# Clinicopathologic Features of Advanced Squamous NSCLC

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**ABSTRACT**

Lung cancer remains the leading cause of cancer-related death worldwide. NSCLC accounts for more than 85% of all lung cancers, and the prognosis for advanced-stage disease is typically poor. In recent years, the importance of histologic subtypes of NSCLC has been recognized, and the distinction between squamous and other NSCLC histologic subtypes is now critical to patient management. Squamous cell lung cancer (sqCLC) represents approximately 25% to 30% of NSCLC. The prognosis for patients with advanced NSCLC is poorer for those with sqCLC than for those with adenocarcinoma. This is partly due to a number of clinical characteristics that distinguish sqCLC from other NSCLC histologic subtypes, such as smoking history, comorbid diseases, age, and molecular profile. Together, these factors make sqCLC an especially challenging disease to manage. Herein, we review some of the key clinicopathologic features of sqCLC. Understanding these features to optimally address many of the unique therapeutic challenges of this disease is likely to be central to ultimately improving outcomes for patients with squamous NSCLC.



*Keywords:* NSCLC; Squamous cell lung cancer; Histologic Subtype; Etiology

## Introduction

Lung cancer remains the leading cause of cancer-related death worldwide, accounting for 19.4% of cancer-related deaths and 1.59 million deaths annually.[1] Most patients present with locally advanced or metastatic disease, as until recently there had been no effective screening measures and metastases occur early in the natural history of lung cancer. Consequently, the prognosis for advanced-stage disease is poor, with an overall 5-year survival rate of 17% in the United States.[2] Tobacco consumption is well recognized as the major risk factor for lung cancer, and because of the ongoing tobacco epidemic in many parts of the world, the incidence of lung cancer and related mortality is expected to rise over the coming decades.[1] As a result, lung cancer will remain a considerable global burden and a major focus of research.

NSCLC accounts for more than 85% of all lung cancers.[2] In the past, advanced NSCLC was viewed as a single disease entity and managed as such, with systemic therapies the mainstay of treatment.[3] More recently, the subclassification of NSCLC into adenocarcinoma, large cell carcinoma, and squamous cell carcinoma has gained importance because of the therapeutic implications of underlying histologic subtype.[4,5] Several currently available treatment options developed in the past decade for NSCLC (e.g., bevacizumab, pemetrexed, and nintedanib) are not approved or are not suitable for use in patients with the squamous histologic subtype owing to toxicity and/or efficacy issues.[6,7] Unfortunately, recent improvements in patient outcomes have largely been confined to patients with adenocarcinoma.[6–9] Indeed, a comparison of outcomes in recent clinical trials reveals that the median survival of patients with an advanced case of the squamous histologic subtype who are receiving first-line therapy was approximately 30% shorter than that of patients with other NSCLC subtypes.[10–14]

In this context, this article reviews some of the key characteristics of squamous cell lung cancer (sqCLC), including pathologic features, clinical features, patient characteristics, epidemiology, and molecular profile, to bring into focus the specific challenges and opportunities relating to this disease.

## Pathologic and Histologic Subtype

In the vast majority of patients, advanced lung cancer is diagnosed on the basis of small biopsy and cytologic specimens rather than resection specimens; as a result, the 2015 WHO classification of lung cancers provides criteria for both scenarios. In the case of small biopsy and cytologic samples, the main diagnostic categories are adenocarcinoma, squamous cell carcinoma, and NSCLC not otherwise specified (Table 1).[15] The classification of NSCLC according to small biopsy and cytologic samples in the WHO guidelines is based closely on the 2011 International Association for the Study of Lung Cancer, American Thoracic Society, and European Respiratory Society classification.[4] Given the importance of specific histologic diagnosis in NSCLC because of its impact on subsequent genetic testing and ultimately treatment, the classification also aims to reduce the number of patients in whom NSCLC not otherwise specified is diagnosed, not least by the use of terminology in which one subtype is favored.

