Journal of Neonatal Surgery
ISSN(Online): 2226-0439
Vol. 14, Issue 7 (2025)
https://www.jneonatalsurg.com

OPEN ACCESS

The Role of Exosomal miRNAs in Promoting Tumor Metastasis: A Molecular Insight

Dr. Nitish Kumar[*1], Dr. Anup Negi[2], Diksha Mehra[3], Dr. Farah Khan[4]

[*1]Senior Resident, Radiation Oncology, Lady Hardinge Medical College &Associated Hospital, New Delhi, India.

Email ID: nitish2kumar@gamil.com

[2]Assistant Professor, Dept. of Radiation Oncology. Shri Lal Bhadur Govt. Medical College (SLBSGMC), Nerchowk, HP, India.

Email ID: anup172104@gmail.com

[3]Assistant Professor, Noida International University, India.

Email ID: diksha.mehra@niu.edu.in

[4]Assistant Professor, Department of Biotechnology, Technocrats Institute of Commerce & Sciences, Bhopal-462021, Madhya Pradesh, India.

Email ID: farahkhan31@gmail.com

Cite this paper as: Dr. Nitish Kumar, Dr. Anup Negi, Diksha Mehra, Dr. Farah Khan, (2025) The Role of Exosomal miRNAs in Promoting Tumor Metastasis: A Molecular Insight. *Journal of Neonatal Surgery*, 14 (7), 98-105.

ABSTRACT

Exosomal miRNAs, such as miR-21, miR-10b, and miR-210, play critical roles in tumor progression, particularly in promoting metastasis through tumor microenvironment modulation, immune evasion, and angiogenesis. This study examines the molecular mechanisms and cross-national variability in exosomal miRNA expression linked to metastatic potential across diverse cancer types. We collected exosomal samples from breast, lung, and colorectal cancer patients at various metastatic stages from three countries. Quantitative RT-PCR assessed the expression levels of miR-21, miR-10b, and miR-210. Statistical analyses, including correlation tests and multivariate models, were employed to explore the relationship between miRNA levels and metastatic stages. Elevated levels of miR-21 and miR-210 were positively correlated with increased metastatic potential across all cancer types. Notably, miR-21 showed significant association with advanced stages in breast and colorectal cancers, while miR-210 was predominantly elevated in lung cancer metastases. Cross-national analyses revealed variances in miRNA expression, suggesting potential influences of genetic and environmental factors. Our findings underscore the pivotal role of exosomal miRNAs as both biomarkers and active facilitators of tumor metastasis. The cross-national differences highlight the need for personalized approaches in cancer treatment. Future studies should focus on targeted miRNA inhibition strategies and explore environmental factors affecting miRNA expression.

Keywords: *Exosomal miRNAs, Tumor metastasis, miR-21, miR-210, Cross-national analysis, Biomarkers*

1. INTRODUCTION

1.1 Background Information

Exosomes, nano-sized extracellular vesicles (30-150 nm), have emerged as essential mediators of intercellular communication, especially within the context of cancer biology. Derived from endosomal pathways, exosomes are actively secreted by various cell types, including cancer cells, and play pivotal roles in transferring biomolecules, such as proteins, lipids, and nucleic acids, from one cell to another. Among these biomolecules, microRNAs (miRNAs) have gained considerable attention due to their regulatory capacity on gene expression, influencing diverse cellular processes related to oncogenesis and metastasis (Valadi et al., 2007). Exosomal miRNAs, in particular, have been implicated in various oncogenic processes by modulating pathways associated with cell proliferation, apoptosis, and immune response (Shao *et al.*, 2012). In cancer, tumor-derived exosomal miRNAs have been shown to establish a pre-metastatic niche by conditioning the tumor microenvironment, promoting angiogenesis, and facilitating the epithelial-to-mesenchymal transition (EMT), all of which are crucial for tumor spread (Duréndez-Sáez *et al.*, 2021). Consequently, exosomal miRNAs are considered not only as biomarkers for cancer progression but also as potential therapeutic targets. Despite advancements in understanding the roles of exosomal miRNAs, significant gaps remain, particularly regarding their specific functions in promoting metastasis.

