Immunohistochemical study of tumour-infiltrating lymphocytes in non-small cell lung cancer

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BACKGROUND

According to the International Agency for Research on Cancer (IARC) of the World Health Organization (WHO) lung cancer remains the most frequent cancer worldwide with fatal consequences, as it is the leading cause of cancer-related mortality in many countries [1].

Adenocarcinoma and squamous cell carcinoma are most common histotypes, and they belong to the group of non-small cell lung cancers (NSCLC) [2]. Increased density of tumour-infiltrating lymphocytes (TILs) in NSCLC is associated with better prognosis [3, 4].

MATERIALS AND METHODS

The material covers 210 routinely processed, formalin-fixed and paraffin-embedded (FFPE) tissues of primary squamous cell lung carcinoma (SQC) and adenocarcinoma (AC) from patients who underwent pulmonary resection.

Squamous cell carcinoma samples comprise 53% and adenocarcinoma 47%. A tissue microarray (TMA) is a paraffin block that can contain small tissue samples (narrow cores) from many patients.

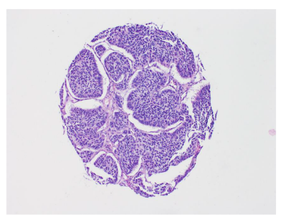
Four 2 mm in diameter cores were taken from every paraffin block containing NSCLC tissue (from one patient) using a manual tissue arrayer (MTA) machine (Beecher Instruments, USA) and were constructed the paraffin blocks containing tissue cores.

The tumour tissue microarrays (TMAs) were made and stained with hematoxylin and eosin (HE), and antibodies focused on immune markers (CD3, CD8, CD56, CD163) on the slides containing 4 micrometres sections of a TMA block with the positive tissue controls (tonsil, lymph node, spleen, hepar).

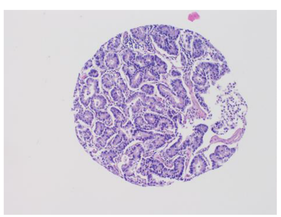
Histological analysis of the cores was performed on HE and IHC slides, and assessment of the histopathological type of lung carcinoma, the percentage of tumour-infiltrating lymphocytes (TILs) and other mononuclear immune cells were included.

The percentage of positively stained cells, their staining pattern, and distribution of immune cells in both adenocarcinoma and squamous cell carcinoma were evaluated. The histopathological type is based on the WHO Classification [2].

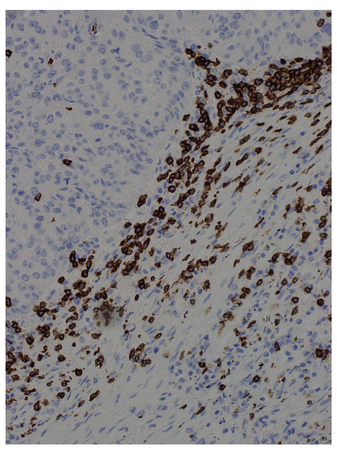
RESULTS



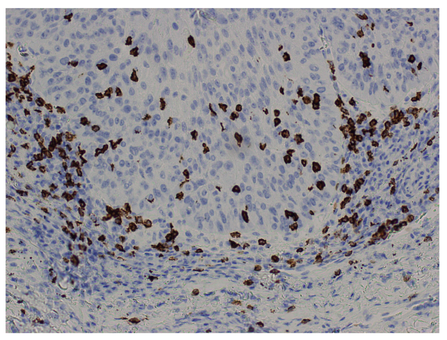
TMA cores with squamous cell carcinoma, HE stain.



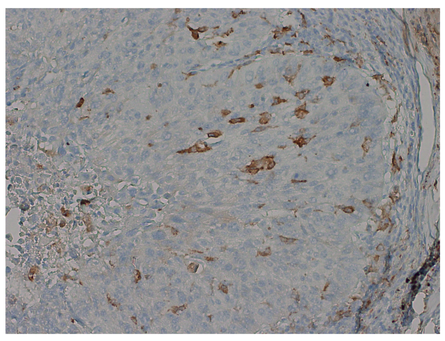
TMA cores with adenocarcinoma, HE stain.



CD3 positive T cell lymphocytes in squamous cell carcinoma



CD8 positive T cell lymphocytes in squamous cell carcinoma



CD163 positive histiocytes in squamous cell carcinoma

The results point to the variability of the mononuclear immune cells infiltration in NSCLC.

The cancer area is not linked to the total number of immune cells.

It is evident that the stroma and the cancer area, the distribution and the number of the immune cells differ between the cores of the same patient.

It is found that CD3 positive T cell lymphocytes are the most frequent group of immune infiltration cells, and its significant part is CD8 positive cells. The histiocytes (CD163 positive) are the second group, but much less numerous.

Almost all CD3 positive T cell lymphocytes are CD8 positive in between cancer cells, but the total number of CD3 positive T cell lymphocytes is not related to the number of CD3 positive cells between cancer cells.

The number of CD3 positive T cell lymphocytes at the edge of tumour is likely to be higher than in central parts of the tumour.

There is a tendency for squamous cell carcinoma to have more abundant immune infiltration than adenocarcinoma.

NK cells (CD56 positive) are scattered in NSCLC, mainly exist in the stroma.

There is the association of the total number of CD3 positive T cell lymphocytes in immune infiltration with CD3 positive T cell lymphocytes adjacent to cancer cells of squamous cell carcinoma.

The findings perhaps suggest that the histological grade of squamous cell carcinoma is linked to the total number of immune cells.

It is noticeable that the immune cells (in the focal arrangement and as tertiary lymphoid structures) are abundant at the edge of tumour in adenocarcinoma samples.

CONCLUSIONS

Our study provides some useful insights into the NSCLC-immune crosstalk - CD3+ CD8+ lymphocytes are the most common cells of immune infiltration.
On the basis of our preliminary data, it is required to delve into other T lymphocyte subpopulations, which have not yet been studied in NSCLC, and other immune checkpoints. We expect to find them.

References:

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