The microscopic features of sqCLC can help to distinguish it from adenocarcinoma (Table 2). Squamous cell carcinoma is characterized histologically by areas of keratinization and the presence of intercellular bridges, whereas adenocarcinoma typically shows gland formation and papillary structures on histologic examination.[9,16] The presence of morphological components of both adenocarcinoma and sqCLC histologic subtypes, with each representing at least 10% of the tumor cells, could mean adenosquamous carcinoma.[15]

In addition to morphological examination, immunohistochemical analysis can be used to help identify the histologic subtype. Thyroid transcription factor-1, p63, and p40 are the principal markers used currently for subtyping NSCLC: thyroid transcription factor-1 as a

**Table 1.** 2015 WHO Classification of NSCLC for Small Biopsy and Cytologic Specimens

| Terminology | Morphologic Pattern and Staining |
|---|---|
| Adenocarcinoma | |
| Adenocarcinoma with lepidic pattern<br>Invasive mucinous adenocarcinoma<br>Adenocarcinoma with colloid features<br>Adenocarcinoma with fetal features<br>Adenocarcinoma with enteric features | Adenocarcinoma morphologic patterns clearly present |
| NSCLC, favor adenocarcinoma | Morphologic patterns not present, but staining supports adenocarcinoma (e.g., thyroid transcription factor-1) |
| Squamous cell carcinoma | Squamous cell morphologic patterns clearly present |
| NSCLC, favor squamous cell carcinoma | Morphologic patterns not present, but staining supports squamous cell carcinoma (e.g., p40) |
| NSCLC, not otherwise specified | No clear adenocarcinoma or squamous morphologic patterns or staining pattern<br>Squamous cell and adenocarcinoma morphologic patterns present (adenosquamous) |

*Note:* This table relates to diagnoses made on the basis of small biopsy and cytologic samples.

marker for adenocarcinoma, and p63 and p40 as markers for sqCLC.[17] Studies have shown that p63 has an extremely high sensitivity for sqCLC[18]; however, a main limitation of p63 is low specificity owing to its reactivity in adenocarcinoma, as well as in other tumor types, particularly lymphomas.[19] p40 is a relatively new predictive marker for sqCLC that has demonstrated sensitivity comparable to that of p63 but superior specificity.[20] Cytokeratin 5/6 may also be a useful marker for sqCLC, particularly in cases of poorly differentiated or undifferentiated NSCLC.[21] Adenosquamous carcinoma may also be diagnosed if some populations of cells immunostain for thyroid transcription factor-1 and others immunostain for squamous markers described later in this article.[15] It should be noted that in some cases, it may be difficult to distinguish metastatic squamous cell cancer originating in a distant site from a true primary sqCLC solely on the basis of immunohistochemical analysis or microscopy.[22]

## Clinical Features

NSCLC histologic subtype correlates with the site of origin. SqCLC is usually centrally located, typically beginning in early versions of flat cells that line the inside of the lung airways and arising in the proximal bronchi. Conversely, adenocarcinoma is usually located peripherally and related to surface alveolar epithelium or bronchial mucosal glands.[9,23]

As a consequence of its central location, sqCLC is more likely to invade larger blood vessels and vital structures in the mediastinum and is more likely to cause bronchial obstruction.[9,23] Furthermore, cases of peripherally located sqCLC usually present once the tumor has grown larger and invaded the chest wall. In addition, the squamous histologic subtype is associated with tumor cavitation, with more than 80% of cavitating tumors identified as sqCLC.[24,25] It is also noteworthy that, on average, sqCLC tumors grow more quickly than adenocarcinoma tumors: in a study of 63 patients with NSCLC in the United States, the median doubling time was 160 days for sqCLC compared with 387 days for adenocarcinoma/bronchoalveolar carcinoma ($p = 0.0031$).[26]

These clinical features, particularly cavitation, may explain why patients with sqCLC are at increased risk for potentially fatal pulmonary hemorrhage compared with

**Table 2.** Key Cellular Characteristics of Squamous NSCLC and Adenocarcinoma

| Characteristics | Squamous Cell Lung Cancer | Adenocarcinoma |
|---|---|---|
| Cell arrangements | Cells appear in cohesive aggregates, usually in flat sheets, and may appear as irregular shapes (e.g., spindle-shaped and tadpole-shaped cells) | Single cells or arranged in three-dimensional clusters; borders of cell clusters may be sharply defined |
| Cytoplasmic features | Abundant cytoplasm in irregularly shaped cells | Relatively abundant cytoplasm, typically more translucent than squamous cell carcinoma. Cytoplasm can range from distinctly homogeneous to granular to foamy because of the presence of vacuoles |
| Nuclear features | Central, irregular hyperchromatic nuclei, exhibiting one or more small nucleoli | Single, eccentric, and round to oval with relatively smooth contours and minimal nuclear irregularity. Chromatin tends to be finely granular and evenly dispersed |

patients with adenocarcinoma.[25,27] In one retrospective cohort of patients who had died because of lung cancer (n = 100), pulmonary hemorrhage was significantly correlated with sqCLC, with 50% (six of 12) of those who died as a direct result of pulmonary hemorrhage having sqCLC.[25] In a retrospective study of 583 patients with advanced NSCLC, all patients who had baseline cavitation (n = 38) and who experienced a fatal pulmonary hemorrhage (n = 3) had sqCLC.[27] In the same study, multivariate analysis found baseline major cavitation (OR = 17.9) and the squamous histologic subtype (OR = 5.5) to be significant independent risk factors for fatal pulmonary hemorrhage.[27]

