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■ REVIEW ARTICLE

The Applicability of Liquid Biopsy in Triple-Negative Breast Cancer: A Focused Review

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ABSTRACT: Liquid biopsy has been gaining increased attention in recent years with its considerable potential in diagnostic, predictive, screening, and prognostic assays in general oncology. Triple-negative breast cancer (TNBC) is the subtype of breast cancer that presents no established target for treatment selection thus limiting strategies for treatment. With its high metastatic potential, it has been identified to have a marked genetic instability and mutational burden. To date, traditional chemotherapy and immunotherapy have been used to attempt tumor control; however, TNBC still illustrates a high risk of death and shows a need for novel approaches that may be more effective. Liquid biopsy used at different time periods of TNBC treatment may help to identify unique features that allow for the application of strategies to provide a better outcome. The ability to achieve such over time is due to its ease of application with the ability to obtain information from a minimally invasive approach via blood draw. With the interest to learn more about this technology in the context of TNBC, the objective of this study is to analyze the status of liquid biopsy in TNBC patients through conducting a comprehensive review of current literature.

KEYWORDS: Translational Medical Sciences; Disease Detection and Diagnosis; Breast Cancer; Triple-Negative Breast Cancer; Liquid Biopsy.

■ Introduction

Breast cancer is a pathological condition that occurs in the breast tissue. As the number of breast cancer diagnoses substantially rises worldwide, the current and future role of oncologists in the professional care of cancer patients is progressively challenging. In fact, breast cancer has become the second leading cause of cancer deaths among women across the globe.[1]

Triple-negative breast cancer (TNBC) is a subtype of breast cancer that lacks the fundamental receptors — estrogen, progesterone, and the overexpression of the human epidermal growth factor receptor 2 (HER2) gene.[2] Such results indicate that TNBC does not respond to traditional hormonal therapy medicines or the medicines that target the HER2 protein; the only effective therapeutic treatment for TNBC is chemotherapy.[3] Correspondingly, TNBC is known for its aggressive and invasive nature, with low 5-year survival and a much higher recurrence rate compared to hormone- or HER2-positive breast cancers.[4] TNBC represents about 15% of all invasive breast cancers,[5] and it is proved to be more common in women younger than 40 years of age, those of African or Hispanic descent,6 and with the presence of BRCA1 mutation.[7] In recent years, the use of liquid biopsy and the sampling and analysis of circulating tumor DNA (ctDNA) or tumor cell fragments from blood circulation,[8] was successfully tested with clinical trials in non-small cell lung cancer detection and treatments. Such findings contribute to a greater rate of early diagnosis and play a notable role in early detection and cancer treatment. It also aids the development of individualized therapy and the prediction of cancer prognosis. Several biomarkers have been obtained in previous findings such as exosomes, tumor-associated antigens (TAAs), and tumor-associated autoantibodies (TAAbs);[9] among others, the most studied and used one being ctDNA. Although all biomarkers are still the subject of ongoing research, understanding their role in cancer behavior and the process of carcinogenesis is crucial to further develop efficient diagnostic and screening tools.

Through discussing published literature in the relevant landscape, this review summarizes the latest findings on the clinical application of ctDNA-based liquid biopsy technology in triple-negative breast cancer, including its ultimate efficacy, early diagnosis, personalized treatment, prognosis prediction, and the future research progress of oncology technology.

■ Discussion

Understanding Triple-Negative Breast Cancer and the Triple-Negative Paradox:

Triple-negative breast cancer (TNBC) is a heterogeneous disease that is estrogen receptor (ER) negative, progesterone receptor (PR) negative, and human epidermal growth factor receptor 2 (HER2) negative (Figure 1), based on immunohistochemistry.[10] TNBC only affects 10-20% of the overall breast cancer-positive population and 13 in 100,000 women yearly.[5,11]

For now, established drugs such as trastuzumab or tamoxifen which specifically apply to ER-, PR-, or HER2-positive breast cancer seem unforeseeable in the clinical treatment of TNBC. TNBC is characterized by its typical ductal histology, high grade, high proliferation, and mitotic rates. It is associated with poor prognosis, a high risk of local recurrence, and poor disease-free and cancer-specific survival.[12] In fact, a previous study of 906 early-stage breast cancer patients demonstrated that the TNBC subtype and the increasing number of cancer-positive lymph nodes are largely correlated with a superior local recurrence,[13] further indicating the high malignancy of the disease. Other studies have also proven the