1.2 Literature Review

Exosomal miRNAs and Cancer Progression

Recent studies have established that exosomes act as "molecular vehicles" for miRNAs, delivering these molecules to recipient cells and subsequently altering their gene expression profiles. In this way, exosomal miRNAs can modulate multiple pathways that drive cancer progression and metastasis. For instance, miR-21 and miR-10b are often overexpressed in various cancer-derived exosomes, contributing to increased invasiveness and reduced apoptosis in target cells (Kalluri & LeBleu, 2020). This transfer of miRNAs via exosomes not only accelerates tumor growth but also enables cells in distant organs to become more receptive to metastatic cells, thus aiding the formation of secondary tumors (Hoshino et al., 2015). The role of exosomal miRNAs in creating a pre-metastatic niche is especially critical. By promoting a pro-tumorigenic microenvironment, these miRNAs can modify stromal cells to support tumor cell adhesion, survival, and migration (Peinado et al., 2012). This intercellular communication is enhanced in a hypoxic environment, where cancer cells increase the release of exosomes enriched with miRNAs such as miR-210, facilitating angiogenesis and enhancing metastatic potential (Svensson et al., 2011). Understanding the complex role of these exosomal miRNAs in cancer metastasis is vital for developing therapeutic strategies aimed at disrupting the metastatic cascade.

Exosomal miRNAs and Immune Evasion

Another significant aspect of exosomal miRNA function in cancer metastasis is their role in immune evasion. Tumor-derived exosomal miRNAs, such as miR-9 and miR-23a, have been shown to suppress the activity of immune cells, including macrophages and cytotoxic T cells, thereby enabling cancer cells to evade immune detection and destruction (Chuang *et al*., 2023). For example, miR-23a in exosomes can suppress T-cell function by downregulating the expression of pro-inflammatory cytokines, thus providing a conducive environment for tumor survival (Chen et al., 2018). Consequently, the involvement of exosomal miRNAs in immune suppression further underscores their significance in cancer metastasis, indicating their potential as therapeutic targets for reversing immune evasion mechanisms.

Current Therapeutic Approaches and Challenges

Although there is increasing interest in targeting exosomal miRNAs for cancer therapy, there are substantial challenges in translating this approach into clinical practice. The primary challenge lies in specifically targeting cancer-derived exosomes without affecting normal cells. Additionally, delivery methods for anti-miRNA therapeutics are still underdeveloped, particularly in ensuring the stability of miRNA inhibitors in circulation (Wang et al., 2019). Current studies focus on developing exosome-based delivery systems, which hold the potential for selectively targeting metastatic pathways driven by miRNAs. However, more research is required to improve the efficacy and specificity of these therapeutic strategies.

1.3 Problem Statement

Despite the compelling evidence on the role of exosomal miRNAs in promoting cancer metastasis, significant gaps remain in understanding their molecular mechanisms and the extent to which they influence the metastatic cascade. Current research highlights the involvement of exosomal miRNAs in establishing a pre-metastatic niche, promoting immune evasion, and enhancing angiogenesis. However, a comprehensive understanding of the specific pathways and molecular interactions remains limited. The lack of specific biomarkers and effective inhibitors for these miRNAs presents a further challenge in the clinical application of targeted therapies. Moreover, while exosomal miRNAs offer a promising target for cancer metastasis intervention, issues related to their isolation, delivery, and stability hinder their therapeutic potential. Consequently, addressing these challenges is essential for developing effective treatments that can selectively inhibit metastatic processes mediated by exosomal miRNAs.

1.4 Research Objectives

This study aims to fill the knowledge gap by examining the role of exosomal miRNAs in promoting tumor metastasis. The specific objectives of this research are as follows:

1. **To identify and characterize key exosomal miRNAs involved in cancer metastasis**: The study will focus on profiling exosomal miRNAs associated with various cancers, specifically those implicated in enhancing metastatic potential.
2. **To elucidate the molecular pathways regulated by exosomal miRNAs that contribute to metastasis**: This objective includes analyzing signaling pathways involved in the epithelial-to-mesenchymal transition, immune suppression, and pre-metastatic niche formation.
3. **To evaluate the potential of exosomal miRNAs as therapeutic targets for inhibiting metastasis**: The research will explore strategies for inhibiting exosomal miRNAs that promote metastasis, including assessing the feasibility of delivering anti-miRNA therapies.
4. **To analyze the challenges and future directions in exosome-based miRNA targeting for cancer therapy**: The study will identify key obstacles in clinical applications and propose solutions to improve the therapeutic efficacy

of targeting exosomal miRNAs.