## Etiology

Tobacco smoking is the underlying cause of more than 80% of all lung cancers and is the major risk factor for sqCLC, as well as other NSCLC subtypes.[28] Importantly, smoking cessation can substantially reduce the risk for development of lung cancer. In a population-based study, there was an inverse relationship between the risk for development of lung cancer and the time since smoking cessation, with a cumulative risk for death due to lung cancer by age 75 years of 15.9% for men who continued to smoke cigarettes, compared with 9.9%, 6.0%, 3.0%, and 1.7% for those who stopped at age 60, 50, 40, and 30 years, respectively.[29] Smoking cessation is also an important goal in patients in whom lung cancer has already been diagnosed, as it is associated with increased efficacy of treatments and improved quality of life.[30–33] In a meta-analysis, overall mortality in active smokers after surgery for stage I to stage IIIA NSCLC was 2.94 times higher than in nonsmokers.[33] In a retrospective study of patients with stage I or II NSCLC, active smokers had a 2-year survival rate of 41% after radiotherapy compared with 56% for patients who had ceased smoking or who had never smoked.[31] Smoking has also been reported as a negative predictor of response to chemotherapy in patients with advanced lung cancer and can also affect the efficacy of targeted therapy in this setting.[30,32] Hence, not only is smoking central to lung cancer development but smoking cessation is also critical to disease prevention and improved outcomes.

Active tobacco smoking has a stronger association with squamous disease than with adenocarcinoma.[34,35] In a large pooled analysis of case-control studies (13,169 patient cases), the ORs for squamous NSCLC in current or former smokers versus in those who had never smoked were 45.6 and 14.7, respectively, compared with 10.8 and 4.2 for adenocarcinoma (Fig. 1).[35] In addition, in a population-based study of 20,561 patients in Poland in whom lung cancer was diagnosed between 1995 and 1998, the patients with adenocarcinoma smoked less intensely than the patients with squamous NSCLC: 31 versus 36 packs per year in women and 38 versus 42 packs per year in men.[36]

In addition to the etiologic association with lung cancer, smoking has been formally established as a causative link to 21 diseases, including chronic obstructive pulmonary disease and cardiovascular disease. Moreover, pooled data from five cohort studies (n = 954,029 patients age ≥55 years) recently showed that a significant portion of excess mortality among smokers was due to causes not previously recognized as being related to smoking, such as renal failure, hypertensive heart disease, and intestinal ischemia.[37] The additional potential disease burden due to smoking is a contributor to poor outcomes in patients with sqCLC, as well as in patients with other histologic subtypes of lung cancer, and emphasizes the importance of cessation.

Smoking is declining in many Western countries but is currently peaking in other parts of the world, such as China, Eastern Europe, and several countries in Africa.[38,39] At present, the highest incidence rates of lung cancer are still seen in North America and northern

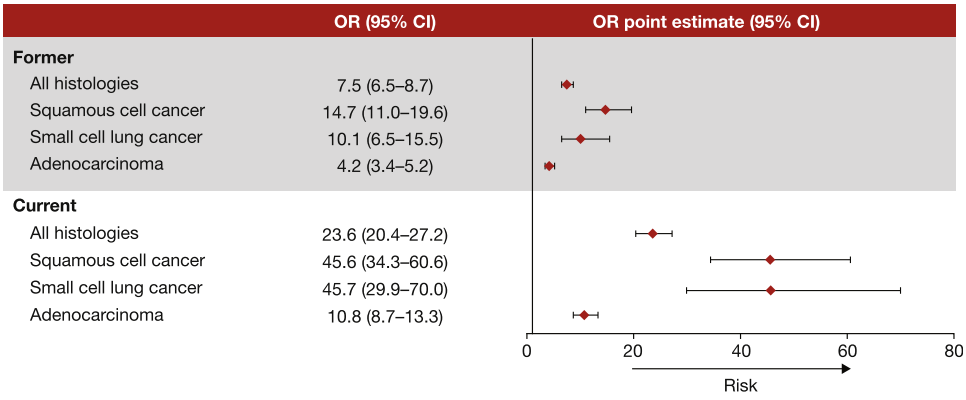


**Figure 1.** Relative risk for lung cancer based on smoking status in men. CI, confidence interval. Adapted with permission from Pesch et al.[35] 

Europe, where smoking rates have historically been high; however, the geographic distribution of lung cancer cases is expected to change, reflecting trends in tobacco consumption.