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risk for recurrence in TNBC patients is significantly higher in the first 3 to 5 years after diagnosis than in patients with PR- and ER-positive breast cancers. Another key contributing factor to TNBC's malignancy is that it is not amenable to current targeted therapies. Lack of estrogen, progesterone receptors, and the overexpression of HER2 excludes tailored therapeutic approaches with endocrine and HER2 targeted therapies. With the absence of targets, the mainstay of treatment has been with the use of cytotoxic chemotherapy and more recently the incorporation of immune checkpoint inhibitors.[14] It has also been understood that in triple-negative tumors, there is a clear benefit of introducing these agents at diagnosis prior to surgery (neoadjuvant/preoperative chemotherapy) due to the high risk early on of metastases and the aim of this treatment is to control local disease by reducing local and distant recurrence.[15]

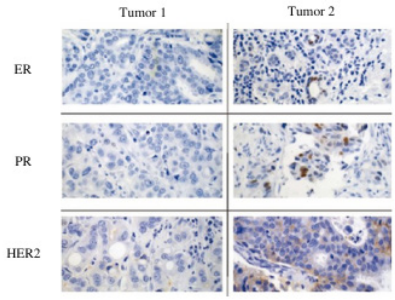

Figure 1: Tumor 1 is a triple-negative tumor (ER, PR, and HER2 negative) whereas Tumor 2 is ER, PR, and HER2 positive. The image is an example of stained breast cancer tissue; the brown color indicates the presence of ER, PR, and HER2 receptors, which indicates the microscopic absence of the 3 key targeting receptors in TNBC.

The "paradox of TNBC" is that early on the disease is extremely chemotherapy responsive in general but the effects can be short-lived as there are many patients who do enjoy the reduction of disease but those who do not achieve complete remission of cancer (referred to as a pathologic complete remission or pCR) at the time of surgical resection after this treatment, experience high rates of recurrence and death.[16] This paradox of TNBC is also used because the more additional exposure to patients with TNBC after the initial administration of cytotoxic chemotherapy, the more observations of relative resistance to chemotherapy in spite of initial responsiveness. Additionally, patients with metastatic TNBC rather than non-TNBC typically have poorer survival. The CALGB 9342 study concluded that overall survival was significantly shorter in those with TNBC compared with those with other subtypes, despite a similar rate of objective response and times to treatment failure.[17] Furthermore, studies focusing on patients with metastatic TNBC concluded a more rapid progression during multiple rounds of chemotherapy, which is another consolidative evidence of its high relapse rates and low survival.[18,19]

TNBC's extreme malignancy, poor prognosis, and high recurrence rate further highlight an urgent need for more effective treatment options and more focused diagnostic tools to assist early detection of the disease.

Circulating Tumor DNA in Triple-Negative Breast Cancer Sample: Predictor of Prognosis:

Circulating tumor DNA (ctDNA) derived from tumors or cancer cells is present in the cancer patients' plasma. Essentially, ctDNA is the extracellular DNA molecule (double-stranded and mitochondrial DNA) that came from cells found in the patients' body fluids.[20] Early studies showed that ctDNA possessed many cancer-associated molecular characteristics, such as single-nucleotide mutations, methylation changes and cancer-derived viral sequences, and therefore were considered to be derived from tumor tissue. These findings were significant for the development of future ctDNA detection technology.

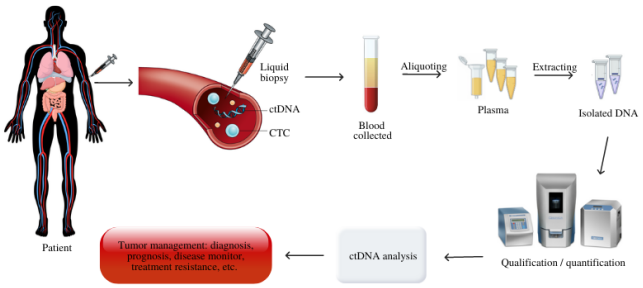

Figure 2: The process of liquid biopsy ctDNA analysis.

