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■ REVIEW ARTICLE

Liquid Biopsy in Non-Small Cell Lung Cancer: A Systematic Review of Diagnostic and Prognostic Applications

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ABSTRACT: Lung cancer is the primary cause of cancer death worldwide, in addition to being the most prevalent type of cancer. Non-small cell lung cancer (NSCLC) is the most common form of lung cancer, accounting for 85% of all forms of lung cancer. Liquid biopsy in NSCLC provides a non-invasive tool that determines biomarkers in body fluids with the aim of early detection, treatment monitoring/selection, prognosis estimation, and minimal residual disease (MRD) detection. This review focuses on NSCLC. Articles with diagnostic and prognostic applications of liquid biopsy in NSCLC were collected, and 19 articles published in the last five years (2019-2024) were selected, excluding systematic and literature reviews. All the original papers examined biomarkers in blood in NSCLC patients. Human studies involving solely lung cancer were included. CTCs, ctDNAs, circulating miRNAs, and exosomes were promising in diagnosis and prognosis. However, most studies were done on advanced-stage patients with liquid biopsy's prognostic applications. The most mutated genes were EGFR, KRAS, TP53, ALK, and HER2. Consequently, a need exists to focus on early-stage NSCLC and develop multi-analyte tests using ctDNA features for diagnostic applications.

KEYWORDS: Translational Medical Sciences; Disease Detection and Diagnosis; Non-Small Cell Lung Cancer; Liquid Biopsy; Biomarkers.

■ Introduction

Lung cancer is the primary cause of cancer death worldwide, in addition to being the most prevalent type of cancer. About a fifth of every cancer death is linked to lung cancer. Tobacco use is the major cause of the development of lung cancer.[1] Previous lung disease (such as chronic obstructive pulmonary disease) can also be associated with cancer formation.[2] Non-small cell lung cancer (NSCLC) and small cell lung cancer (SCLC) are the two dominant forms of lung cancer. Roughly 85% of all forms of lung cancer are NSCLC. Mutations in KRAS, EGFR, TP53, ALK, and HER2 are widespread in NSCLC.[1] Distant metastases are present in approximately 50% of lung cancer patients at diagnosis, highlighting the importance of identifying new biomarkers to enable early diagnosis before metastasis and personalized treatment according to cancer type.[3]

Liquid biopsy is the sampling and analysis of tumor DNA fragments or cells originating from the tumor in various body fluids such as blood. Detecting tumor cells or DNA released into the bloodstream by apoptosis, necrosis, or secretion makes liquid biopsies a valuable tool in the elucidation of the features of cancer. Hence, liquid biopsies can identify important tumor features, including significant gene alterations such as point mutations, large deletions, and duplications in the cancer genome. Liquid biopsies provide a non-invasive method that can surmount the limitations of tumor tissue biopsy, such as the difficulty of the biopsy site in obtaining a sample and complications associated with the biopsy process. Biomarkers (measurable, accurate, and sensitive patterns that are specific to a type of cancer) found in blood are circulating tumor DNAs p_1^3 (ctDNA), circulating tumor cells (CTC), circulating microRNAs (miRNA), exosomes, and tumor-educated platelets (TEP).

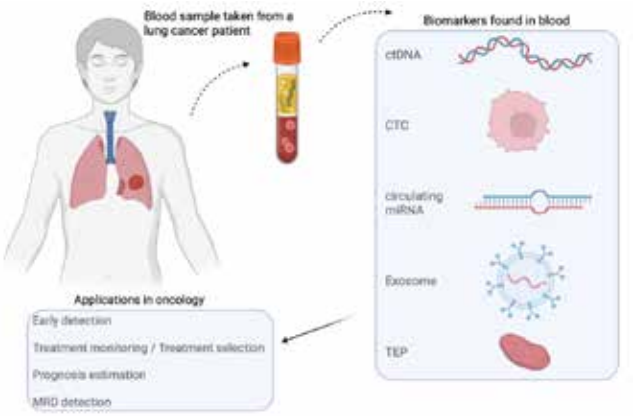

Figure 1: Liquid biopsy biomarkers in blood and clinical applications. ctDNAs, CTCs, microRNAs, exosomes, and TEPs have early detection, treatment monitoring/selection, prognosis estimation, and MRD detection applications.

