

Lung cancer in the northeast microregion of Rio Grande do Sul, Brazil: epidemiological aspects, molecular and immunohistochemical profile

Câncer de pulmão na microrregião nordeste do Rio Grande do Sul, Brasil: aspectos epidemiológicos, perfil molecular e imuno-histoquímico

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ABSTRACT

Introduction: lung cancer is one of the leading causes of mortality in Brazil and worldwide. Due to the low survival rate, more targeted therapies have been used for effective treatments; thus, studies that identify the profile of patients are essential. **Objective:** to analyse the epidemiological profile of patients affected by lung cancer between 2005 and 2021 in the northeastern microregion of Rio Grande do Sul, Brazil, and to describe the molecular and immunohistochemical profile through the PD-L1, EGFR and ALK biomarkers. **Methods:** this was a cross-sectional, retrospective study with quantitative and qualitative analysis carried out with access to the Hospital Cancer Registry database located in the northeast microregion of Rio Grande do Sul. Epidemiological parameters were histological type, age, gender, stage at diagnosis, family history, smoking and alcohol consumption, as well as data on PD-L1, EGFR and ALK biomarkers. **Results:** a total of 1139 records were analysed. Adenocarcinoma was the most frequent histological type (43.9%), the median age of patients was 65 years, and men were more affected, corresponding to 66.4%. The most prevalent staging was IV (metastatic), and most patients were smokers, non-alcoholics and had a family history of cancer. Regarding the biomarkers, only 49 requests were made during the period. PD-L1 positivity, EGFR alterations and ALK gene rearrangements were observed in 43.8%, 20% and 7.5% of the cases, respectively. **Conclusion:** the epidemiological data corroborate previous studies evaluating lung cancer profiles. Some variations of the analysed biomarkers were verified compared to other studies; nonetheless, more investigation is necessary to assess the profile of biomarkers in the region.

Keywords: Lung neoplasm; Biomarkers; Tumor.

RESUMO

Introdução: o câncer de pulmão é uma das principais causas de mortalidade no Brasil e no mundo. Devido às baixas taxas de sobrevivência, terapias mais direcionadas têm sido utilizadas para tratamentos mais eficazes, logo, estudos que identifiquem o perfil dos pacientes são essenciais. **Objetivo:** Analisar o perfil epidemiológico dos pacientes acometidos pelo câncer de pulmão entre 2005 e 2021 na microrregião nordeste do Rio Grande do Sul, Brasil e descrever o perfil molecular e imuno-histoquímico através dos biomarcadores PD-L1, EGFR e ALK. **Métodos:** trata-se de um estudo transversal, retrospectivo, com análise quantitativa e qualitativa realizado por meio de consulta ao banco de dados do Registro Hospitalar de Câncer na microrregião nordeste do Rio Grande do Sul. Foram avaliados os parâmetros epidemiológicos de tipo histológico idade, sexo, estadiamento ao diagnóstico, histórico familiar, tabagismo e etilismo, e dados dos biomarcadores PD-L1, EGFR e ALK. **Resultados:** um total de 1139 registros foram analisados. O adenocarcinoma foi o tipo histológico mais encontrado (43,9%), a mediana de idade foi de 65 anos e o sexo masculino foi o mais atingido (66,4%). O estadiamento mais prevalente foi o IV (metastático), e os pacientes, em sua maioria, eram fumantes, não-etilistas e com histórico familiar de câncer. Em relação aos biomarcadores, apenas 49 requerimentos foram realizados durante o período. A positividade de PD-L1, alterações no EGFR e rearranjos no gene ALK foram encontrados em 43,8%, 20% e 7,5% dos casos, respectivamente. **Conclusões:** Os dados epidemiológicos corroboram estudos anteriores que avaliaram o perfil do câncer de pulmão. Algumas variações dos biomarcadores analisados foram observadas em comparação com outros estudos; logo, há a necessidade de realização de mais estudos para melhor analisar o perfil de biomarcadores na região.

Palavras-chave: Câncer de pulmão; Biomarcadores; Tumor.