Initially described in 1948, both diseased and normal cells release circulating cell-free DNA (cfDNA) into circulation through active and passive transport. Typically, cfDNA is fragmented and its length strands suggest that DNA is released into the bloodstream through cell death.[20] In healthy individuals, cfDNA is thought to be mainly of hematopoietic origin and is usually found at lower levels (approximately 1–10 ng/mL of plasma). However, patients with cancer can have levels 5 to 10 times those of healthy individuals. There is evidence that a proportion of fragmented DNA is derived from the tumor itself; this phenomenon is termed circulating tumor DNA (ctDNA). The seminal research by Stroun revealed that ctDNA fragments harbor similar genomic alterations as a patient's tumor. Since then, others have detected ctDNA in various solid malignancies. Intriguingly, tumor-derived DNA fragments appear to be sizably smaller (approximately 143 to 145 bp) than the normal cfDNA fragment-size distribution (peaks around 166 bp). Therefore, genomic changes, epigenetic markers, and fragment size can be used to detect the presence of ctDNA in the circulation and to infer the tissue of origin.[20]

The majority of available studies on the use of ctDNA deal with genetic analysis and mutation detection. The analysis of ctDNA (Figure 2) is often discussed in the context of the non-invasive approach to mutation detection that leads to mechanism resistance and therapeutic management in patients.[21] With the development of highly sensitive methods and the ability to interrogate a large quantity of genes, substantial advancements have been made in this area of cancer detection. Interestingly, such a method is also used to analyze different features of the sample's DNA, such as methylation status, size fragment patterns, transcriptomics, and viral load,[21] which opens new paths for analyzing samples of liquid biopsy from cancer patients.

Past clinical investigations of ctDNA-based liquid biopsy present major success in non–small cell lung cancer, where examination and diagnosis of peripheral blood with tumor antigens are routinely used in clinical examinations.[9,22] In the TRACERx study, results showed that ctDNA could be detected up to 6–12 months prior to a lung cancer diagnosis with imaging, because cancer-related chemical abnormalities and genetic mutation precede the appearance of detectable abnormalities, such as visible masses, in chest imaging.[9,23] This significantly reduces or even possibly eliminates the risk of exposure to radiation by multiple follow-up imaging for stable lung lesions and reduces high false-positive rates and overdiagnosis seen in imaging screening.[23] As a result, with previous evidence of success, liquid biopsy is emerging as a promising method for disease progression identification for cancer after adjuvant treatments or surgery, as well as the longitudinal monitoring for therapy response and disease progression.

Circulating Tumor DNA in Triple-Negative Breast Cancer Sample: Predictor of Prognosis:

In previous findings of retrospective studies, ctDNA has an excellent ability to predict relapse and prognosis in TNBC patients who have completed their primary treatment.[24] With sufficient supporting data, further stratification of TNBC prognosis can be achieved by incorporating ctDNA detection through liquid biopsy after the delivery of curative treatment. The ongoing phase II trial, cTRAK TN (NCT03145961), aims to investigate whether ctDNA screening can be used to detect cancer residue following patients' primary TNBC treatment.[25] Dr. Nicholas C. Turner and his colleagues called this study "the first study to assess whether ctDNA assays can guide therapy for patients with early-stage TNBC".[26] Patients undergo double-blinded ctDNA screening every 3 months after the completion of primary treatment for TNBC. The trial's recent data found that using ctDNA early often assists the process of determining relapse of TNBC.[25,26] This finding, if confirmed, would have significant implications for future study designs and allow for earlier intervention for cancer control which could impact tumor management and survival. It has also been observed that using liquid biopsy to identify mutations earlier in the patient experience may afford an opportunity to identify the development of variants and with more frequent monitoring, changes like these and other changes may be helpful for treatment approaches. Though this technology appears to be helpful in many patients, there have been findings that show it is not always helpful in some patients at risk for relapse.[27] With this limitation acknowledged, this detection method, because of its ease in utilization, has the potential to be an invaluable tool in many cancers including TNBC. Ongoing studies and measures to finetune this technology to provide clinically meaningful application will hopefully be forthcoming.