As illustrated in Figure 1, biomarkers circulating in the bloodstream have the advantages of detecting minimal residual disease (MRD), early detection, treatment monitoring, aiding in drug selection, and estimating prognosis. Molecular characterization of cancer through liquid biopsy can help clinicians identify medications that are not likely effective based on the tumor's molecular profile. Based on molecular tumor profiling, specific drugs could be selected to better respond to individuals' characteristics, avoiding the side effects of ineffective medications. Hence, patients' life qualities are enhanced, and the "trial

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and error" method is eliminated as the more effective drug is identified before the therapy. This displays the use of liquid biopsy in personalized medicine.

Biomarker values such as CTC concentration over time can be examined to determine how well the patient responds to the treatment. If healthcare providers decide that the patient would benefit more from another medication based on results from tracking cancer biomarkers (or if any resistance evolves to the drug), a change in medication may be made. Moreover, liquid biopsy is valuable not only for treatment monitoring but also for post-treatment monitoring purposes. For example, MRD can be tracked by monitoring specific cancer biomarkers. MRD detection can indicate whether cancer will relapse. Liquid biopsy allows prognosis prediction by detecting tumor-specific mutations and monitoring biomarker concentration. As a result, liquid biopsy in lung cancer is an advantageous tool with diagnostic and prognostic applications. That said, it should also be noted that liquid biopsy involves some limitations and challenges. These limitations include the sensitivity and reliability of the current tests, mainly due to the low concentration of biomarkers in blood. They can be surmounted with the introduction of enhanced technologies and larger volumes of blood sampling. The use of bioinformatics tools can increase the accuracy of tests. Challenges involve the high cost of liquid biopsy assays, the ambiguity regarding the frequency of ctDNA tests for monitoring or screening, and the requirement of 10+ years in randomized trials to evaluate liquid biopsy tests.

Additionally, different research employs different time points in the conduction of liquid biopsies, making it challenging to compare studies. Another major challenge is acquiring assay approval from regulators; however, it is possible to ensure that newly developed liquid biopsy assays comply with regulations by interacting with regulatory authorities promptly during the development process. The potential in liquid biopsy & NSCLC research can be achieved by resolving these limitations and challenges through enhanced clinical integration, standardization, and technological developments, translating into improved outcomes for both diagnosis and prognosis. It is also important to consider the ethical and legal aspects of liquid biopsy procedures. Liquid biopsies provide genetic data that raises questions regarding privacy and security. In addition, to enable patients to make informed decisions, it is imperative to ensure that patients understand the potential impact of liquid biopsy on their treatment plan before the procedure and give informed consent.

Methods

PubMed listed articles with the search command "liquid biopsy lung cancer." Articles were narrowed to "diagnostic and prognostic applications" of liquid biopsy. Of these, 19 articles published in the last five years were selected, excluding systematic and literature reviews. Human studies involving solely lung cancer were included. An overview of these articles is included in Table 1.

Table 1: "First author and publication year, research question, biomarker, targeted gene, method, type of blood collection tube, tumor stage, and number of cases" information regarding the articles included in the review.

