Exosomes: A Key Role in the Molecular Mechanism of Brain Metastases of Bronchopulmonary Origin

ABSTRACT

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Brain metastases represent over 50% of all brain tumors encountered in clinical practice and constitute a frequent and severe complication of lung cancer, the leading cause of cancer-related mortality worldwide. In recent years, research has increasingly focused on identifying the molecular mechanisms involved in metastatic progression, emphasizing the critical role of extracellular vesicles, particularly exosomes. These significantly contribute to organotropic metastatic dissemination by increasing blood-brain barrier permeability and preparing the premetastatic niche. Thus, exosomes have become promising biomarkers for liquid biopsies and potential drug carriers, capable of enhancing early diagnosis and therapeutic efficacy in patients diagnosed with brain metastases.

This doctoral thesis initially aimed to analyze systemic inflammatory markers relevant for assessing prognosis in patients with lung cancer, with or without brain metastases. Subsequently, we investigated the role of exosomes in the cerebral metastatic process, identifying and quantifying essential proteins involved in angiogenesis, extracellular matrix remodeling, and tumor metabolic reprogramming. Consequently, Angiopoietin-2 (ANGPT2), cell migration-inducing protein (CEMIP), and the ketolytic enzyme 3-oxoacid CoA-transferase 1 (OXCT1) were analyzed.

The first prospective study included 228 subjects divided into three distinct groups: patients diagnosed with lung cancer and brain metastases (n=67), patients with lung cancer without identified metastases (n=88), and healthy subjects (n=73). For each group, systemic inflammatory indices NLR, PLR, LMR, and SII were evaluated, revealing statistically significant increases for SII, NLR, and PLR in the oncologic groups compared to healthy subjects. These values demonstrated a clear upward trend proportional to disease severity and progression toward the metastatic stage.

In the subsequent second study, we analyzed for the first time the presence and expression of proteins ANGPT2, CEMIP, and OXCT1 in exosomes isolated from the plasma of enrolled patients, using advanced techniques such as density gradient ultracentrifugation, electron microscopy, flow cytometry, and Western blot analysis. Results indicated significantly elevated expressions of all three biomarkers in exosomes derived from oncologic patients compared to healthy subjects. CEMIP levels were significantly higher (by approximately 59%) in patients with brain metastases compared to those with localized lung cancer. OXCT1 expression was significantly higher in

metastatic stages compared to patients without metastases, the percentage difference being almost doubled in the metastatic group.

Evaluating the therapeutic effect of surgical intervention on these biomarker levels revealed a significant postoperative reduction in ANGPT2 and CEMIP levels both in patients with localized lung cancer (36% and 8.5%, respectively) and in those with brain metastases (40% and 4.6%, respectively). Regarding OXCT1 expression, surgery resulted in a significant reduction of approximately 20% in patients without brain metastases, whereas the decrease was minimal (below 1%) and statistically insignificant in patients with brain metastases.

These findings support the potential of exosomes and associated biomarkers—ANGPT2, CEMIP, and OXCT1—alongside systemic inflammatory indices, as tools for early diagnosis, disease progression monitoring, and prognosis evaluation in lung cancer-derived brain metastases. Moreover, the differential expression of OXCT1 among the studied groups confirms metabolic reprogramming as a central mechanism in cerebral metastatic progression, offering valuable perspectives for developing novel therapeutic strategies targeting tumor energetic metabolism.

Future research directions should further explore detailed molecular mechanisms and complex interactions mediated by exosomes in the tumor microenvironment, as well as therapeutic opportunities derived from manipulating these biomarkers to control brain metastases.