Comparing Tissue Biopsy and Liquid Biopsy: The Effects of Liquid Biopsy on Triple-Negative Breast Cancer:

General biopsy is a fundamental tool for clinical assessment of cancer, aiding diagnosis and disease management.[28] In an oncology setting, tissue biopsy is traditionally used for identifying and classifying diseases and profiling genetic mutations of cancer. However, such a historical method presents many restraints such as the concerns over single-biopsy (Figure 3). Previous findings demonstrate a significant intertumoral and intratumoral heterogeneity between primary tumor sites and metastasis sites. Consequently, a traditional surgical biopsy is proven to be not feasible due to its underestimation of tumor complexity and failure to see an overall picture of the disease.[29] These limitations highlighted the need for a minimally-invasive, more modern approach to the real-time monitoring of TNBC. Unlike traditional biopsy, which usually involves either a surgical approach or a minimally-invasive procedure to collect tumor tissue, liquid biopsy can easily be done from a simple blood draw of the patient's plasma.[29,30] Briefly, mutation detection in ctDNA-based liquid biopsy is mainly used for the purpose of:[31]

- Diagnosing cancers prior to traditional detection such as radiology
- Customizing cancer treatment through genotyping
- Identifying mechanisms of resistance occurred in therapies to aid a better understanding of the disease
- Measuring and monitoring residual disease after treatment
- Inspecting cancer resistance or relapse to the already-administered treatments

Although tissue biopsy can sometimes be more affirmative, liquid biopsy is performed on a basis of easy access, allowing easier and more cost-efficient use,[32] particularly among those who cannot have surgery. The use of liquid biopsy can decrease the time to treatment based on tumor detection, improve staff and resource efficiency, and can be used to test other diseases with greater scope. Moreover, it avoids postoperative complications of standard tissue biopsy, including the risk of more cancer spread, injury to surrounding tissues, and severe bleeding from the incision. In cases of tumors that show regional differences, liquid biopsy is able to test tumor DNA fragments from different sites of the tumor and thus supply a more thorough genomic picture.[33] With that being said, oncologists can more flexibly use molecularly-targeted therapies, especially in cases of TNBC where targeted therapies are difficult to plan and execute. Likewise, liquid biopsy may bring a faster and more innovative method to select future TNBC clinical trial candidates and monitor response to the tested drug or agent.

Type of Biopsy	Advantage	Disadvantage
 Tissue Biopsy	• Important for research and discovery • Clinically validated • Critical if non-DNA biomarkers are needed	• Invasive, risky, time-consuming • Failure to detect metastasis at distant sites • Impractical for periodic monitoring of treatment response • Surgical post-operative complications
 Liquid Biopsy	• Non-invasive, easy to obtain samples • Captures heterogenous alterations • Provides comprehensive tissue profile • Minimal pain/risk • Rapid turnaround & high sensitivity • Real-time monitoring of drug response/resistance	• Clinical practices not yet established • Can not assess non-DNA biomarkers • Does not provide histological evaluation • Higher false positive rates

Figure 3: Comparing the advantages and disadvantages between tissue and liquid biopsy.

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Potential Challenges of the Clinical Adoption of Liquid Biopsy:

Although a growing number of liquid biopsies are being performed, they still must reach two crucial goals before full adoption: its full clinical validation and the development of standards/regulations for their future adoption. In addition, a few areas include:[34-36]

- In order to receive appropriate treatment decisions, strengthened clinical data must be established to create clinically beneficial outcomes: experimental studies must gain more knowledge on the biology of liquid biopsy markers (including the emerging markers beyond ctDNA such as extracellular vesicles, tumor-educated platelets or circulating microRNAs) which can then in turn be retranslated to the bedside to improve the clinical use of liquid biopsy analytes.
- Requires a deeper understanding of aspects of biomarkers and cancer biology, otherwise, risks for false results of liquid biopsy are possible: results from previous studies support the clinical validity of ctDNA liquid biopsy for improved risk assessment (staging), monitoring of cancer therapies and early detection of relapse in cancer patients. However, it should be noted that the concentration of ctDNA is also low in early-stage cancer patients, which has stimulated the recent development of ultrasensitive ctDNA assays. Applying these assays has revealed a background of cancer-associated mutations in normal white blood cells, which may contaminate the ctDNA fraction.
- Despite all the progress being made in blood-based technology, there are still limited standards to address specific processes: these crucial steps include extraction methods and analytic criteria (such as determination of ctDNA, circulating tumor cell positivity assay cutoffs interpretation and reporting, and clinical utility).
- Before liquid biopsies can become an integrated part of regular oncologic practices, physician education must be ensured: their sophisticated understanding of the utility and limitations of liquid biopsies enables the correct and most effective implementation of the technology for each patient.

Estimates for Application:

Research reports estimated a $6 billion market value and $26 billion market size for liquid biopsy by 2030.[35] Currently, it has been utilized for diagnosing mainly lung cancer and screening for other types of cancer. It is often applied in clinical settings as a companion diagnostic for cancer detection which confirms the accuracy of performed tests.