Given the prominent role of smoking in lung cancer, screening of at-risk populations has long been suggested. The National Lung Screening Trial, which was launched in 2002, found that annual screening using low-dose helical computed tomography scans of individuals with a 30–pack-year or longer history of cigarette smoking (or smoking cessation $\leq$15 years if a former smoker) was associated with a 20% relative reduction in lung cancer mortality.[40] Screening of individuals age 55 to 80 years who meet this criterion is now recommended by the U.S. Preventive Services Task Force.[41] In the National Lung Screening Trial, adenocarcinoma was the most common histologic subtype detected by low-dose helical computed tomography (39.9%), with 21.1% of detected tumors being sqCLC.[40] A post hoc analysis of this trial reported that the survival benefit of low-dose helical computed tomography versus chest radiography observed for adenocarcinoma was not observed for sqCLC (mortality risk ratio = 0.75, 95% confidence interval: 0.60–0.94 for adenocarcinoma versus 1.23, 95% confidence interval: 0.92–1.64 for sqCLC).[42] Screening trials are ongoing in many parts of the world to further evaluate the benefits, harm, and cost-effectiveness of lung cancer screening.[43,44]

## Epidemiology

In recent decades, the predominant NSCLC histologic type has shifted from sqCLC to adenocarcinoma, especially in developed countries.[45] This may be due in part to the shift to low-tar filter cigarettes, in which small particles are inhaled more deeply into the periphery of the lung.[46] Importantly, however, sqCLC still accounts for 25% to 30% of lung cancers,[47] thereby representing approximately half a million cases in 2012.[1]

Although the histologic distribution of NSCLC differs geographically, in relative terms sqCLC is more common in men. On the basis of a review of international sources of cases diagnosed between 1998 and 2002, sqCLC accounted for 27% to 46% of NSCLC cases in men, compared with 11% to 23% in women.[38] Indeed, according to these data, in a number of countries including the United Kingdom, sqCLC was the most common NSCLC histologic subtype in men (Fig. 2).[38] In contrast, adenocarcinoma is more than twice as common as any other histologic subtype of lung cancer in women.[47]

Male sex has been established as a negative prognostic factor for NSCLC, as survival rates for women have been shown to be significantly better than those for men.[48] In a cohort study of 4618 patients in whom NSCLC was diagnosed between 1997 and 2002 in the United States, the 1- and 5-year survival rates in women were 60% and 19%, respectively, compared with 51% and 15% in men, respectively.[49]

NSCLC, including sqCLC, is associated with increased age. The median age at diagnosis for patients in the United States with lung cancer diagnosed between 2008 and 2012 was 70 years.[50] Patients with sqCLC tend to be slightly older than those with adenocarcinoma. In the Surveillance, Epidemiology, and End Results Registry of more than 50,000 patients with advanced NSCLC diagnosed between 1988 and 2003, the mean age of patients with sqCLC was 66.9 years compared with 64.2 years for patients with adenocarcinoma, and 62% of patients with sqCLC were 65 years or older at diagnosis, compared with 51% for adenocarcinoma.[8] Increased age is associated with worse outcomes in NSCLC; in the Surveillance, Epidemiology, and End Results Registry of patients in the United States in whom NSCLC (of any stage) was diagnosed between 2004 and 2010, those younger than 65 years had a 5-year survival rate of 23%, compared with 19% for those older than 65 years and 17% for those older than 75 years.[51] In the same registry, patients older than 65 years were less likely to receive care in line with current guidelines,[52] a finding supported by the Netherlands Cancer Registry, in which the proportion of patients with stage IV NSCLC receiving chemotherapy decreased with increasing age, from 60% in patients younger than 65 years to 19% in those age 75 years or older.[53]

## Comorbidities

As sqCLC is associated with increased age and has a stronger association with smoking, it is not surprising that patients with squamous disease have a high degree of smoking- and age-related comorbidities and a higher rate of comorbidity than patients with other lung cancers.[54,55] In one large population-based study, 64% of patients with the squamous histologic subtype had a concomitant disease, compared with 52% of patients with other histologic subtypes ($p = 0.001$).[55] In particular, patients with sqCLC had a higher incidence of chronic obstructive pulmonary disease and cardiovascular disease.[54,55]