First author and publication year	Research question/ Researched link	Biomarker	Targeted gene	Method	Type of blood collection tube	Tumor stage	Number of cases
Melanie Janning 2019	The link between PD-L1 expression of CTCs and resistance to immunotherapy	CTC	PD-L1	EpCAM-based CellSearch system	EDTA	96% metastatic 4% stage III	127
Heather Scharpenseel 2019	New markers for CTC detection in NSCLC	CTC	EGFR, HER3	EpCAM-based CellSearch system	EDTA	I, II, III, IV	45
Francesca Chemi 2019	The link between PV-CTC and prognosis in NSCLC	CTC	-	CellSearch system, WES	-	Early (I-II)	100
Minji Lim 2020	The link between CTC and tumor progression& drug response in EGFR-targeted therapy	CTC	EGFR, T-790M	CellSearch system, PCR	EDTA	Early (I-II)	40
Chimbu Chinniah 2019	CTC and disease recurrence	CTC	-	Adenoviral probe, OncoQuick	-	LA-NSCLC stage (II-III)	48
Jingsi Dong 2019	CTC count and prognosis	CTC	PD-L1	CanPatrol, DXS, Sanger sequencing, FISH	EDTA	I-III	114
Anna Buder 2021	Copy number alterations (CNA) in ctDNA of EGFR-mutated lung adenocarcinoma and prognosis	ctDNA	rrSCNAs (resistance-related genes)	ddPCR, WGS	EDTA	Advanced	43
Edward S. Kim 2022	Applications of blood tumor mutational burden in the prediction of atezolizumab benefit in advanced lung cancer patients	ctDNA	PD-L1	WES, bTMB	-	Locally advanced or metastatic	152
Angela Alama 2019	Quantification of CTC&cell free DNA and prognosis of metastatic NSCLC	CTC and cfDNA	-	ScreenCell CYTO, QIAamp Circulating Nucleic Acid Kit (Qiagen), qPCR	EDTA	Metastatic	89
Hiroaki Akamatsu 2019	The clinical significance of liquid biopsy in disease monitoring	cfDNA	EGFR	QIAmp Circulating Nucleic Acid Kit, digital PCR	EDTA	IIIB: 2 patients IV: 39 Post-operative relapse: 16	57
Pei N. Ding 2021	The link between osimertinib response and plasma T790M mutation load	ctDNA	EGFR	Digital PCR	-	Advanced	139
Julie A. Vendrell 2020	Is combining liquid and tissue biopsy important in predicting NSCLC to SCLC change?	ctDNA	EGFR	QIAamp Circulating Nucleic Acid Kit, NGS, ddPCR	Streck cfDNA blood collection tube	Advanced	3
Yajie Zhang 2019	Use of circulating miRNA ratio signatures to predict prognosis	Ct miRNA	-	miRNeasy Mini Kit	-	Advanced	530
Alfons Navarro 2019	Exosome size and relapse	Exosome	-	ultracentrifugation in a Sorvall MX Plus	EDTA	I-IIIA	61

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				Micro-Ultracentrifuge with S140AT Rotor			
Chuling Li 2019	• Exosomal miRNAs and early NSCLC diagnosis • Diagnostic value of GAS5 in exosomes (Exo-GAS5)	Exosomal miRNA	GAS5	total Exosome Isolation Kit (from serum; Thermo, Carlsbad, CA), PCR	Pro-Coagulation Tube	I-II-III-IV	140
Yi Zhang 2019	Circulating exosomal miRNA and diagnosis of NSCLC	Exosomal miRNA	-	ExoQuick, miRNeasy Micro Kit, qRT-PCR	Eppendorf tube	I-II-III	172
Rafal Dziadziuszko 2022	cfDNA concentration and prognosis of ALK+ NSCLC	cfDNA	ALK	D1000 screentape, NGS	EDTA-2K	Advanced	303
Ran Zhong 2023	ctDNA and MRD detection (meta-analysis)	ctDNA	-	-	-	-	16 studies
Leora Horn 2019	ctDNA in ALK+ NSCLC and monitoring	ctDNA	ALK	Circulating Nucleic Acid Extraction Kit (Qiagen), NGS	EDTA	-	76

Results

Circulating tumor cells (CTCs):

CTCs are extravasated into the bloodstream from the primary tumor site. Inspection of their genome reveals information about cancer's characteristics. CTCs are valuable in terms of their prognostic applications in NSCLC. Even though further research is required to validate pulmonary venous CTCs' therapeutic value, it was shown that patients with detectable CTCs had a higher risk of cancer recurrence. [4] In another study, the use of serial CTC analysis and traditional imaging to identify recurrences in patients with locally advanced non-small cell lung cancer following therapy was investigated. Authors suggested that detectable CTC levels in many individuals substantially predate radiologic signs of cancer recurrence. Thus, when assessing a patient's risk of cancer recurrence, longitudinal monitoring of CTCs in the blood circulation is more significant than radiological evidence. [5] Other research disclosed that the prognosis for disease-free survival (DFS) was poor in patients with many CTCs, MCTCs, or PD-L1+ CTCs in the pulmonary vein. The gene subgroups in tumor tissue and the CTC subtype of non-small cell lung cancer exhibited clear connections. [6] In addition, CTC analyses are valuable in detecting resistance toward therapy. An increase in PD-L1+ CTCs was associated with immunotherapy resistance. [7] Another study demonstrated that examining each pretreatment CTC's unique epithelial/mesenchymal signature was crucial to forecasting a patient's response to medication. [8] The heterogeneity of NSCLC CTCs led a research team to recommend using several markers for CTC enrichment and identification. The authors claim that in NSCLC patients with brain metastases, EpCAM enrichment combined with EGFR and HER3 enrichment would result in more CTCs being detected. [9]