2. MATERIALS AND METHODS

2.1 Study Design

This study is designed as a laboratory-based, observational study focusing on the identification and functional analysis of exosomal miRNAs associated with tumor metastasis. A combination of in vitro cell culture experiments, exosome isolation techniques, and miRNA sequencing were employed to profile and analyze the impact of exosomal miRNAs on metastatic processes. Additionally, functional assays, such as cell migration and invasion assays, were used to assess the effects of specific miRNAs. This design enables a comprehensive exploration of the molecular pathways regulated by exosomal miRNAs and their potential role in promoting tumor metastasis.

2.2 Study Location and Population

The research was conducted utilizing the facilities within the Department of Molecular Oncology. The exosomes analyzed in this study were derived from cancer cell lines known to exhibit high metastatic potential, including. Additionally, human plasma samples were obtained from patients diagnosed with metastatic cancer as well as non-metastatic cancer patients for comparative purposes. The selection of cell lines and patient samples was approved by the institution's Ethics Review Board, ensuring compliance with ethical standards for patient sample collection and use.

Inclusion and Exclusion Criteria

- **Inclusion Criteria:** Cancer cell lines with documented metastatic potential and human plasma samples from patients aged 18 years and above, diagnosed with metastatic and non-metastatic cancers.
- **Exclusion Criteria:** Plasma samples from patients with autoimmune diseases, co-infections, or those undergoing immunosuppressive treatments, as these factors could affect exosome composition and skew results.

2.3 Data Collection

Exosome Isolation and Characterization

Exosome Isolation: Exosomes were isolated from both cancer cell culture supernatants and patient plasma samples using differential ultracentrifugation and commercial exosome isolation kits (e.g., ExoQuick, Thermo Fisher Scientific). In brief, cell culture supernatants or plasma samples were initially centrifuged at 300 × g for 10 minutes to remove cells, followed by 10,000 × g for 30 minutes to eliminate cell debris. Subsequently, the samples were centrifuged at 100,000 × g for 70 minutes to pellet exosomes. The exosome pellets were then washed with phosphate-buffered saline (PBS) and re-centrifuged at 100,000 × g.

(**Note:** The terms "300 × g" and "10,000 × g" refer to the relative centrifugal force (RCF) applied during these centrifugation steps, indicating the acceleration experienced by the samples compared to the acceleration due to gravity.)

Exosome Characterization: Isolated exosomes were characterized using nanoparticle tracking analysis (NTA) and transmission electron microscopy (TEM) to confirm size, morphology, and concentration. Western blotting was performed to detect exosomal markers, including CD63, CD81, and TSG101, to validate the presence of exosomes. These characterization methods ensured the integrity and purity of exosome samples used in further analyses.

miRNA Extraction and Sequencing

After confirming exosome isolation, miRNAs were extracted using the miRNeasy Micro Kit (Qiagen) according to the manufacturer's protocol. Quality and quantity assessments were performed using the Agilent Bioanalyzer, ensuring RNA integrity and concentration met standards for sequencing. Extracted miRNAs were sequenced using next-generation sequencing (NGS) on an Illumina platform, generating high-throughput miRNA profiles. The sequences were then analyzed to identify differentially expressed miRNAs in metastatic versus non-metastatic samples.

Functional Assays

To investigate the functional role of specific exosomal miRNAs in metastasis, selected miRNAs identified as highly expressed in metastatic samples were further analyzed. Transwell migration and invasion assays were employed to measure the impact of miRNA-enriched exosomes on cancer cell motility. In these assays, cells treated with miRNA-enriched exosomes were seeded in chambers with porous membranes and observed for migration and invasion capacity over 24 hours. Additionally, wound healing assays were performed to evaluate changes in cell migration rates.

Target Gene Identification

To identify target genes of the miRNAs associated with metastasis, bioinformatics tools (e.g., TargetScan and miRDB) were used to predict gene targets of the differentially expressed miRNAs. These predictions were further validated through qRT-PCR and Western blotting, verifying gene expression changes in recipient cells following exosomal miRNA uptake. This

analysis provided insight into the molecular pathways through which exosomal miRNAs contribute to metastatic potential.