Generally, as a breast cancer detection tool, many believe liquid biopsy can improve the diagnostic outcome, and evidence agrees that the more practical use of liquid biopsy technique is for high-risk patients who have a history of cancer or detected genetic mutation.[37] Currently, liquid biopsies are commonly applied as a complementary technique to tissue biopsy;[38] traditional tissue biopsy is still the main tool used for breast cancer and TNBC diagnosis, but for those patients who are unable to undergo treatment or whose tumors do not allow an invasive biopsy, liquid biopsy holds great promise. And hopefully, with more established research, liquid biopsies can gradually be adopted in not only triple-negative breast cancer detection but also for all types of cancer.

The Future of Liquid Biopsies:

In 2020, the United States Food and Drug Administration (FDA) approved Foundation Medicine's companion diagnostic test "FoundationOne® Liquid CDx", which can detect changes in all solid tumor masses with the initial diagnosis. This sequencing test can analyze more than 300 genes and their signatures which will be utilized to guide treatment selection and possible clinical trials for patients.[39] Data from prospective studies have shown that liquid biopsies can even deliver advantages to patients beyond being a non-invasive approach, such as increasing quality-adjusted life years.[40,41] As numerous companies have already taken action, it is closer and closer to making the widespread adoption of liquid biopsies a reality.

Conclusion

Despite an inability to perform clinical experiments to add original knowledge to the field of triple-negative breast cancer, through the format of a literature review, this paper made valuable derivations in the areas of summarizing previous studies and predicting the future role of liquid biopsies. Previous studies and research examined in this review have shown a high potential for the successful application of liquid biopsies in TNBC. However, its large-scale adoption will require further clinical evidence of clear and direct benefits for patients in order to complete full application and transition in hospitals, research institutions, clinics, private practices, etc.[42,43] Ultimately, the present literature supports the validity of liquid biopsy as a minimally invasive diagnostic tool for the early diagnosis and monitoring of therapeutic response, cancer screening in high-risk populations, assessment of tumor heterogeneity, and detection of novel cancer driver mutations. The future of liquid biopsies is believed to have the potential to increase diagnostic options and individualized care for breast cancer.[41] Therefore, with further research and investigations, liquid biopsy marks a promising future in oncology and medicine.

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References

1. Sun, Y.-S.; Zhao, Z.; Yang, Z.-N.; Xu, F.; Lu, H.-J.; Zhu, Z.-Y.; Shi, W.; Jiang, J.; Yao, P.-P.; Zhu, H.-P. Risk Factors and Preventions of Breast Cancer. International Journal of Biological Sciences 2017, 13 (11), 1387–1397. https://doi.org/10.7150/ijbs.21635.
2. Kumar, P.; Aggarwal, R. An Overview of Triple-Negative Breast Cancer. Archives of Gynecology and Obstetrics 2015, 293 (2), 247–269. https://doi.org/10.1007/s00404-015-3859-y.
3. Wahba, H. A.; El-Hadaad, H. A. Current Approaches in Treatment of Triple-Negative Breast Cancer. Cancer biology & medicine 2015 , 12 (2), 106–116. https://doi.org/10.7497/j.issn.2095-3941.2015.0030.
4. Tečić Vuger, A. Characteristics and Prognosis of Triple-Negative Breast Cancer Patients: A Croatian Single Institution Retrospective