Circulating tumor DNA (ctDNA):

ctDNAs are DNA molecules that circulate in the bloodstream. They are single or double-stranded DNAs released from tumors by necrosis, apoptosis, or secretion.[10] ctDNAs are valuable in terms of their prognostic uses in NSCLC. A study indicated that a high concentration of cell-free DNA is a poor prognostic factor for both OS and PFS, regardless of factors such as age, smoking status, type of tumor, and treatment regimen.[11] Additionally, another study found that low ctDNA levels (maximum somatic allele frequency (MSAF) <1%) indicated a substantially improved PFS with atezolizumab monotherapy than MSAF ≥ 1%.[12] Moreover, plasma cell-free DNA concentration was shown to be useful in terms of prognosis estimation for ALK+ advanced NSCLC patients.[13] Another study proposed using cell-free DNA to predict disease progression in patients treated with afatinib.[14] A study that focused on the histological transformation from SCLC to NSCLC reported that tissue and liquid biopsy should be integrated before and during osimertinib treatment to forecast this kind of histological alteration.[15] ctDNAs are also useful for predicting drug response in NSCLC. Researchers revealed that the presence of resistance-related genes (rrSCNAs) in ctDNAs before the start of osimertinib leads to shorter progression-free survival (PFS) and overall survival (OS). Therefore, the detection of rrSCNAs before starting osimertinib treatment was suggested to have the capacity to help guide treatment.[16] In another study, authors identified a link between plasma T790M mutation load on ctDNA and response to osimertinib treatment.[17] A meta-analysis encompassing 16 studies presented ctDNA MRD as a promising biomarker to predict relapse.[18] ctDNAs were also demonstrated to have clinical utility in identifying patients who would benefit from ensartinib and monitoring whether any resistance develops.[19]

Circulating miRNA:

Circulating miRNAs are small RNA fragments found in the bloodstream. In advanced NSCLC patients, authors identified a 5-miRNA signature that was linked to 3-year survival and thus could predict prognosis.[20]

Exosomes:

Exosomes are extracellular vesicles that contain proteins, nucleic acids, and metabolites. Exosomes have diagnostic uses in NSCLC. Researchers identified noncoding RNA growth arrest-specific transcript 5 (GAS5) in circulating exosomes (Exo-GAS5) that can be an early detection tool.[21] Furthermore, another study demonstrated that exosomal miR-17-5p could have a diagnostic value as it was significantly elevated in NSCLC patients.[22] Exosomes are also valuable in predicting recurrence. A study showed that examining exosome sizes was better than clinical data at predicting relapse in patients who had undergone curative surgery.[23]

Of the studies with specified cancer stages, most were focused on advanced-stage cancer, and of the studies that specified the type of blood collection tubes used, approximately all involved EDTA. Various methods to isolate ctDNA, CTC, circulating miRNA, and exosomes were applied, as listed in Table 1. FDA-approved NSCLC assays are listed below in Table 2.

The table underscores the need to develop new assays for NSCLC that are accessible and reliable.

Table 2: FDA-approved assays for NSCLC.

Assay name	Genetic target	Date of approval
COBAS EGFR Mutation test V2	EGFR	June 2016
Guardant 360 CDx	BRAF, KRAS, EGFR	August 2020
FoundationOne Liquid CDx	BRCA1, BRCA2, ALK, PIK3CA, ATM	August 2020
ONCO/Reveal Dx Lung and Colon Cancer Assay	EGFR	July 2021

Discussion

The variation in the methods applied for blood collection and isolation of ctDNA, CTC, circulating miRNA, and exosomes can lead to pre-analytical issues. Since official bodies have yet to establish standardization of these methods, there are still inconsistencies between studies. Although the type of blood collection tube used is another pre-analytical factor, studies included in our review used almost exclusively EDTA. Implementing standardized and optimized methods would lead to more successful results in translating research into clinical practice. Most studies were done on advanced-stage lung cancer patients. Therefore, there is a need to focus on early-stage NSCLC and liquid biopsy diagnostic applications. Combining multiple biomarkers to develop multi-model tests can enhance the sensitivity of liquid biopsy assays.