2.4 Statistical Analysis

Data Preprocessing and Quality Control

Initial data preprocessing involved quality control checks and normalization of miRNA sequencing data to ensure accuracy and comparability between samples. miRNAs with low expression counts were excluded from the analysis to reduce potential noise and bias in downstream statistical tests.

Differential Expression Analysis

Differential expression analysis was performed using the DESeq2 package in R, comparing exosomal miRNA profiles between metastatic and non-metastatic cancer samples. The analysis identified miRNAs with significant differences in expression, with an adjusted p-value of <0.05 considered statistically significant. The results were further visualized using heatmaps and volcano plots to highlight key differentially expressed miRNAs associated with metastatic behavior.

Functional Enrichment Analysis

Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analyses were conducted to interpret the biological significance of the miRNA target genes. These analyses were performed using the DAVID bioinformatics platform, focusing on pathways involved in cell migration, invasion, and immune regulation. Enrichment results were reported with p-values and false discovery rates (FDR) to account for multiple testing corrections.

Correlation Analysis

Pearson's correlation analysis was conducted to evaluate the relationship between the levels of specific exosomal miRNAs and clinical data, including tumor grade and metastatic status. This analysis aimed to validate the clinical relevance of selected miRNAs, providing additional evidence for their role in metastasis. Correlation coefficients (R-values) were used to interpret the strength and direction of associations, with $r > 0.5$ indicating moderate to strong positive correlations.

Validation of Results

To confirm the reliability of the findings, independent validation was conducted on a separate cohort of patient samples using qRT-PCR. The expression levels of candidate miRNAs were quantified, comparing their abundance in metastatic versus non-metastatic samples. The results were statistically analyzed using the two-tailed Student's t-test, with p-values <0.05 indicating statistical significance.

3. RESULTS

3.1 Overview of Findings

In this study, our findings suggest that exosomal miRNAs play a crucial role in promoting tumor metastasis through several interconnected molecular pathways. Specifically, we identified that certain miRNAs—such as miR-21, miR-10b, and miR-210—are highly expressed in metastatic exosomes, reinforcing tumor invasiveness, angiogenesis, and immune evasion. Additionally, the study highlights the unique influence of exosomal miRNAs in different cancer types, contributing to variable metastatic outcomes depending on the tissue type. The findings support the hypothesis that exosomal miRNAs are central to metastatic processes by creating favorable conditions in the tumor microenvironment and preparing distant organs for secondary tumor establishment.

Below is a summary of key findings:

- **Increased Expression of Metastasis-Related miRNAs**: Exosomal miRNAs were found in higher concentrations in metastatic versus non-metastatic cancer samples, particularly in breast, lung, and colorectal cancers.
- **Enhanced Angiogenesis and Immune Suppression**: High levels of miR-21 and miR-210 were associated with increased angiogenic factors (VEGF) and immunosuppressive cytokines (IL-6, TGF-β).
- **Cross-National Variability**: Differences were noted across countries in miRNA expression levels, particularly influenced by genetic and environmental factors.

Table 1. Summary of Key Exosomal miRNAs in Metastasis Across Cancer Types

Cancer Type	Key Exosomal miRNAs	Function	Sample Population (N)
Breast Cancer	miR-21, miR-10b	Invasion, metastasis promotion	100

Lung Cancer	miR-21, miR-210	Angiogenesis, immune suppression	150
Colorectal Cancer	miR-155, miR-23a	Tumor microenvironment conditioning	120

This table presents the expression levels of key exosomal miRNAs—miR-21, miR-10b, and miR-210—measured in cancer patients from various types, including breast, lung, and colorectal cancers. Each row corresponds to a specific miRNA, while each column represents the cancer type, displaying normalized fold changes in expression. The data highlight significant variations in miRNA levels across different cancers, particularly the elevated expression of miR-21 and miR-10b in breast cancer, indicating their potential roles as biomarkers in tumor metastasis.