Cohort Study. Acta Clinica Croatica 2020, 59. https://doi. org/10.20471/acc.2020.59.01.12.
5. Cleveland Clinic. Triple Negative Breast Cancer: What Is It, Symptoms, Treatment & Survival Rate https://my.clevelandclinic.org/health/diseases/21756-triple-negative-breast-cancer-tnbc.
6. Clarke, C. A.; Keegan, T. H. M.; Yang, J.; Press, D. J.; Kurian, A. W. ; Patel, A. H.; Lacey, J. V. Age-Specific Incidence of Breast Cancer Subtypes: Understanding the Black–White Crossover. JNCI: Journal of the National Cancer Institute 2012, 104 (14), 1094–1101. https://doi.org/10.1093/jnci/djs264.
7. Chen, H.; Wu, J.; Zhang, Z.; Tang, Y.; Li, X.; Liu, S.; Cao, S.; Li, X. Association between BRCA Status and Triple-Negative Breast Cancer: A Meta-Analysis. Frontiers in Pharmacology 2018, 9. https://doi.org/10.3389/fphar.2018.00909.
8. National Cancer Institute. NCI Dictionary of Cancer Terms https://www.cancer.gov/publications/dictionaries/cancer-terms/def/liquid-biopsy.
9. Revelo, A. E.; Martin, A.; Velasquez, R.; Kulandaisamy, P. C.; Bustamante, J.; Keshishyan, S.; Otterson, G. Liquid Biopsy for Lung Cancers: An Update on Recent Developments. Annals of Translational Medicine 2019, 7 (15). https://doi.org/10.21037/ atm.2019.03.28.
10. Aysola, K.; Desai, A.; Welch, C.; Xu, J.; Qin, Y.; Reddy, V.; Matthews, R.; Owens, C.; Okoli, J.; Beech, D. J.; Piyathilake, C. J.; Reddy, S. P.; Rao, V. N. Triple Negative Breast Cancer – an Overview. Hereditary Genetics 2012. https://doi.org/10.4172/2161-1041.s2-001.
11. Sun, B. Triple-Negative Breast Cancer https://www.hopkinsmedicine.org/health/conditions-and-diseases/breast-cancer/triple-negative-breast-cancer.
12. Jiao, Q.; Wu, A.; Shao, G.; Peng, H.; Wang, M.; Ji, S.; Liu, P.; Zhang, J. The Latest Progress in Research on Triple Negative Breast Cancer (TNBC): Risk Factors, Possible Therapeutic Targets and Prognostic Markers. Journal of Thoracic Disease 2014, 6 (9), 1329–1335. https://doi.org/10.3978/j.issn.2072-1439.2014.08.13.
13. Russo, A. L.; Arvold, N. D.; Niemierko, A.; Wong, N.; Wong, J. S.; Bellon, J. R.; Punglia, R. S.; Golshan, M.; Troyan, S. L.; Brock, J. E.; Harris, J. R. Margin Status and the Risk of Local Recurrence in Patients with Early-Stage Breast Cancer Treated with Breast-Conserving Therapy. Breast Cancer Research and Treatment 2013, 140 (2), 353–361. https://doi.org/10.1007/s10549-013-2627-6.
14. Thomas, R.; Al-Khadairi, G.; Decock, J. Immune Checkpoint Inhibitors in Triple Negative Breast Cancer Treatment: Promising Future Prospects. Frontiers in Oncology 2021, 10. https://doi.org/10.3389/fonc.2020.600573.
15. Medina, M. A.; Oza, G.; Sharma, A.; Arriaga, L. G.; Hernández Hernández, J. M.; Rotello, V. M.; Ramirez, J. T. Triple-Negative Breast Cancer: A Review of Conventional and Advanced Therapeutic Strategies. International Journal of Environmental Research and Public Health 2020, 17 (6), 2078. https://doi.org/10.3390/ijerph17062078.
16. Carey, L. A.; Dees, E. C.; Sawyer, L.; Gatti, L.; Moore, D. T.; Collichio, F.; Ollila, D. W.; Sartor, C. I.; Graham, M. L.; Perou, C. M. The Triple Negative Paradox: Primary Tumor Chemosensitivity of Breast Cancer Subtypes. Clinical Cancer Research 2007, 13 (8), 2329–2334. https://doi.org/10.1158/1078-0432.ccr-06-1109.
17. Harris, L. N.; Broadwater, G.; Lin, N. U.; Miron, A.; Schnitt, S. J. ; Cowan, D.; Lara, J.; Bleiweiss, I.; Berry, D.; Ellis, M.; Hayes, D. F.; Winer, E. P.; Dressler, L. Molecular Subtypes of Breast Cancer in Relation to Paclitaxel Response and Outcomes in Women with Metastatic Disease: Results from CALGB 9342. Breast Cancer Research 2006, 8 (6). https://doi.org/10.1186/bcr1622.
18. Lin, N. U.; Claus, E.; Sohl, J.; Razzak, A. R.; Arnaout, A.; Winer, E. P. Sites of Distant Recurrence and Clinical Outcomes in Patients with Metastatic Triple-Negative Breast Cancer. Cancer 2008, 113 (10), 2638–2645. https://doi.org/10.1002/cncr.23930.
