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Review

Integrating multi-omic liquid biopsies and artificial intelligence: The next frontier in early cancer detection

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ABSTRACT

The integration of multi-omic liquid biopsies with artificial intelligence (AI) represents a rapidly evolving frontier in early cancer detection, offering the potential to enhance personalized medicine and improve patient outcomes. This review explores the current state and emerging directions of this approach, focusing on the synergistic value of combining genomics, epigenomics, transcriptomics, proteomics, and metabolomics with AI-driven analytics. We discuss advances in multi-analyte blood tests such as CancerSEEK, which have demonstrated promising multi-cancer detection capabilities in early studies, as well as efforts to integrate liquid biopsy data with imaging modalities to improve diagnostic performance. The review also highlights ongoing challenges, including the need for greater analytical sensitivity, improved specificity for early-stage disease, standardization of workflows, and harmonization with existing screening modalities. We outline the prospective—but still largely investigational—impact of these technologies on cancer management, including early detection, treatment monitoring, and minimal residual disease assessment, along with their potential economic implications. Ultimately, we envision a future in which multi-omic liquid biopsies integrated with AI may contribute to more effective, noninvasive cancer detection strategies, while recognizing that substantial validation, regulatory approval, and health-system integration are required before widespread clinical adoption can occur.

1. Introduction

1.1. The need for early cancer detection

Cancer is a leading cause of death worldwide, accounting for nearly 10 million deaths in 2020, or nearly one in six deaths. Early detection of aggressive tumors allows for more effective treatment.[1] There is strong evidence that its occurrence will keep rising in the future, driven by lifestyle shifts and the growing elderly population.[2] It has been known that patients diagnosed in the early stages have higher cure rates and improved 5-year survival rates. Medical costs increase considerably with each advancing stage, as treatments become more complex and prolonged. Early interventions are less expensive, while later stages require intensive care, driving up overall healthcare expenses.[3] Cancer is frequently detected through symptoms such as a breast lump, rectal bleeding, persistent cough, swollen lymph nodes, or unexplained weight loss. However, many patients may choose not to seek medical care because they are asymptomatic. This can be caused by factors such as lack of health knowledge, high medical costs, cultural attitudes toward seeking care, fear of a cancer diagnosis, and difficulties dealing with the healthcare system. As a result, many patients tend to delay seeking medical attention until their symptoms have become more pronounced or greatly disruptive to daily life. By this stage, the disease has often progressed to an advanced stage, at which curative treatment options are substantially limited, and management typically shifts towards disease control and palliative care rather than curative approaches. When symptoms appear and patients seek medical attention, available clinical treatment may be inaccessible. Cancer detection at a late stage renders treatment difficult due to disease progression and metastasis.[2] Early detection of cancer would not only improve the quality of life for cancer patients but also significantly increase survival rates for many types of cancer. As the disease progresses, surgery becomes much less effective, radiotherapy is more frequently required, and chemotherapy drugs tend to be more toxic. Delays in diagnosis lead to poorer outcomes, while treatment costs, including medication, medical visits, and hospital care, rise sharply with each advancing stage.[4]