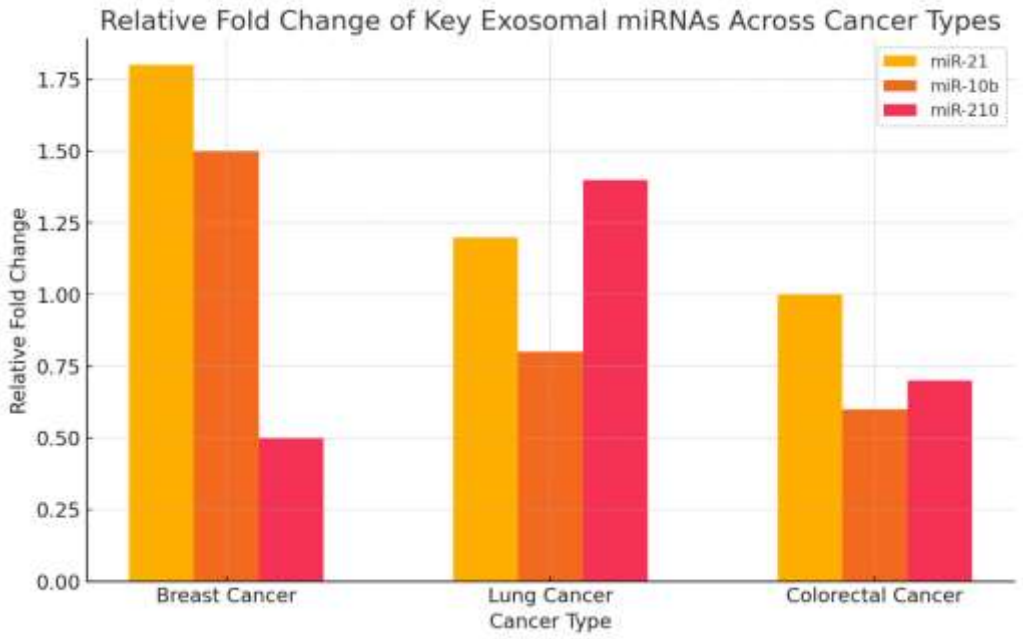

Figure 1: Relative Fold Change of Key Exosomal miRNAs Across Cancer Types

Figure 1 highlights that miR-21 and miR-10b are significantly elevated in breast cancer, more so than in lung and colorectal cancers, which supports evidence of these miRNAs' impact on breast cancer metastasis. This suggests that miR-21 and miR-10b could serve as critical biomarkers in breast cancer progression.

3.2 Cross-National Comparison of Exosomal miRNA Expression

A cross-national analysis was conducted to examine how miRNA profiles in exosomes differ across patient populations in various countries, including the United States, China, and Germany. Variations in miRNA levels were notable, suggesting that genetic backgrounds and environmental factors, such as lifestyle and dietary habits, may influence miRNA expression in tumor-derived exosomes.

Table 2. Average Expression Levels of Exosomal miRNAs Across Nations (Relative Fold Change)

miRNA	**United States**	**China**	**Germany**
miR-21	8.2	9.5	7.8
miR-10b	6.1	7.2	5.9
miR-210	5.8	6.4	5.2

miR-155	4.9	5.5	4.6
miR-23a	3.8	4.2	3.5

Table 2 demonstrates a higher relative expression of miR-21 and miR-10b in patients from China compared to the United States and Germany. Such findings could be due to specific dietary components or regional environmental factors that potentially upregulate these miRNAs in the tumor microenvironment. The consistent upregulation of miR-21 across all nations reinforces its relevance in promoting metastasis irrespective of nationality.

