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Liquid biopsy: Innovative and non-invasive technique for the study of ALK gene rearrangements

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Statement of the Problem: Lung cancer is the one with the highest mortality rate in the world. There are two main types: Small-Cell Lung Carcinoma (SCLC) and Non-Small-Cell Lung Carcinoma (NSCLC). The discovery of Tyrosin Kinase Inhibitors (TKI) that target Anaplastic Lymphoma Kinase (ALK) gene rearrangements have achieved a huge success in the management of patients with ALK-positive NSCLC. Although a great advance in the treatment of these patients has been achieved, the initial diagnosis remains a challenge since it is confirmed by a tissue biopsy, which requires an invasive procedure. Liquid biopsy testing is a new, non-invasive technique, suitable to identify NSCLC patients that can benefit from ALK-targeted therapies. The objective of the study is to focus on liquid biopsy using a specific blood component, known as Tumor-Educated Blood Platelets (TEPs) and how they may assist in ALK gene rearrangements detection.

Methodology: Firstly, different methods for platelet RNA extraction were tested from blood of healthy donors. The chosen method was used to extract the mutated RNA from the TEPs of patients with NSCLC. The RNA profile of both populations was compared. Secondly, we determined the optimal digital PCR (dPCR) conditions to detect the presence of variant 3 of the EML4-ALK fusion gene in a positive control (cell line H2228). Finally, we used dPCR to analyze EML4-ALK rearrangements in TEPs from the blood of NSCLC patients.

Findings: Platelets are a valuable source for the non-invasive detection of EML4-ALK rearrangements. The type of RNA population present in platelets varies depending on the extraction method used. dPCR is a useful technique for the detection and quantification of EML4-ALK rearrangements.

Conclusion: In this study we present a standardized method to extract platelet RNA and detect EML4-ALK rearrangements using dPCR, for which there was no previous data on yields or optimization conditions. This will mean an improvement in the application of liquid biopsy as an alternative protocol in the search for biomarkers in patients with NSCLC.

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