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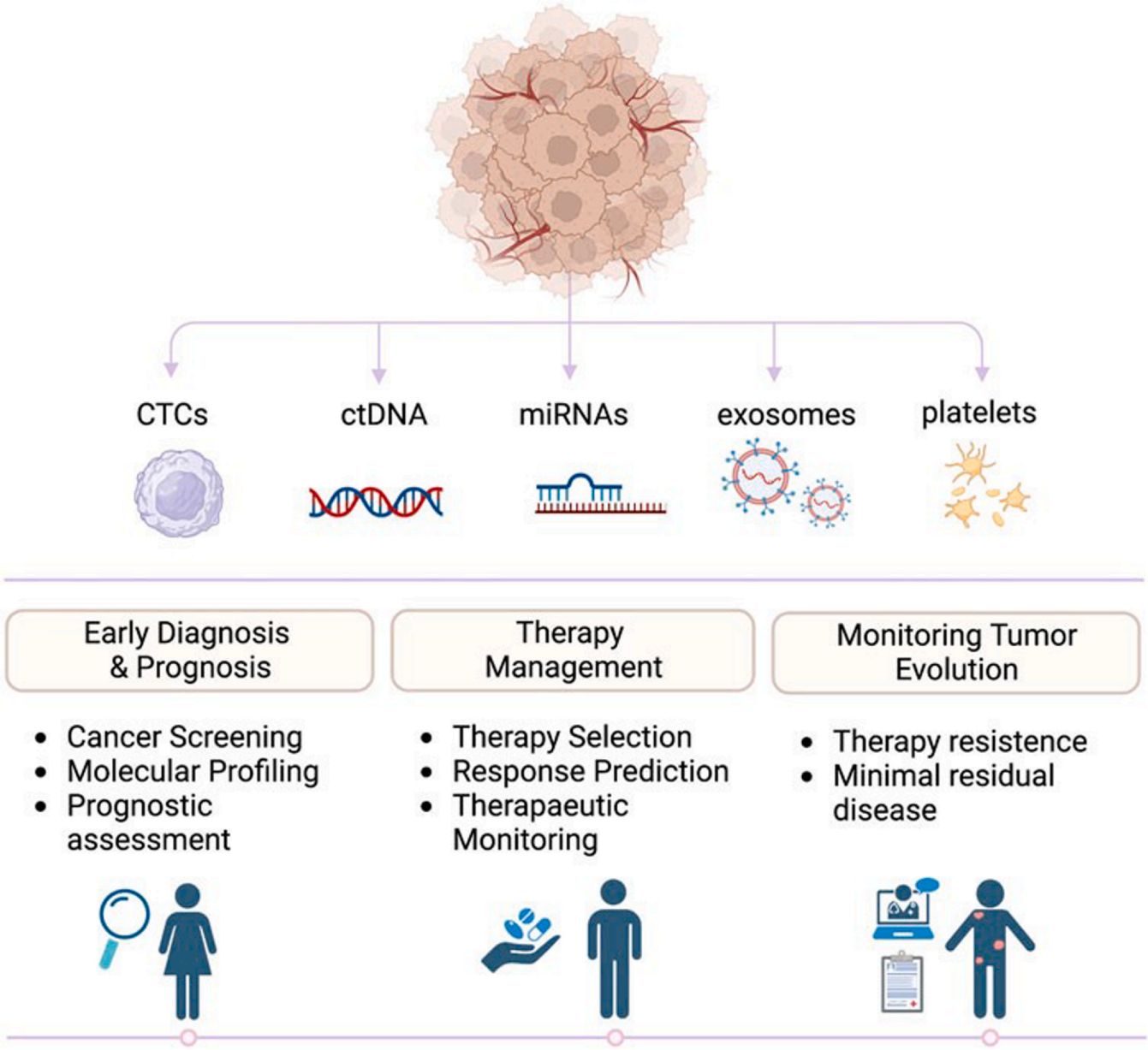


Figure 1. Liquid biopsy analytes and clinical applications. Tumor-derived CTCs, ctDNA, circulating miRNAs, exosomes and platelet-based biomarkers detected by liquid biopsy support early detection, prognosis, therapy management and monitoring of tumor evolution. Figure 1 was obtained with permission from Di Sario et al.[9] Abbreviations: 1. CTCs: circulating tumor cells 2. ctDNA: circulating tumor DNA.

Although early detection programs have grown in popularity, particularly in developed countries, they are typically based on symptomatic presentation. Early cancer symptoms, on the other hand, can be nonspecific and easily misdiagnosed as other conditions, contributing to disease progression and delayed diagnosis.[2,5] To effectively conduct large-scale cancer screening among healthy individuals in the future, a more comprehensive and cost-efficient approach is required. In recent years, an increasing number of scientists and companies have directed their attention toward liquid biopsy, recognizing its potential as a groundbreaking method for early detection and screening.[6,7] Blood contains a variety of biological materials, including circulating cells, platelets, extracellular vesicle (EVs), messenger RNA (mRNA), microRNA (miRNA), protein, and cell-free DNA (cfDNA).[8] These biomarkers can serve as a new type of cancer indicator, aiding in the identification of cancer patients within a group of healthy individuals (Figure 1).