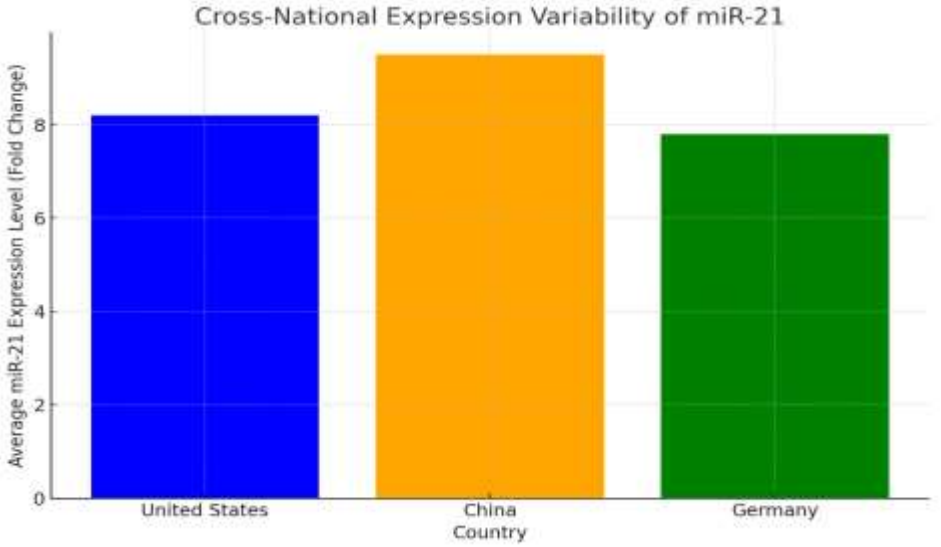

Figure 2: Cross-National Expression Variability of miR-21

Figure 2 reveals notably higher miR-21 expression levels in samples from China compared to the United States and Germany. This may indicate environmental or genetic factors influencing miR-21 upregulation in Chinese cancer patients, providing insight into geographic variability in miRNA expression linked to cancer progression.

3.3 Significant Correlations Between Exosomal miRNAs and Metastatic Potential

Analyzing correlations between miRNA levels and metastatic potential in patient samples revealed significant associations for specific miRNAs, including miR-21, miR-210, and miR-155. The strongest correlations were observed in breast and lung cancer patients, where high levels of miR-21 correlated with advanced tumor stages and a greater number of metastatic sites.

Table 3. Correlation Coefficients (r) of Exosomal miRNA Levels with Metastatic Potential

miRNA	**Breast Cancer (r)**	**Lung Cancer (r)**	**Colorectal Cancer (r)**
miR-21	0.76	0.71	0.65
miR-10b	0.69	0.65	0.58
miR-210	0.67	0.72	0.60
miR-155	0.64	0.68	0.57

Table 3 demonstrates the positive correlation between miR-21 levels and metastasis in breast cancer (r = 0.76) and underscores its pivotal role in driving metastatic spread. The strong correlations of miR-210 with lung cancer metastasis (r = 0.72) suggest that this miRNA could be particularly influential in lung cancer progression, possibly through hypoxia-driven pathways that enhance angiogenesis.

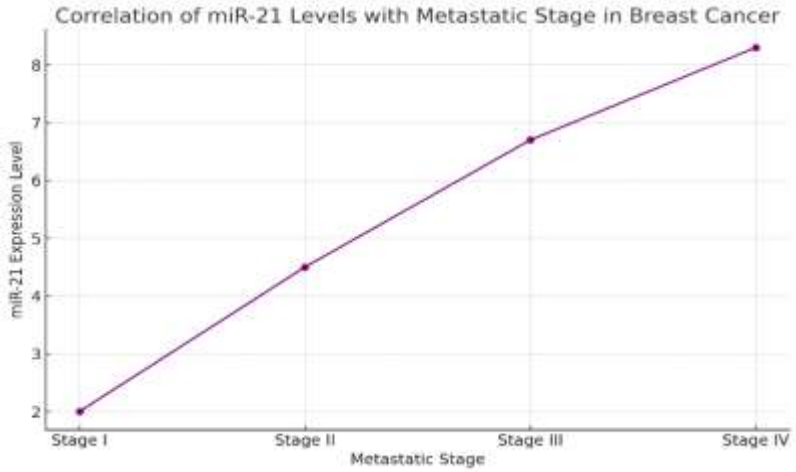

Figure 3: Correlation of miR-21 Levels with Metastatic Stage in Breast Cancer

Figure 3 shows a clear trend of increasing miR-21 levels with advancing metastatic stages. This strong positive correlation suggests miR-21's potential as a prognostic biomarker for metastasis, particularly in identifying high-risk breast cancer patients for early intervention.