INTRODUCTION

Substantial changes have emerged globally in recent decades related to the prevention, treatment, and screening of different types of lung cancer. Nonetheless, lung cancer is still responsible for the highest number of

cancer deaths worldwide¹. Nationally, Rio Grande do Sul presents the highest number of cases of trachea, bronchi and lung cancer. According to data from the National Cancer Institute (INCA)², the state has a rate of 30.07 cases for men and 16.87 cases for women per 100 thousand inhabitants. These data contrast with other Brazilian states, where men and women have the lowest rates of 9.14 and 6.23 cases, respectively.

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Cancers are frequently classified as early-stage disease (stages I and II), locally advanced disease (stage III), and advanced/metastatic disease (stage IV). Staging is an important step in the diagnostic process, including standardising the main treatment modalities for each stage, estimating prognosis, and comparing the results of several therapies and therapy combinations in different institutions^{3,4}.

Regarding the types of cancer, lung cancer can be divided into two main groups: small-cell lung cancer (SCLC) and non-small-cell lung cancer (NSCLC). This primary differentiation is extremely important due to clinical differences in presentation and therapeutic response⁵. According to data from the American Cancer Society,⁶ NSCLCs account for up to 85% of lung cancer cases. NSCLC is subdivided into large or undifferentiated cell cancer, adenocarcinoma, and squamous cell cancer, with the last two being the most frequent⁷.

The Biomarkers Definition Working Group defines a biomarker as an indicator of normal biological processes, pathogenic processes or responses to an exposure or intervention, including therapeutic interventions. With the advancement of personalised medicine, the definition of lung tumours' immunohistochemical and molecular profile has been fundamental in directing targeted therapies for each patient. In the case of lung tumours, the most widely used are tyrosine kinase inhibitors⁸. Some of the main oncogenes targeted by this therapy are the anaplastic lymphoma receptor kinase (ALK) and the epidermal growth factor receptor (EGFR). Mutations or rearrangements in these oncogenes can outline a defining molecular profile for targeted treatments. In addition, the protein expression of programmed death ligand 1 (PD-L1) in tumour tissue has been a director for the patients most suitable for the use of immunotherapy^{9,10}.

Clinical trials evaluating patients with NSCLC with a positive expression of PD-L1 showed promising results in the treatment of the disease using immunotherapies, such as pembrolizumab¹¹. The efficacy of pembrolizumab used together with conventional platinum-based chemotherapy was also evaluated, showing an increase in survival and progression-free survival¹².

Due to the advancement of personalised medicine and greater knowledge about the prevalence of different biomarkers, targeted therapy in cancer patients has become a promising reality. Currently, in Brazil, there are only a few studies characterising the epidemiological profile of lung cancer, especially over a long period and with recent data^{9,13-15}. In this context, the objective of this study is to identify the scenario of lung cancer in the northeast microregion of Rio Grande do Sul, Brazil, analysing the epidemiological profile of the disease, and describe the molecular and immunohistochemical profile, characterised by the analysis of the biomarkers EGFR, PD-L1 and ALK.

METHODS

This is a cross-sectional, retrospective study with quantitative and qualitative analysis carried out from the electronic database of the Hospital Cancer Registry of a High Complexity Oncology Unit for 24 municipalities in the northeastern micro-region of Rio Grande do Sul. The Research Ethics Committee from Centro Universitário CNEC de Bento Gonçalves and Hospital Tacchini approved this study (protocol n. 5.492.181).

For the analysis of the epidemiological profile, all cases of lung cancer treated at the institution included in the hospital database, which uses the Tumor Registration Form – INCA, were included in the analysis. Considering only full records of patients by the time the present research was conducted, data from January 2005 to December 2021 were collected. The following data were collected: age, gender, stage at diagnosis, family history, smoking, alcohol consumption and histological type. For the biomarkers, considering that the information was received without the identification of the patients, the data were not related to other parameters of the epidemiological profile, given the single and exclusive analysis of their results.

Among the analysed results, the immunohistochemistry method performed a data sample referring to the expression of PD-L1. The results varied between 0 and 100% of expression, and when some degree of expression was verified, the result was considered positive. For the EGFR, the test was performed using the Polymerase Chain Reaction (PCR) method, and its results constituted the molecular profile of the present study, which could have wild or present variations. A sample of data referring to the ALK test was carried out using the immunohistochemical method, allowing a positive or negative result for the detection of rearrangements in the gene.

A basic descriptive analysis of the data was performed. The results were expressed in absolute, relative (%), and mean values.