1.2. Limitations of current screening methods

Current cancer screening and diagnostic methods face several substantial limitations that hinder their effectiveness and accessibility. The invasive nature of tissue biopsies, a primary tool for cancer detection, poses risks to patients and incurs considerable costs. Obtaining adequate tissue samples for genetic testing is particularly challenging for certain cancer types, often leading to insufficient material for accurate analysis. For instance, in advanced or metastatic non-small cell lung cancer (NSCLC), up to 31% of cases lack accessible tissue for biopsy. Even when tissue is available, preservation techniques, such as formalin fixation, can induce C→T transitions (cytosine → thymine) through cytosine deamination, potentially resulting in false-positive outcomes in genetic tests.[10,11]

Another major limitation of biopsies is their susceptibility to sampling bias due to tumor heterogeneity. Biopsies typically reflect the

genetic profile of only a small portion of the tumor at a specific point in time. However, tumors are dynamic and can evolve, changing their dominant mutation patterns or acquiring new mutations, especially in response to drug treatments.[12] This temporal limitation is particularly problematic when making treatment decisions, as relying on historical mutation profiles from past biopsies may not accurately represent the tumor's current state. For example, approximately 50% of NSCLC patients develop resistance to tyrosine kinase inhibitors due to the emergence of the epidermal growth factor receptor (EGFR) T790M mutation, yet this mutation is detectable in less than 5% of primary biopsies. Additionally, in NSCLC, up to one-third of tumors with EGFR-activating mutations develop resistance mutations such as EGFR-T790M within months after initiating EGFR-targeted therapy.[13,14]

Meanwhile, tissue biopsies, despite being the most commonly used method for cancer detection, staging, and prognosis, are not always feasible, particularly in metastatic diseases such as late-stage lung cancer, where obtaining tumor tissue is challenging. Furthermore, tissue biopsies are impractical for cancer screening and early diagnosis, especially when tumors have not yet formed. While there are some established screening methods, such as mammograms for breast cancer, Pap tests for cervical cancer, and regular screenings for colorectal and lung cancers, these methods also have limitations. They often suffer from limited sensitivity and specificity and are applicable only to specific types of cancer. Routine monitoring through biopsies necessitates that patients repeatedly endure painful procedures over the course of their treatment, significantly adding to the physical discomfort and emotional strain associated with cancer care.[15,16] This highlights the need for more effective, less invasive, and broadly applicable screening technologies that can address these gaps in cancer detection and diagnosis.

1.3. Advances in screening technologies and the emerging role of AI

Recent improvements in cancer screening have prompted the development of technologies including high-throughput sequencing, multi-omics profiling, and liquid biopsy–based assays. These technologies work together to generate large amounts of complicated biological data. These modern tools have the potential to detect tumors at fairly early stages, even before symptoms appear, but their performance is strongly dependent on the ability to interpret small molecular patterns that are frequently undetectable using traditional analytical methods. Artificial intelligence (AI) has emerged as an important tool for bridging this gap by allowing for sensitive, high-dimensional data analysis, recognizing weak or nonlinear biomarker signals, and integrating information across numerous biological layers. AI-driven models can promote liquid biopsy performance by increasing specificity, lowering false positives, and predicting cancer tissue-of-origin (TOO) more precisely. Combining advanced screening platforms with AI-powered analytics can improve early detection accuracy, scalability, and accessibility, paving the way for population-level screening and multi-cancer early detection systems.[17,18] Several studies have demonstrated that AI-powered models can analyze cfDNA methylation, RNA signatures, and circulating biomarker patterns to identify weak signals, predict TOO, and reduce false-positive rates more effectively than traditional pipelines. By combining advanced screening technologies with AI-driven analytics, early detection becomes more accurate, scalable, and clinically actionable, supporting the development of population-level multi-cancer early detection strategies.[19,20] Together, these advancements provide a promising pathway to overcome the limitations of current screening programs and establish a more proactive approach to cancer diagnosis. However, the clinical translation of these technologies remains challenged by issues such as false-positive results, cost-effectiveness considerations, and the need for standardized analytical and pre-analytical protocols.

Over the last ten years, tools including next-generation sequencing (NGS), detailed multi-omics studies, and liquid biopsy tests have transformed cancer screening by creating massive amounts of complex biological data. These approaches hold solid potential for detecting tumors at a fairly early stage before patients notice any signs, but the interpretation of the faint, multi-layered molecular clues they reveal remains a major hurdle for conventional analytical techniques. That is where AI comes in—it has become essential for advancing screening by improving detection rates and accuracy, reducing false positives, and providing tailored risk assessments for different cancer types.[21] In tandem with standard imaging approaches—such as mammography for breast cancer, low-dose computed tomography (LDCT) for lung cancer, colonoscopy for colorectal cancer, and Pap tests for cervical cancer—AI models are now widely used to evaluate radiographic data. These models uncover subtle abnormalities, predict interval cancers missed by routine protocols, and customize follow-up schedules based on individual risk, thereby reducing the workload for radiologists. In this context, AI serves as a unifying element, linking risk prediction, initial screening, and long-term monitoring into a cohesive, data-informed system.[22] This review, building on this momentum, demonstrates how AI can integrate liquid biopsy data across genomics, epigenomics, transcriptomics, proteomics, and metabolomics to achieve concrete benefits, including calculated cancer risk levels, predictions of tumor origin, and ongoing tracking systems designed for large-scale early detection efforts.[23]