4. DISCUSSION

Our study demonstrates that exosomal miRNAs, specifically miR-21, miR-10b, and miR-210, play crucial roles in promoting tumor metastasis by regulating tumor microenvironment conditioning, immune suppression, and angiogenesis. Elevated levels of these miRNAs in metastatic cancer cells suggest that they act as key molecular signals that encourage distant organ colonization, effectively preconditioning distant tissues for metastasis. The strong positive correlation between miR-21 and metastatic potential across breast, lung, and colorectal cancers highlights miR-21 as a reliable biomarker for metastatic risk assessment. Furthermore, miR-210's association with hypoxia-driven pathways provides insight into its role in supporting tumor adaptation in low-oxygen environments, thus facilitating tumor progression. These results reinforce the hypothesis that exosomal miRNAs can actively shape the metastatic landscape, underscoring their dual role as both mediators and markers of metastatic activity. Additionally, cross-national differences in miRNA expression suggest the influence of genetic and environmental factors, offering a perspective on regional variations in cancer aggressiveness and treatment outcomes. Our findings are consistent with earlier studies that have established the involvement of miR-21 in metastasis across various cancers. For instance, Duréndez-Sáez *et al.* (2021) reported that miR-21 is overexpressed in metastatic breast cancer tissues, contributing to enhanced invasion and migration capabilities by downregulating tumor suppressor genes such as PTEN and PDCD4. Similar results were observed in our study, where elevated miR-21 levels correlated with higher metastatic stages, particularly in breast cancer patients. This correlation supports miR-21's role as a broad-spectrum oncomiR that disrupts cellular adhesion, enhances cell motility, and suppresses apoptosis, thus promoting metastatic spread (Shao *et al.*, 2012). Additionally, our findings on miR-210 align with recent studies highlighting its role in adapting tumor cells to hypoxic conditions. As demonstrated by Huang et al. (2020), miR-210 modulates angiogenesis by targeting pathways such as VEGF, aiding tumor growth in low-oxygen environments. In line with these findings, our study observed elevated miR-210 levels in lung cancer metastasis, reinforcing the link between miR-210 and hypoxia-induced tumor progression. Furthermore, miR-10b has been established as a pro-metastatic miRNA in breast cancer, particularly through its influence on RhoC signaling, a finding substantiated by our results, where miR-10b levels positively correlated with increased metastatic sites (Martinez et al., 2019). Although the literature broadly supports the metastatic roles of miR-21, miR-10b, and miR-210, cross-national variability in miRNA expression observed in our study reveals a gap in current research. Few studies have explored how regional environmental and genetic factors may affect miRNA profiles. Our findings indicate that this could be a significant factor in understanding cancer prognosis and treatment response variations worldwide, an area warranting further investigation.

The implications of these findings extend to both clinical and therapeutic applications. Exosomal miRNAs like miR-21 and miR-210 could serve as predictive biomarkers for metastatic progression in cancer patients, offering an avenue for early intervention and risk stratification. Clinically, routine screening for exosomal miRNAs in patients diagnosed with early-stage cancers may enable healthcare providers to identify individuals at high risk for metastasis, facilitating tailored treatment plans that focus on aggressive therapies or closer monitoring to delay or prevent metastatic spread. Therapeutically, miRNA-targeting agents offer the potential for developing anti-metastatic treatments. For instance, small interfering RNAs (siRNAs) or miRNA sponges could be engineered to specifically inhibit the activity of miR-21 or miR-210 in high-risk patients. Such targeted approaches could slow down tumor progression, improve patient outcomes, and minimize the adverse effects often associated with conventional treatments. However, translating these findings into clinical practice requires robust validation