RESULTS

The epidemiological profile of 1139 patients diagnosed with lung cancer from 2005 to 2021 was evaluated considering the following variables: histological type, age, sex, stage, smoking, alcoholism, and family history (Table 1).

Table 1 – Characteristics of patients with lung cancer treated at a High Complexity Oncology Unit located in the northeast of Rio Grande do Sul, Brazil.

Variables	N (%)
Histological type	
Adenocarcinoma	500 (43.9%)
Squamous cell carcinoma	226 (19.8%)
Small cell carcinoma	136 (11.9%)
Poorly differentiated non-small cell carcinoma	133 (11.7%)

Carcinoma – NOS (not otherwise specified)	47 (4.1%)
Undifferentiated malignant neoplasm	97 (8.5%)
Age	
> 60 years	806 (70.8%)
≤ 60 years, n (%)	333 (29.4%)
Gender	
Male	756 (66.4%)
Female	383 (33.6%)
Stage	
I	58 (5.1%)
II	44 (3.9%)
III	139 (12.2%)
IV	494 (43.4%)
No information	404 (35.5%)
Smoking	
Yes	650 (57.1%)
No	179 (15.7%)
Ex-consumer	202 (17.7%)
No information	108 (9.5%)
Alcoholism	
Yes	238 (20.9%)
No	454 (39.9%)
Ex-consumer	70 (6.1%)
No information	377 (33.1%)
Family history	
Yes	294 (25.8%)
No	148 (13.0%)
No information	697 (61.2%)
Total	1139

Considering the histology, six distinct types were verified: adenocarcinoma, squamous cell carcinoma, small cell carcinoma, poorly differentiated non-small cell carcinoma, carcinoma without other specifications (NOS) and undifferentiated malignant neoplasm. Adenocarcinoma was the most common histological type, with 500 cases (43.9%). This type, together with squamous cell carcinoma (19.8%) and poorly differentiated non-small cell carcinoma (11.7%), corresponded to NSCLC cases, totalling 859 cases (75.4% of the total number of cases evaluated).

Age (in years) ranged between 28 and 95 years (median 65 years), with 806 (70.8%) patients being over 60 years. In addition, male patients were the most affected, corresponding to 756 (66.4%). A total of 494 (43.4 patients presented stage IV (metastatic). Although a considerable percentage was found in this staging, the information was missing in 404 (35.5%) cases.

In the present study, 650 (57.1%) patients were smok-

ers, in addition to another 202 (17.7%) considered former users. Regarding alcoholism, the highest percentage was of non-alcoholics, with 454 (39.9%) patients. The presence of a family history of cancer was little computed, as 697 (61.2%) patients did not present this information. Among those who had the information, 294 (25.8%) were positive for the family history, and 148 (13.0%) were not.

Concerning biomarkers, their requests at the institution began in 2015 and are restricted to PD-L1, ALK and EGFR biomarkers in lung cancer cases. The first analyses were carried out for lung cancer cases in 2017, demonstrating an increase in the requests until 2021, totalling 49 requests. Some included the three biomarkers analysed in the present study, while others included only one or two. Separately, during the survey period, the most requested exam was PD-L1 (48 requests), followed by EGFR (45 requests) and, finally, ALK (40 requests).

Of these 48 PD-L1 results, 27 (56.3%) were negative, and 21 (43.7%) were positive. According to the tumour proportion score (TPS), cut-off values commonly used in other clinical studies, a PD-L1 TPS ≥ 50% is defined as a cut-off point, being considered a high level of PD-L1 expression¹¹. Although a considerable percentage of the total had some level of expression of the biomarker, only 9 (18.8%) had a TPS ≥ 50%.

Considering the molecular profile, variations in the EGFR gene were evaluated in 45 patients. The wild profile was more frequent, without alterations in the gene structure, corresponding to 80% (36/45) of the patients. Among the 20% (9/45) who had some alteration, 66.6% had a deletion in exon 19, and only one patient was identified as an L858R variant in exon 21, one as a G719X variant in exon 18 and one with an insertion in exon 18. exon 20 (Table 2). Regarding immunohistochemistry, ALK gene rearrangements were evaluated in 40 patients, only 7.5% (3/37) were positive (Table 3).