Nevertheless, the clinical application of AI-driven multi-omic liquid biopsy approaches still encounters significant challenges that must be addressed before they can be routinely implemented in clinical practice. One of the primary concerns in early screening studies is the prevalence of false-positive results, necessitating a thorough cost–benefit analysis to assess the validity of these complex tests for large-scale use. Additionally, standardization of both analytical and pre-analytical procedures across different centers presents considerable difficulty, potentially introducing variability in test performance and undermining the reliability of the results.[24] Beyond these technical hurdles, several broader challenges to clinical implementation persist. These include issues related to data privacy and sharing restrictions, the complexity of regulatory requirements, the limited representativeness of many training cohorts, and concerns regarding algorithmic fairness across diverse demographic and clinical subgroups. Furthermore, the heterogeneity in AI study designs, difficulties with reproducibility and external validation, and obstacles to clinical integration and validation, further complicate the translation of these technologies into practice.[25] For successful and equitable adoption in real-world healthcare settings, it is imperative to ensure the reliability, scalability, and ongoing performance monitoring of these tools, while also considering their broader social impact.[26]

2. Fundamentals of liquid biopsy

2.1. Definition and principles

Liquid biopsy is an advanced technique with versatile applications across various stages of cancer screening and treatment. For asymptomatic individuals, it offers a powerful tool for early cancer detection, enabling improved diagnosis and timely intervention.[3] Recent oncology research has focused on cancer-derived components circulating in the bloodstream.[27] Cancer cells, through apoptosis or necrosis, release fragments of circulating cfDNA, known as circulating tumor DNA (ctDNA), as well as exosomes-membrane-encapsulated subcellular structures containing proteins and nucleic acids. Additionally, primary tumors and metastatic sites can shed viable cells into the bloodstream, known as circulating tumor cells (CTCs). The isolation of these tumor-derived components from peripheral blood and their subsequent genomic or proteomic analysis has led to the development of a new diagnostic approach termed 'liquid biopsy'.[28] Initially, the technique faced challenges due to the scarcity of nucleic acids and the difficulty in differentiating between normal and tumor-derived nucleic acids. However, advancements in NGS have significantly improved the

sensitivity of liquid biopsy, enabling the accurate detection of genetic and epigenetic abnormalities. The current liquid biopsy offers high specificity, providing robust and reproducible data in a simple, non-invasive manner from a single blood sample.[7,29] Although it is not yet a standard therapeutic medical test, the potential applications of liquid biopsy are rapidly expanding—from diagnostic genomic profiling to monitoring surgical outcomes, assessing response or resistance to systemic therapies, and quantifying minimal residual disease.

The clinical adoption of liquid biopsy has grown substantially since 2004, when Guardant Health introduced the first commercially available liquid biopsy test. By 2016, liquid biopsy, specifically for ctDNA analysis, had been officially recognized as a diagnostic tool and integrated into major reference laboratories. Today, several U.S. Food and Drug Administration (FDA)-approved liquid biopsy assays are available on the market, with some even being recognized by insurance companies as proficient for guiding treatment decisions. A notable example is the FDA's approval of the cobas® EGFR Mutation Test v2 in 2016, which is used to determine the eligibility of patients with NSCLC for specific EGFR tyrosine kinase inhibitors. This approval marked a significant milestone, highlighting the growing clinical importance of liquid biopsy in personalized cancer treatment.[30–32]

Liquid biopsy shows promise in diagnosing hard-to-reach cancers, such as brain tumors. Annually, 3.7 and 2.6 out of every 100,000 males and females, respectively, are diagnosed with primary malignant brain tumors.[33] CT and magnetic resonance imaging scans detect cancerous lesions, while needle biopsies target tumor tissue in sensitive or inaccessible areas.[34] Recently, non-blood-based liquid biopsies have emerged as effective tools for detecting and monitoring brain tumors.[35] In cases of inconclusive imaging, mutant DNA/RNA in cerebrospinal fluid (CSF) or blood can support a neoplasia diagnosis or detect residual disease post-surgery. Targeted sequencing of CSF or blood analytes (EV RNA/DNA, CTCs, cfDNA) can sometimes replace craniotomy for obtaining tissue biopsies to identify actionable genetic alterations. Moreover, the minimally invasive nature of biofluid sampling enables ongoing monitoring.[36]