Conclusion

Lung cancer deaths account for the majority of cancer deaths worldwide. Promising studies indicate that liquid biopsy can be used diagnostically and prognostically in NSCLC by detecting ctDNA, CTCs, exosomes, and circulating miRNAs. However, the standardization of liquid biopsy methods remains an important issue that needs to be addressed. Liquid biopsy is expected to become widespread to obtain important tumorigenic properties that can be used in prognosis and monitoring and for tissues where biopsy is difficult.

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References

1. Wild, C.; Weiderpass, E.; Steward, B. World Cancer Report. *Wold Health Organization* **2020**.
2. Brenner, D. R.; McLaughlin, J. R.; Hung, R. J. Previous Lung Diseases and Lung Cancer Risk: A Systematic Review and Meta-analysis. *PLoS ONE* **2011**, *6* (3), e17479. https://doi.org/10.1371/journal.pone.0017479.
3. Perisano, C.; Spinelli, M. S.; Graci, C.; Scaramuzzo, L.; Marzetti, E.; Barone, C.; Fabbriciani, C.; Maccauro, G. Soft Tissue Metastases in Lung Cancer: A Review of the Literature. *PubMed* **2012**, *16* (14), 1908–1914.
4. Chemi, F.; Rothwell, D. G.; McGranahan, N.; Gulati, S.; Abbosh, C.; Pearce, S. P.; Zhou, C.; Wilson, G. A.; Jamal-Hanjani, M.; Birkbak, N.; Pierce, J.; Kim, C. S.; Ferdous, S.; Burt, D. J.; Slane-Tan, D.; Gomes, F.; Moore, D.; Shah, R.; Al Bakir, M.; Hiley, C. Pulmonary Venous Circulating Tumor Cell Dissemination before Tumor Resection and Disease Relapse. *Nature Medicine* **2019**, *25* (10), 1534–1539. https://doi.org/10.1038/s41591-019-0593-1.
5. Chinniah, C.; Aguarin, L.; Cheng, P.; Decesaris, C.; Cutillo, A.; Berman, A. T.; Frick, M.; Doucette, A.; Cengel, K. A.; Levin, W.; Hahn, S.; Dorsey, J. F.; Simone, C. B.; Kao, G. D. Early Detection of Recurrence in Patients with Locally Advanced Non–Small-Cell Lung Cancer via Circulating Tumor Cell Analysis. *Clinical Lung Cancer* **2019**, *20* (5), 384-390.e2. https://doi.org/10.1016/j.cllc.2019.04.011.
6. Dong, J.; Zhu, D.; Tang, X.; Qiu, X.; Lü, D.; Li, B.; Lin, D.; Zhou, Q. Detection of Circulating Tumor Cell Molecular Subtype in Pulmonary Vein Predicting Prognosis of Stage I–III Non-Small Cell Lung Cancer Patients. *Frontiers in Oncology* **2019**, *9*. https://doi.org/10.3389/fonc.2019.01139.
7. Janning, M.; Kobus, F.; Babayan, A.; Wikman, H.; Velthaus, J.-L.; Bergmann, S.; Schatz, S.; Falk, M.; Berger, L.-A.; Böttcher, L.-M.; Päsler, S.; Gorges, T. M.; O'Flaherty, L.; Hille, C.; Joosse, S. A.; Simon, R.; Tiemann, M.; Bokemeyer, C.; Reck, M.; Riethdorf, S. Determination of PD-L1 Expression in Circulating Tumor Cells of NSCLC Patients and Correlation with Response to PD-1/PD-L1 Inhibitors. *Cancers* **2019**, *11* (6), 835. https://doi.org/10.3390/cancers11060835.