Table 2 – Frequency of EGFR gene testing in patients with NSCLC treated at a High Complexity Oncology Unit in northeastern Rio Grande do Sul, Brazil.

Mutation	N (%)
Wild	36 (80%)
Exon 19 deletion	6 (13.3%)
L858R Exon 21 variant	1 (2.2%)
Exon 20 insertion	1 (2.2%)
G719X Exon 18 variant	1 (2.2%)

Table 3 – PD-L1 expression, EGFR gene mutation status and ALK gene expression in patients with NSCLC treated at a High Complexity Oncology Unit located in northeast Rio Grande do Sul, Brazil.

Histological condition	PD-L1 expression (N = 48)		EGFR gene mutation (N = 45)		ALK gene expression (N = 40)	
	Negative N (%)	Positive N (%)	Wild N (%)	Mutant N (%)	Negative N (%)	Positive N (%)
NSCLC	27 (56.2%)	21 (43.8%)	36 (80%)	9 (20%)	37 (92.5%)	3 (7.5%)

DISCUSSION

The present study aimed to characterise the epidemiological profile and biomarkers of patients diagnosed with lung cancer from a microregion of Rio Grande do Sul. Adenocarcinoma was the most prevalent histological type, which, together with squamous cell carcinoma and poorly differentiated non-small cell carcinoma, correspond to cases of NSCLC, totalling 75.4% of all cases evaluated. The percentage found in the present study is lower than the data reported by the American Cancer Society, which mentions a prevalence of more than 80% of NSCLC concerning the total number of cases of lung cancer⁶. Likewise, a study evidenced that non-small cell lung cancer corresponds to 80-85% of all cases of lung cancer⁷. The amount of undifferentiated malignant neoplasia, corresponding to 8.5% of the total cases, could contribute to this difference.

The median age of 65 years was similar to previous studies performed in Brazil, which found values of 67 and 60 years^{9,16}. A retrospective study carried out in Brazil that evaluated cases of lung cancer from 2013 to 2020 found that the most significant number of diagnoses were made in patients aged between 60 and 69 years¹⁷. These data demonstrate that patients over 60 years old are considered critical for the diagnosis of the disease. Thus, the implementation of public policies aiming to encourage the population to seek health care services earlier could result in the diagnosis of the disease in the initial stages.

In the present study, 66.4% of cases of lung cancer occurred in men. This finding is similar to data from INCA, which estimated 17,760 new cases of lung cancer in men and 12,440 in women². Moreover, a study performed through administrative databases from Brazilian private healthcare systems has also evidenced more cases in men, corresponding to 58.1%¹⁸. Pereira et al.¹⁵ (2023) have also found a higher prevalence in men from the Brazilian Amazon and reported that the frequency of lung neoplasm in this population is typically related to tobacco consumption since the ratio of men smokers is more significant than that of women smokers. In contrast, a retrospective study carried out in Brazil and a study evaluating biomarkers carried out in Latin America found higher frequencies of lung cancer in women, corresponding to 55% and 53.5%, respectively^{17,19}. This could be due to sex hormones that play a decisive role in tobacco smoke-induced lung cancer development, as reviewed previously²⁰. Nonetheless, it is important to highlight that lung carcinogenesis is a very complex event and that multiple specific risk factors contribute to its development.

Most patients with lung cancer were in stage III or IV, being the stage IV (which indicates locally advanced or metastatic disease) the most prevalent. These findings corroborate a pattern already known about lung cancer, which shows a low percentage of early diagnosis and 5-year survival of less than 30% for stages III and IV^{21,22}. A study evaluating epidemiological variables in cancer

in Brazil evidenced that 85% of the analysed cases were in stage III or IV¹⁶. Further revisions to the tumour, node and metastasis (TNM) staging system²³ and histological classifications²⁴, as well as the implementation and greater availability of new technologies for the diagnosis of metastatic lesions, are possibly the main factors related to the increased proportion of lung cancer cases diagnosed at an advanced stage in Brazil²⁵. However, it is worth mentioning that this information was not available in 35% of the reports analysed in the present study.

Regarding smoking, the vast majority of patients used or were former users (74.8%). Considering that another 9.5% of the cases did not have this information, the number of smokers could be even higher. The majority of lung cancer cases diagnosed among these individuals suggest that the accumulation of molecular damage during cigarette exposure sets a cascade of events in motion that leads to the diagnosis of cancer, often decades after smoking cessation²⁶.