19. Kassam, F.; Enright, K.; Dent, R.; Dranitsaris, G.; Myers, J.; Flynn, C.; Fralick, M.; Kumar, R.; Clemons, M. Survival Outcomes for Patients with Metastatic Triple-Negative Breast Cancer: Implications for Clinical Practice and Trial Design. Clinical Breast Cancer 2009, 9 (1), 29–33. https://doi.org/10.3816/cbc.2009.n.005.
20. Alese, O. B., Cook, N., Ortega-Franco, A., Ulanja, M. B., Tan, L., & Tie, J. (2022). Circulating tumor DNA: An emerging tool in gastrointestinal cancers. American Society of Clinical Oncology Educational Book, (42), 279–298. https://doi.org/10.1200/edbk_349143
21. Keller, L.; Belloum, Y.; Wikman, H.; Pantel, K. Clinical Relevance of Blood-Based CtDNA Analysis: Mutation Detection and Beyond. British Journal of Cancer 2020, 1–14. https://doi. org/10.1038/s41416-020-01047-5.
22. Li, W.; Liu, J.-B.; Hou, L.-K.; Yu, F.; Zhang, J.; Wu, W.; Tang, X. -M.; Sun, F.; Lu, H.-M.; Deng, J.; Bai, J.; Li, J.; Wu, C.-Y.; Lin, Q. -L.; Lv, Z.-W.; Wang, G.-R.; Jiang, G.-X.; Ma, Y.-S.; Fu, D. Liquid Biopsy in Lung Cancer: Significance in Diagnostics, Prediction, and Treatment Monitoring. Molecular Cancer 2022, 21 (1). https://doi.org/10.1186/s12943-022-01505-z.
23. Jamal-Hanjani, M.; Wilson, G. A.; McGranahan, N.; Birkbak, N. J.; Watkins, T. B. K.; Veeriah, S.; Shafi, S.; Johnson, D. H.; Mitter, R.; Rosenthal, R.; Salm, M.; Horswell, S.; Escudero, M.; Mathews, N.; Rowan, A.; Chambers, T.; Moore, D. A.; Turajlic, S.; Xu, H.; Lee, S.-M. Tracking the Evolution of Non–Small-Cell Lung Cancer. New England Journal of Medicine 2017, 376 (22), 2109–2121. https://doi.org/10.1056/nejmoa1616288.
24. Dawson, S.-J.; Tsui, D. W. Y.; Murtaza, M.; Biggs, H.; Rueda, O. M.; Chin, S.-F.; Dunning, M. J.; Gale, D.; Forshew, T.; Mahler-Araujo, B.; Rajan, S.; Humphray, S.; Becq, J.; Halsall, D.; Wallis, M.; Bentley, D.; Caldas, C.; Rosenfeld, N. Analysis of Circulating Tumor DNA to Monitor Metastatic Breast Cancer. New England Journal of Medicine 2013, 368 (13), 1199–1209. https://doi.org/10.1056/nejmoa1213261.
25. Institute of Cancer Research, United Kingdom; National Institute for Health Research Biomedical Research Centre at the Royal Marsden / Institute of Cancer Research UK; Merck Sharp & Dohme LLC. c-TRAK TN: A Randomised Trial Utilising ctDNA Mutation Tracking to Detect Minimal Residual Disease and Trigger Intervention in Patients With Moderate and High Risk Early Stage Triple Negative Breast Cancer https://clinicaltrials.gov/ct2/show/NCT03145961.
26. Turner N, Swift C, Jenkins B, et al. Primary results of the cTRAK TN trial: A clinical trial utilising ctDNA mutation tracking to detect minimal residual disease and trigger intervention in patients with moderate and high risk early stage triple negative breast cancer. Presented at: 2021 San Antonio Breast Cancer Symposium; December 7-10, 2021; virtual. Abstract GS3-06.
27. Alba-Bernal, A.; Lavado-Valenzuela, R.; Domínguez-Recio, M. E. .; Jiménez-Rodriguez, B.; Queipo-Ortuño, M. I.; Alba, E.;Comino -Méndez, I. Challenges and Achievements of Liquid Biopsy Technologies Employed in Early Breast Cancer. EBioMedicine 2020, 62. https://doi.org/10.1016/j.ebiom.2020.103100.
28. Mayo Clinic. How biopsy procedures are used to diagnose cancer https://www.mayoclinic.org/diseases-conditions/cancer/in-depth/biopsy/art-20043922.
29. Osei-Bordom, D. C.; Sachdeva, G.; Christou, N. Liquid Biopsy as a Prognostic and Theranostic Tool for the Management of Pancreatic Ductal Adenocarcinoma. Frontiers in Medicine 2022, 8. https://doi.org/10.3389/fmed.2021.788869.
30. Ilié, M.; Hofman, P. Pros: Can Tissue Biopsy Be Replaced by Liquid Biopsy? Translational Lung Cancer Research 2016, 5 (4), 420–