8. Lim, M.; Park, J.; Lowe, A. C.; Jeong, H.; Lee, S.; Park, H. C.; Lee, K.; Kim, G. H.; Kim, M.-H.; Cho, Y.-K. A Lab-On-a-Disc Platform Enables Serial Monitoring of Individual CTCs Associated with Tumor Progression during EGFR-Targeted Therapy for Patients with NSCLC. *Theranostics* **2020**, *10* (12), 5181–5194. https://doi.org/10.7150/thno.44693.
9. Scharpenseel, H.; Hanssen, A.; Loges, S.; Mohme, M.; Bernreuther, C.; Peine, S.; Lamszus, K.; Goy, Y.; Petersen, C.; Westphal, M.; Glatzel, M.; Riethdorf, S.; Pantel, K.; Wikman, H. EGFR and HER3 Expression in Circulating Tumor Cells and Tumor Tissue from Non-Small Cell Lung Cancer Patients. *Scientific Reports* **2019**, 9 (1). https://doi.org/10.1038/s41598-019-43678-6.
10. Thierry, A. R.; El Messaoudi, S.; Gahan, P. B.; Anker, P.; Stroun, M. Origins, Structures, and Functions of Circulating DNA in Oncology. *Cancer and Metastasis Reviews* **2016**, *35* (3), 347–376. https://doi.org/10.1007/s10555-016-9629-x.
11. Alama; Coco; Genova; Rossi; Fontana; Tagliamento; Giovanna Dal Bello; Rosa; Boccardo; Rijavec; Biello; Longo; Cavalieri; Bruzzo; Grossi. Prognostic Relevance of Circulating Tumor Cells and Circulating Cell-Free DNA Association in Metastatic Non-Small Cell Lung Cancer Treated with Nivolumab. *Journal of Clinical Medicine* **2019**, *8* (7), 1011. https://doi.org/10.3390/jcm8071011.
12. Kim, E. S.; Velcheti, V.; Mekhail, T.; Yun, C.; Shagan, S. M.; Hu, S.; Chae, Y. K.; Leal, T. A.; Dowell, J. E.; Tsai, M. L.; Dakhil, C. S. R.; Stella, P.; Jin, Y.; Shames, D. S.; Schleifman, E.; Fabrizio, D. A.; Phan, S.; Socinski, M. A. Blood-Based Tumor Mutational Burden as a Biomarker for Atezolizumab in Non-Small Cell Lung Cancer: The Phase 2 B-F1RST Trial. *Nature Medicine* **2022**, *28* (5), 939–945. https://doi.org/10.1038/s41591-022-01754-x.
13. Rafal Dziadziuszko; Peters, S.; Mok, T.; D. Ross Camidge; Gadgeel, S. M.; Sai-Hong Ignatius Ou; Konopa, K.; Johannes Noé; Malgorzata Nowicka; Bordogna, W.; Morcos, P. N.; Vlatka Smoljanovic; Shaw, A. T. Circulating Cell-Free DNA as a Prognostic Biomarker in Patients with Advanced ALK+ Non–Small Cell Lung Cancer in the Global Phase III ALEX Trial. *Clinical Cancer Research* **2022**, *28* (9), 1800–1808. https://doi.org/10.1158/1078-0432.ccr-21-2840.
14. Akamatsu, H.; Koh, Y.; Okamoto, I.; Fujimoto, D.; Bessho, A.; Azuma, K.; Morita, S.; Yamamoto, N.; Nakagawa, K. Clinical Significance of Monitoring EGFR Mutation in Plasma Using Multiplexed Digital PCR in EGFR Mutated Patients Treated with Afatinib (West Japan Oncology Group 8114LTR Study). *Lung Cancer* **2019**, *131*, 128–133. https://doi.org/10.1016/j.lungcan.2019.03.021.