The alcoholism variable was primarily composed of people who did not use alcohol (39.9%). This variable is important to consider because, according to the review conducted by García-Lavandeira et al. (2016)²⁷, the relationship between alcohol consumption and lung cancer risk is still inconclusive. However, a study demonstrated that alcohol consumption might be a risk factor for lung cancer, with alcohol types (wine, beer, spirits) having a different effect on the onset of the disease²⁸. In this context, according to Ahrendt et al. (2000)²⁹, alcohol use seems to have an impact on the occurrence of p53 mutations, which are related to the development of cancer.

Unfortunately, information about family history was not available in most medical records (61.2%). The lack of this information characterises a limitation in the comprehension of the profile of patients with lung cancer. Understanding the aetiology of the disease becomes even more complex in these cases, especially for those who do not have other associated risk factors, such as smoking.

Considering the immunohistochemistry, a previous study identified the EML4-ALK fusion gene and reported its presence in 6.7% of cases of NSCLC³⁰. Similarly, the present study found a frequency of 7.5% for gene rearrangements. On the other hand, at the national level, a higher frequency, corresponding to 10.4%, was verified in a study carried out in the northeast region⁹, whereas a lower frequency, corresponding to 3.2%, was verified in a study conducted in the Southeast region³¹. Another study carried out with patients with adenocarcinoma in southern Brazil found 4% positivity in the analysed ALK samples, evidencing, together with other analyses, a distinction in the profile of biomarkers compared to other regions of Brazil¹³.

The present study observed that 20% of the patients had some variation in the EGFR gene. This finding is similar to other national studies carried out in the Northeast and Southeast regions, which reported frequencies of 22.1% and 19.16%, respectively^{9,13}, suggesting a non-disparity

in the molecular profile of the gene in the country. Conversely, the data are divergent when compared to studies carried out in other countries. In Asian patients, positive values greater than 50% of the total lung cancer cases evaluated were found³². Other studies conducted in Latin America reported EGFR mutation frequencies between 24 and 35% of the prevalence of mutation^{19,33}.

The expression of PD-L1 was found in 43.8% of patients analysed in the present study. Similarly, another study carried out in São Paulo, Brazil, found expression in 37.9% of the analysed cases³⁴. In contrast, 50.9% of patients with NSCLC had some degree of PD-L1 expression in the northeast region⁹. Based on preliminary studies that evaluated the expression of PD-L1 considering a TPS $\geq$ 50% as a high expression, only 18.8% of the samples in the present study presented this value. Two other studies carried out in the country evaluating the expression of PD-L1 in tumour cells found 16.6% and 18.2% of cases with a TPS $\geq$ 50%^{9,35}. Thus, verifying a similar prevalence in the PD-L1 expression was possible nationally. It should be noted that discrepancies between the studies may be due to different baseline clinical characteristics of the patients and the lack of standardisation of the definition of PD-L1 positivity⁹. However, additional studies are needed to assess possible differences in the expression level of PD-L1 in different regions of the country, given distinct demographic characteristics, such as gender and smoking behaviour, and genetic backgrounds.

The present study has some limitations, considering the lack of information related to the epidemiological profile of patients, especially alcoholism and family history, which may have interfered with the definition of the profile of patients with lung cancer. This is possibly due to the lack of definition in filling out the data, mainly because it is an analysis of 17 years of data. It should be noted that smoking, an extremely important variable associated with neoplasia, showed a lack of standardisation regarding habits of use, including time of use. Regardless, the data point to a relationship between smoking and lung cancer, as already described in the literature.

CONCLUSION

The epidemiological data assessed in the present study cover 17 years of cases and corroborate previous studies that evaluated the profile of lung cancer. However, some variations of the analysed biomarkers were verified in comparison with other national and international studies, mainly concerning ALK and EGFR. Due to the lack of data on the subject, it is still impossible to define a profile of biomarkers in the region. Nonetheless, the importance of performing this type of examination is highlighted, considering the benefits of the targeted therapy. Therefore, it is necessary to encourage permission for this type of examination to obtain more data on the subject. Consequently, this information could be used for the benefit of patients treating lung cancer.

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