As for most cancers, the presence of comorbid conditions in NSCLC is associated with a worse prognosis.[55–57] Comorbidity influences survival by affecting treatment choice and treatment adherence, as well as by direct comorbidity-related deaths. In addition, comorbidity in elderly patients with lung cancer has been associated with increased cancer-specific mortality.[57] In a study of 1255 patients with NSCLC, a Charlson comorbidity index score of 1 or higher was associated with significantly shorter survival,[56] and in a further study, severe comorbidity

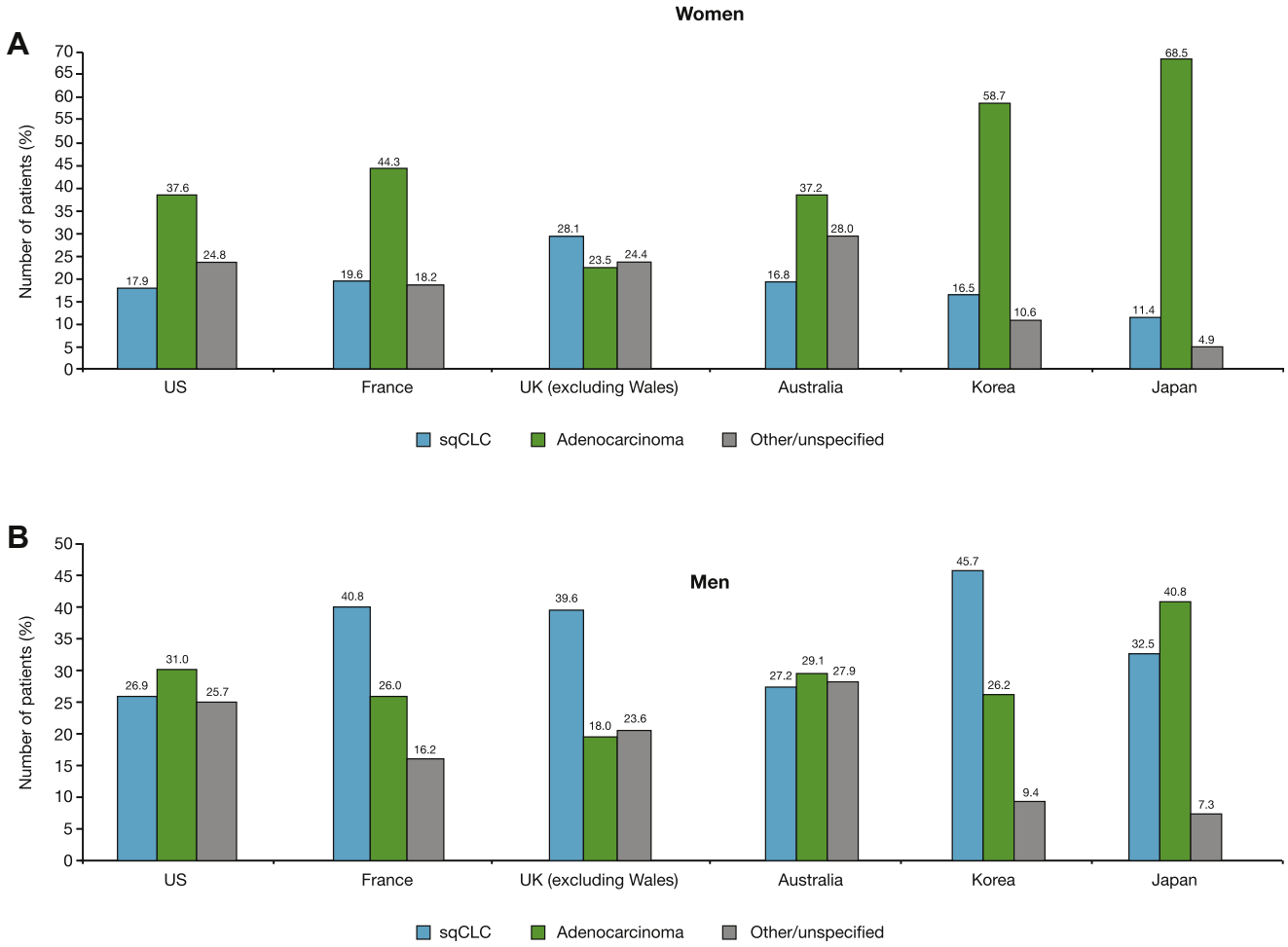


**Figure 2.** Distribution of lung cancer by histologic type in (*A*) women and (*B*) men, 1998–2002. US, United States; UK, United Kingdom; sqCLC, squamous cell lung cancer. Adapted with permission from Youlden et al.[38]

(Charlson comorbidity index score >3) was associated with increased mortality.[57] Indirect estimates suggest that mortality rates for patients with sqCLC who have smoked are 11 times higher than those of patients who have never smoked,[58] which may partly reflect the burden of smoking-related comorbid conditions.