DOI: 10.36838/v6i9.11

15. Vendrell, J. A.; Quantin, X.; Serre, I.; Jérôme Solassol. Combinati on of Tissue and Liquid Biopsy Molecular Profiling to Detect Tra nsformation to Small Cell Lung Carcinoma during Osimertinib Treatment. *Therapeutic advances in medical oncology (Print)* **2020**, *12*, 175883592097419-175883592097419. https://doi. org/10.11 77/1758835920974192.
16. Buder, A.; Heitzer, E.; Waldispühl-Geigl, J.; Weber, S.; Moser, T. ; Hochmair, M. J.; Hackner, K.; Errhalt, P.; Setinek, U.; Filipits, M . Somatic Copy-Number Alterations in Plasma Circulating Tumo r DNA from Advanced EGFR-Mutated Lung Adenocarcinoma Patients. *Biomolecules* **2021**, *11* (5), 618–618. https://doi. org/10.3 390/biom11050618.
17. Ding, P. N.; Roberts, T. L.; Chua, W.; Becker, T. M.; Caixeiro, N. J.; Paul de Souza; Gao, B.; Lee, C. K.; Itchins, M.; Westman, H.; Clarke, S. J.; Prunella Blinman; Steven Chuan-Hao Kao; John, T.; Leal, J. L.; Bray, V. J. Plasma Pre-Treatment T790M Relative Alle lic Frequency in Patients with Advanced EGFR-Mutated Non-Small Cell Lung Cancer Predicts Treatment Response to Subseq uent-Line Osimertinib. *Translational lung cancer research (Print)* **2021**, *10* (4), 1623–1634. https://doi.org/10.21037/tlcr-20-1125.
18. Zhong, R.; Gao, R.; Fu, W.; Li, C.; Huo, Z.; Gao, Y.; Lu, Y.; Li, F.; Ge, F.; Tu, H.; You, Z.; He, J.; Liang, W. Accuracy of Minimal Residual Disease Detection by Circulating Tumor DNA Profiling in Lung Cancer: A Meta-Analysis. *BMC Medicine* **2023**, *21* (1). https://doi.org/10.1186/s12916-023-02849-z.
19. Horn, L.; Whisenant, J. G.; Wakelee, H.; Reckamp, K. L.; Qiao, H.; Leal, T. A.; Du, L.; Hernandez, J.; Huang, V.; Blumenschein, G. R.; Waqar, S. N.; Patel, S. P.; Nieva, J.; Oxnard, G. R.; Sanborn, R. E.; Shaffer, T.; Garg, K.; Holzhausen, A.; Harrow, K.; Liang, C. Monitoring Therapeutic Response and Resistance: Analysis of C irculating Tumor DNA in Patients with ALK+ Lung Cancer. *Jou rnal of Thoracic Oncology* **2019**, *14* (11), 1901–1911. https:// doi.or g/10.1016/j.jtho.2019.08.003.
20. Zhang, Y.; Roth, J. A.; Yu, H.; Ye, Y.; Xie, K.; Zhao, H.; Chang, D. W.; Huang, M.; Li, H.; Qu, J.; Wu, X. A 5-MicroRNA Signa ture Identified from Serum MicroRNA Profiling Predicts Surviv al in Patients with Advanced Stage Non-Small Cell Lung Cancer. *Carcinogenesis* **2018**, *40* (5), 643–650. https://doi.org/10.1093/car cin/bgy132.
21. Li, C.; Lv, Y.; Shao, C.; Chen, C.; Zhang, T.; Wei, Y.; Fan, H.; Lv, T.; Liu, H.; Song, Y. Tumor-Derived Exosomal LncRNA GAS5 as a Biomarker for Early-Stage Non-Small-Cell Lung Cancer Di agnosis. *Journal of Cellular Physiology* **2019**, *234* (11), 20721–2072 7. https://doi.org/10.1002/jcp.28678.
22. Zhang, Y.; Zhang, Y.; Yin, Y.; Li, S. Detection of Circulating Ex osomal MiR-17-5p Serves as a Novel Non-Invasive Diagnostic Marker for Non-Small Cell Lung Cancer Patients. *Pathology - R esearch and Practice* **2019**, *215* (8), 152466. https://doi. org/10.101 6/j.prp.2019.152466.
23. Navarro, A.; Laureano Molins; Marrades, R. M.; Moisés, J.; Núr ia Viñolas; Morales, S.; Canals; Joan Josep Castellano; José Ramir ez; Monzó, M. Exosome Analysis in Tumor-Draining Pulmonary Vein Identifies NSCLC Patients with Higher Risk of Relapse aft er Curative Surgery. *Cancers* **2019**, *11* (2), 249–249. https://doi.or g/10.3390/cancers11020249.

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