS, Karaca G, Wu J, Funkhouser WK, Moore D, et al. Molecular classification of head and neck squamous cell carcinomas using patterns of gene expression. *Cancer Cell* 2004;5: 489-500.
23. Wu YM, Su F, Kalyana-Sundaram S, Khazanov N, Ateeq B, Cao X, et al. Identification of targetable FGFR gene fusions in diverse cancers. *Cancer Discov* 2013;3:636-47.
24. Liao RG, Jung J, Tchaicha J, Wilkerson MD, Sivachenko A, Liu Q, et al. Inhibitor-sensitive FGFR2 and FGFR3 mutations in lung squamous cell carcinoma. *Cancer Res* 2013;73:5195-205.
25. Nogova L, Sequist LV, Cassier PA, Hidalgo M, Delord J-P, Schuler MH, et al. Targeting FGFR1-amplified lung squamous cell carcinoma with the selective pan-FGFR inhibitor BGJ398. *J Clin Oncol* 32:5s, 2014 (suppl; abstr 8034).
26. Bahleda R, Dienstmann R, Adamo B, Gazzah A, Infante JR, Zhong B, et al. Phase 1 study of JNJ-42756493, a pan-fibroblast growth factor receptor (FGFR) inhibitor, in patients with advanced solid tumors. *J Clin Oncol* 32:5s, 2014 (suppl; abstr 2501).
27. Akagi K, Li J, Broutian TR, Padilla-Nash H, Xiao W, Jiang B, et al. Genome-wide analysis of HPV integration in human cancers reveals recurrent, focal genomic instability. *Genome Res* 2014;24: 185-99.
28. Parfenov M, Pedamallu CS, Gehlenborg N, Freeman SS, Danilova L, Bristow CA, et al. Characterization of HPV and host genome interactions in primary head and neck cancers. *Proc Natl Acad Sci U S A* 2014;111:15544-9.
29. Ojesina AI, Lichtenstein L, Freeman SS, Pedamallu CS, Imaz-Rosshandler I, Pugh TJ, et al. Landscape of genomic alterations in cervical carcinomas. *Nature* 2014;506:371-5.
30. Weiss J, Sos ML, Seidel D, Peifer M, Zander T, Heuckmann JM, et al. Frequent and focal FGFR1 amplification associates with therapeutically tractable FGFR1 dependency in squamous cell lung cancer. *Sci Transl Med* 2010;2:62ra93.
31. Dutt A, Ramos AH, Hammerman PS, Mermel C, Cho J, Sharifnia T, et al. Inhibitor-sensitive FGFR1 amplification in human non-small cell lung cancer. *PLoS ONE* 2011;6:e20351.
32. Wolf J, LoRusso PM, Camidge RD, Perez JM, Taberno J, Hidalgo M, et al. A phase I dose escalation study of NVP-BGJ398, a selective pan FGFR inhibitor in genetically preselected advanced solid tumors. In: Proceedings of the 103rd Annual Meeting of the American Association for Cancer Research; 2012 Mar 31-Apr 4; Chicago, IL. Philadelphia (PA): AACR; *Cancer Res* 2012;72:Abstract nr LB-122.
33. Abazeed ME, Adams DJ, Hurov KE, Tamayo P, Creighton CJ, Sonkin D, et al. Integrative radiogenomic profiling of squamous cell lung cancer. *Cancer Res* 2013;73:6289-98.
34. Seiwert TY, Burtneess B, Weiss J, Gluck I, Eder JP, Pai SI, et al. A phase Ib study of MK-3475 in patients with human papillomavirus (HPV)-associated and non-HPV-associated head and neck (H/N) cancer. *J Clin Oncol* 32:5s, 2014 (suppl; abstr 6011).
35. Chen BJ, Chapuy B, Ouyang J, Sun HH, Roemer MG, Xu ML, et al. PD-L1 expression is characteristic of a subset of aggressive B-cell lymphomas and virus-associated malignancies. *Clin Cancer Res* 2013;19:3462-73.
36. Parsa AT, Waldron JS, Panner A, Crane CA, Parney IF, Barry JJ, et al. Loss of tumor suppressor PTEN function increases B7-H1 expression and immunoresistance in glioma. *Nat Med* 2007;13:84-8.