Certain comorbidities, such as lung, cardiovascular, and renal disease, may influence treatment selection, and in clinical practice patients with cancer and comorbidities may be less likely to receive aggressive, or any, chemotherapy.[59] In line with this, in a population-based study of 5428 patients with advanced NSCLC diagnosed between 2001 and 2012, patients with sqCLC were less likely to receive chemotherapy than were those with adenocarcinoma (38% versus 52% [$p < 0.001$]).[53] This may reflect the clinical features of sqCLC already discussed, as well as the high incidence of comorbidities in patients with sqCLC, thereby making these patients less suitable for chemotherapy.

Comorbidity also affects toxicity of chemotherapy. In one study of 402 patients with lung cancer, those with severe comorbidity were more likely to experience thrombocytopenia (46% versus 36%), febrile neutropenia (12% versus 5%), or death caused by neutropenic infection (3% versus 0%) after chemotherapy.[55,59]

Thus, the potential impact of comorbidity on prognosis in sqCLC is multifactorial, and the high burden of comorbidity observed in sqCLC likely contributes to the challenging nature of disease management.

## Molecular Biology

Important differences between sqCLC and other NSCLC histologic subtypes also exist with regard to the tumor genetic profile. In recent years, identification of *EGFR* mutations and anaplastic lymphoma kinase gene *ALK* rearrangements as oncogenic drivers has led to major therapeutic advances for the subset of patients with NSCLC with these alterations.[60–62] However, the incidence of *EGFR* mutations and *ALK* gene rearrangements in sqCLC is low (approximately 2.7% and 1.5–2.5%, respectively),[63–65] and therefore only a few patients with SqCLC are candidates for either EGFR

tyrosine kinase inhibitors (TKIs) or ALK inhibitors. Some studies have suggested that squamous tumors may not harbor *EGFR* mutations at all, and that the low frequency reported is due to samples of mixed histologic subtypes (e.g., adenosquamous) being diagnosed as the squamous subtype.[66] Hence, the benefits of *EGFR* TKIs and *ALK* inhibitors have largely been confined to patients with a nonsquamous histologic subtype.[67,68]

Despite a dearth of *EGFR* mutations or *ALK* gene rearrangements, sqCLC is characterized by a high overall mutation rate and marked genomic complexity.[69] The Cancer Genome Atlas analyzed tumor samples from 178 patients with previously untreated stage I to IV sqCLC and reported that 96% of samples harbored molecular abnormalities, including alterations in fibroblast growth factor receptor gene (*FGFR*), phosphatidylinositol 3-kinase gene (*PI3K*), *EGFR*, and phosphatase and tensin homolog gene (*PTEN*) (Fig. 3).[69] A further cancer cohort study also found that sqCLC is characterized by a high mutational burden, with an average of 261 somatic mutations per tumor, and a mutational spectrum showing a signature of exposure to cigarette smoke.[70] Interestingly, in the same study, comparative analysis between sqCLC samples from Korean and North American patients demonstrated a similar spectrum of alterations in these two populations, which contrasts with lung adenocarcinoma, in which the frequency of genomic alterations in oncogenes was shown to differ among ethnic groups.[70]

The Cancer Genome Atlas study identified two samples with an *EGFR* mutation (1%), although neither of these involved either of the two mutations most commonly seen in adenocarcinoma (exon 21 L858R point mutation or exon 19 deletions). However, 9% of samples had an *EGFR* alteration of some kind,[69] a frequency supported by other studies that found *EGFR* amplification in 7% to 11% of sqCLC cases.[69,71] In fact, high *EGFR* gene copy numbers and protein overexpression have been observed more frequently in squamous disease than in adenocarcinoma (82% versus 44%), and an association between increased gene copy number and poor prognosis has been observed.[71] In addition, *EGFR* gene and protein overexpression have been explored as potential predictive biomarkers of response to EGFR monoclonal antibodies.[72,73] EGFR protein expression and gene copy number are both continuous variables and are measured by immunohistochemical analysis and fluorescence in situ hybridization, respectively. The method used and the cutoff for delineating positive from negative may affect predictive biomarker validity.[73–75] Furthermore, the cut point for detection from background or for a prognostic role may differ from the predictive cut point.[72,74]