DOI: 10.36838/v5i3.17

423. https://doi.org/10.21037/tlcr.2016.08.06.
31. National Library of Medicine. What is circulating tumor DNA and how is it used to diagnose and manage cancer?: MedlinePlus Genetics https://medlineplus.gov/genetics/understanding/testing/circulatingtumordna/.
32. Hovelson, D. H.; Liu, C.-J.; Wang, Y.; Kang, Q.; Henderson, J.; Gursky, A.; Brockman, S.; Ramnath, N.; Krauss, J. C.; Talpaz, M.; Kandarpa, M.; Chugh, R.; Tuck, M.; Herman, K.; Grasso, C. S.; Quist, M. J.; Feng, F. Y.; Haakenson, C.; Langmore, J.; Kamberov, E. Rapid, Ultra Low Coverage Copy Number Profiling of Cell-Free DNA as a Precision Oncology Screening Strategy. Oncotarget 2017, 8 (52), 89848–89866. https://doi.org/10.18632/ oncotarget.21163.
33. Kwo, L.; Aronson, J. The Promise of Liquid Biopsies for Cancer Diagnosis. Evidence-Based Oncology 2021, 27 (7).
34. Neumann, M. H. D.; Bender, S.; Krahn, T.; Schlange, T. CtDNA and CTCs in Liquid Biopsy – Current Status and Where We Need to Progress. Computational and Structural Biotechnology Journal 2018, 16, 190–195. https://doi.org/10.1016/j.csbj.2018.05.002.
35. Morgan, T. M. Liquid Biopsy: Where Did It Come From, What Is It, and Where Is It Going? Investigative and Clinical Urology 2019, 60 (3), 139. https://doi.org/10.4111/icu.2019.60.3.139.
36. Heidrich, I., Ačkar, L., Mossahebi Mohammadi, P., & Pantel, K. (2020). Liquid biopsies: Potential and challenges. International Journal of Cancer, 148(3), 528–545. https://doi.org/10.1002/ ijc.33217
37. Grand View Report. Liquid Biopsy Market Size & Share Report, 2021-2028 https://www.grandviewresearch.com/industry-analysis/liquid-biopsy-market (accessed 2022 -06 -18).
38. Chen, M.; Zhao, H. Next-Generation Sequencing in Liquid Biopsy: Cancer Screening and Early Detection. Human Genomics 2019, 13 (1). https://doi.org/10.1186/s40246-019-0220-8.
39. Patelli, G.; Vaghi, C.; Tosi, F.; Mauri, G.; Amatu, A.; Massihnia, D.; Ghezzi, S.; Bonazzina, E.; Bencardino, K.; Cerea, G.; Siena, S.; Sartore-Bianchi, A. Liquid Biopsy for Prognosis and Treatment in Metastatic Colorectal Cancer: Circulating Tumor Cells vs Circulating Tumor DNA. Targeted Oncology 2021, 16 (3), 309– 324. https://doi.org/10.1007/s11523-021-00795-5.
40. Health Center for Devices and Radiological. FoundationOne Liquid CDx - P200016. FDA 2020.
41. Sánchez-Calderón, D.; Pedraza, A.; Mancera Urrego, C.; Mejía-Mejía, A.; Montealegre-Páez, A. L.; Perdomo, S. Analysis of the Cost-Effectiveness of Liquid Biopsy to Determine Treatment Change in Patients with Her2-Positive Advanced Breast Cancer in Colombia. ClinicoEconomics and Outcomes Research: CEOR 2020, 12, 115–122. https://doi.org/10.2147/CEOR.S220726.
42. Tay, T. K. Y.; Tan, P. H. Liquid Biopsy in Breast Cancer: A Focused Review. Archives of Pathology & Laboratory Medicine 2020. https://doi.org/10.5858/arpa.2019-0559-ra.
43. Ignatiadis, M.; Sledge, G. W.; Jeffrey, S. S. Liquid Biopsy Enters the Clinic — Implementation Issues and Future Challenges. Nature Reviews Clinical Oncology 2021. https://doi.org/10.1038/s41571-020-00457-x.

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