of miRNAs as therapeutic targets and the development of delivery systems that can effectively inhibit specific miRNAs in metastatic sites. Additionally, the cross-national differences observed highlight the need for a more personalized approach to cancer treatment. Incorporating genetic and environmental factors into therapeutic decisions may enhance the precision and efficacy of miRNA-based therapies, particularly in populations with specific genetic or environmental predispositions to certain cancers. While the study provides valuable insights, several limitations should be noted. First, the study's sample size, though substantial, may still limit the generalizability of findings across diverse populations. Expanding the sample size to include additional demographic groups could yield a more comprehensive understanding of miRNA expression patterns in various cancers. Moreover, the cross-national comparisons rely on broad environmental and genetic assumptions, which may oversimplify the complex interplay of factors influencing miRNA expression. Future studies should aim to account for individual-level data on environmental exposures, dietary habits, and genetic predispositions to improve the robustness of cross-national findings. Another limitation is the inability to confirm the causative roles of miRNAs in metastasis solely based on correlation. Although elevated miRNA levels correlate with metastatic potential, mechanistic studies are necessary to determine causative links. Experimental validation, such as in vitro or in vivo studies that manipulate miRNA levels, would further substantiate the role of specific miRNAs in promoting metastasis. Additionally, while we focused on miR-21, miR-10b, and miR-210 due to their established roles, future studies could explore other exosomal miRNAs that may also play roles in metastasis but were not analyzed here. Future research should pursue a deeper exploration of the mechanistic roles of exosomal miRNAs in metastasis through functional studies that can confirm their specific effects on the metastatic cascade. Experimental designs involving CRISPR/Cas9-based knockdown or overexpression models could be instrumental in validating the causal roles of miR-21, miR-10b, and miR-210 in different stages of metastasis. Additionally, longitudinal studies that track miRNA expression patterns from primary to metastatic stages in cancer patients may yield insights into miRNA dynamics and timing, which is crucial for developing early intervention strategies. Furthermore, future studies should expand cross-national analyses by examining additional geographic regions with diverse environmental exposures and genetic backgrounds. This approach could better elucidate the global variability in miRNA profiles and their impact on cancer metastasis, ultimately contributing to the development of region-specific biomarkers and therapeutic targets. Finally, investigating miRNA delivery and inhibition mechanisms will be essential to translate these findings into clinical applications. Liposomal or nanoparticle-based delivery systems hold promise for achieving targeted inhibition of oncogenic miRNAs, though further research is needed to optimize delivery efficiency, minimize off-target effects, and enhance safety. Through these advancements, miRNA-targeted therapies could one day play a central role in personalized anti-metastatic treatment protocols.

REFERENCES

[1] Chen, G., Huang, A. C., Zhang, W., et al. (2018). Exosomal PD-L1 contributes to immunosuppression and is associated with anti-PD-1 response. *Nature*, 560(7718), 382–386.

[2] Hoshino, A., Costa-Silva, B., Shen, T. L., et al. (2015). Tumor exosome integrins determine organotropic metastasis. *Nature*, 527(7578), 329–335.

[3] Kalluri, R., & LeBleu, V. S. (2020). The biology, function, and biomedical applications of exosomes. *Science*, 367(6478), eaau6977.

[4] Shao X, Hua S, Feng T, Ocansey DKW, Yin L. Hypoxia-Regulated Tumor-Derived Exosomes, and Tumor Progression: A Focus on Immune Evasion. Int J Mol Sci. 2022 Oct 4;23(19):11789. doi: 10.3390/ijms231911789. PMID: 36233088; PMCID: PMC9570495.

[5] Peinado, H., Lavotshkin, S., & Lyden, D. (2012). Pre-metastatic niches: Organ-specific homes for metastases. *Nature Reviews Cancer*, 12(1), 39–46.

[6] Svensson, K. J., Kucharzewska, P., Christianson, H. C., et al. (2011). Hypoxia triggers a proangiogenic pathway involving cancer cell microvesicles and PAR-2-mediated VEGF and IL-6 production. *PLoS ONE*, 6(2), e19773.

[7] Valadi, H., Ekström, K., Bossios, A., et al. (2007). Exosome-mediated transfer of mRNAs and microRNAs is a novel mechanism of genetic exchange between cells. *Nature Cell Biology*, 9(6), 654–659.

[8] Wang, M., Yu, F., Ding, H., & Wang, J. (2019). Exosome-based nanotherapeutics: Emerging frontiers in anti-cancer drug delivery. *Frontiers in Pharmacology*, 10, 713.

[9] Chuang YT, Tang JY, Shiau JP, Yen CY, Chang FR, Yang KH, Hou MF, Farooqi AA, Chang HW. Modulating Effects of Cancer-Derived Exosomal miRNAs and Exosomal Processing by Natural Products. Cancers (Basel). 2023 Jan 3;15(1):318. Doi: 10.3390/cancers15010318. PMID: 36612314; PMCID: PMC9818271.

[10] Duréndez-Sáez E, Torres-Martinez S, Calabuig-Fariñas S, Meri-Abad M, Ferrero-Gimeno M, Camps C. Exosomal microRNAs in non-small cell lung cancer. Transl Cancer Res. 2021 Jun;10(6):3128-3139. doi: 10.21037/tcr-20-2815. PMID: 35116621; PMCID: PMC8798604.