Alterations in fibroblast growth factor receptor 1 gene (*FGFR1*) were seen in 7% of samples in The Cancer Genome Atlas study,[69] although *FGFR1* amplification rates of 16% to 21% have been reported in other cohorts of squamous cell tumors.[76,77] Notably, a lower frequency of 3.4% has been observed in adenocarcinoma.[76] However, as with EGFR, the cutoff that defines *FGFR*-positive and *FGFR*-negative varies between studies. In one study of 203 NSCLC cases, a 3.5-fold amplification optimally divided patients into groups with different survival rates. Using the 3.5-fold cutoff, the *FGFR1* amplification rate was approximately 5% in sqCLC and approximately 4% in

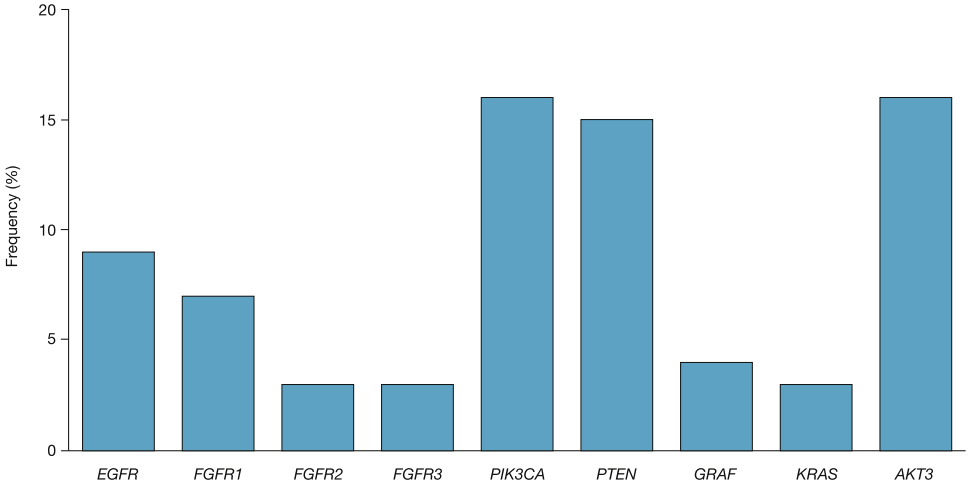


**Figure 3.** Estimated frequency of genetic alterations in squamous cell lung cancer based on analysis of 178 samples from The Cancer Genome Atlas study. Alterations were defined as somatic mutations, homozygous deletions, high-level focal amplifications, or significant upregulation or downregulation of gene expression. *FGFR1*, FGFR1 fibroblast growth factor receptor 1 gene; *FGFR2*, FGFR2 fibroblast growth factor receptor 2 gene; *FGFR3*, FGFR3 fibroblast growth factor receptor 3 gene; *PIK3CA*, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha gene; *PTEN*, phosphatase and tensin homolog; *GRAF*, GRAF Rho GTPase activating protein 26 gene; AKT3, AKT/serine threonine kinase 3 gene. Adapted with permission from the Cancer Genome Atlas Research Network.[69] 

adenocarcinoma.[78] Multiple agents targeting *FGFR1* have been or continue to be evaluated for sqCLC. Initial promising preclinical data have suggested that *FGFR1* inhibition may be an encouraging approach for the subset of squamous tumors harboring an *FGFR1* amplification.[76,79,80] However, clinical response rates have been disappointingly low.[81,82] Recently, evidence has been presented that *FGFR1* mRNA and protein expression, rather than gene copy number, may predict sensitivity to FGFR TKIs across all lung cancer histologic subtypes.[83]

Alterations in the *PI3K* pathway are common in sqCLC, with alterations in one of the three major components of the pathway (phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha gene [*PIK3CA*], *PTEN*, or AKT/serine threonine kinase 3 gene [*AKT3*]) observed in 47% of squamous tumors.[69] *PIK3CA* alterations were identified in 16% of samples in The Cancer Genome Atlas cohort. *PIK3CA* alterations also appear to occur more commonly in sqCLC, with one study finding copy number gains in 42.9% of squamous tumors compared with in 9.5% of adenocarcinomas.[84] *PTEN* mutations also appear to be more common in squamous cell carcinoma than in adenocarcinoma.[85] Of note, a recent phase II study of the pan-PI3K inhibitor buparlisib in relapsed PI3K pathway–activated NSCLC was stopped after failing to meet its primary objective during stage 1 of the study.[86]

Alterations in other genes such as discoidin domain-containing receptor 2 gene (*DDR2*) and nuclear factor erythroid-2-related factor 2 gene (*NRF2*) have been reported in up to 3.8% and 11% of sqCLC, respectively, and may potentially represent therapeutic targets.[85,87] However, a range of *DDR2* mutations, not all of which have been definitely proved to be oncogenic, have been identified.[87,88] To date, clinical trials of agents targeting *DDR2* have been hampered by excessive toxicity, especially in patients with sqCLC.[89] Lack of efficacy has also been noted.[90] Preclinical studies show that mutant *NRF2* is oncogenic and targetable through inhibition of mammalian target of rapamycin.[91]

Together, the frequent genetic alterations in sqCLC tumors suggest common dysfunction in cell cycle control, response to oxidative stress, apoptotic signaling, and/or squamous cell differentiation.[69] Continued focus on this area will be vital to ultimately facilitating a personalized therapeutic approach for sqCLC on par with that seen currently in adenocarcinoma. The Squamous Lung Cancer Consortium is currently studying prognostic classifiers in sqCLC as part of a U.S. National Cancer Institute–initiated collaboration (the Strategic Partnering to Evaluate Cancer Signatures program). The study includes molecular profiling of a large number of sqCLC cases (600–800) and has an additional goal of identifying new potential therapeutic targets for sqCLC.

## Immunological Profile

In addition to molecular aberrations, there are also immunological differences between squamous and nonsquamous histologic subtypes. Squamous tumors more frequently express certain tumor antigens, such as melanoma-associated antigen [MAGE]-A3 and MAGE-A4.[92] Of note, however, in the MAGRIT phase III trial of patients with NSCLC, adjuvant treatment with MAGE-A3 cancer immunotherapy did not increase disease-free survival compared with placebo.[93] Squamous tumors also exhibit more extensive infiltration of $CD8^+$ effector cells, and less extensive infiltration of regulatory T cells, compared with nonsquamous tumors.[94,95] In a study of 65 patients with NSCLC, squamous tumors were associated with a prominent adaptive immune response, whereas greater innate immune responses were observed in adenocarcinomas and large cell carcinomas.[96] Much recent attention has also been paid to immune checkpoint proteins, in particular the programmed death-1 receptor and its ligand programmed death ligand-1 (PD-L1). Initial studies indicate that PD-L1 is expressed in 25% to 39% of NSCLC tumors,[97,98] although the role of PD-L1 expression as a biomarker is yet to be fully determined. The use of multiple assays with widely varying frequencies of positivity has complicated the interpretation of these data.[99–101] Preliminary evidence also suggests that smoking history and a greater mutational burden may correlate with heightened response to anti–PD-L1 agents,[99,102] characteristics that are more often associated with sqCLC. The recent second-line approvals of nivolumab and pembrolizumab across NSCLC histologic subtypes, including squamous cancer, illustrate the new potential of immunotherapy in squamous lung cancer.[103,104] Several anti–programmed death-1 and anti–PD-L1 agents are now in first-line phase III clinical development compared with standard platinum doublets as either monotherapy in PD-L1–enriched populations or in combination with chemotherapy in nonenriched populations.[105–108] The potential influence of differences in the immune environment between sqCLC and other histologic subtypes may be relevant for new and emerging immunotherapies, although this is yet to be fully established.[95]

## Commentary

SqCLC is an especially challenging disease and is associated with a worse prognosis than other histologic subtypes of NSCLC. This is partly due to the aforementioned characteristics, including tumor location, high rate of comorbidity, and genetic complexity of the disease. As a consequence, several recent treatments for NSCLC, including novel drugs targeting oncogenic drivers, new chemotherapeutic agents, and antiangiogenic therapies,

have all demonstrated limitations in terms of efficacy and/or safety in sqCLC.[68,109,110] Therefore, the recommendations for first-line treatment in the vast majority of patients with sqCLC are the same as a decade ago (i.e., platinum-doublet chemotherapy).

It is clear that efforts are needed to address this problem, and one of the main strategies to do so is to improve the understanding of oncogenic drivers specific to the squamous histologic subtype. Genetic profiling of squamous tumors may prove central in discovering novel drug targets and developing new treatments that, together with increased appreciation of the clinical characteristics of sqCLC, will ultimately result in improved patient outcomes.[67] Further investigation of predictive biomarkers for response to immunotherapies will also be crucial.

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