2.2. Types of biomarkers in liquid biopsies

Peripheral blood and other biological fluids can contain cancer-causing agents such as CTCs, ctDNA, exosomes, cell-free RNA (cfRNA), and tumor-educated platelets (TEPs). These components may reflect characteristics of the current tumor stage; therefore, liquid samples—and particularly their components—can be viewed as ideal tools for providing dynamic assessments of tumor genomes.[37] Each biomarker originates from distinct biological processes such as tumor cell shedding, apoptosis or necrosis, EV secretion, and platelet–tumor interactions, making them valuable indicators of tumor activity and progression.

CTCs are intact cells that detach from the primary tumor and enter the bloodstream, providing insights into the tumor's characteristics, metastatic potential, and response to therapy. Interest in studying CTCs via liquid biopsies has grown substantially in cancer research and clinical practice. Various methods, such as immunomagnetic separation, microfluidic devices, and filtration, can isolate CTCs from blood samples. Once identified, molecular and cellular assays can characterize CTCs to reveal their phenotypic and genetic variations. The presence of CTCs in the bloodstream is linked to poor prognosis and an increased metastasis risk in breast, prostate, lung, and colorectal cancers.[38] Studies have shown that higher CTC counts correlate with advanced disease stages and worse treatment outcomes. Because CTCs retain the molecular features of the tumor and circulate throughout the body, they serve as a clinically useful biomarker for prognosis, monitoring of therapeutic response, and guidance of personalized treatment strategies.[39,40]

ctDNA consists of small fragments of tumor DNA released into the bloodstream by apoptotic or necrotic tumor cells.[41] Its short half-life allows ctDNA levels to reflect rapid changes in tumor burden, providing valuable insights into cancer treatment. It holds promise for early cancer detection by identifying tumor-specific changes even when tissue samples are not feasible. Beyond early detection, ctDNA analysis can monitor treatment response and detect minimal residual disease that conventional imaging might miss.[42] Various techniques, such as NGS (including targeted, whole-exome, and whole-genome sequencing) and droplet digital polymerase chain reaction, have been developed for detecting and analyzing ctDNA, with droplet digital polymerase chain reaction offering high sensitivity and specificity for rare mutant alleles.[43,44] Clinically, ctDNA enables real-time tumor genotyping, resistance mutation detection, and longitudinal disease monitoring.

Exosomes, EVs secreted by tumor cells, are present in body fluids including blood, urine, and saliva. Their analysis via liquid biopsies has streamlined cancer diagnosis, prognosis, and treatment monitoring. Exosomes carry proteins, lipids, nucleic acids, and metabolites that reflect their parent cells' molecular traits and influence tumor growth and metastasis.[45] Isolating and characterizing exosomes from liquid biopsies can reveal tumor biomarkers and insights into the tumor microenvironment. Research shows exosomes could serve as cancer detection biomarkers, with specific proteins, nucleic acids, and mutations found in patient samples, including exosomal miRNAs that help differentiate cancer patients from healthy individuals and predict prognosis.[46,47] Their stability and tumor specificity render exosomes clinically valuable for early detection and for tracking therapeutic response.

cfRNAs, also known as extracellular RNAs, are found in microvesicles, exosomes, lipoproteins, and ribonucleoprotein complexes. These release mechanisms protect cfRNA from rapid enzymatic degradation, allowing them to remain stable in body fluids. They function as signaling molecules, mediating communication between cells over various distances. These include miRNA, transfer RNA, long noncoding RNA (lncRNA), and other RNA types from different tissues.[48,49]

Blood-based biomarkers have been the focus of liquid biopsy research, as cfRNAs in plasma are relatively stable, often protected by subcellular particles or exosome packaging. Blood collection is standardized, enabling early detection of resistance and capturing tumor heterogeneity. Noncoding cfRNAs, such as lncRNA and miRNA, are particularly stable and promising as biomarkers in liquid biopsy.[50] Clinically, cfRNA profiles can indicate tumor subtype, treatment response, and dynamic transcriptional activity within the tumor.

Platelets, abundant cell fragments in the blood, perform various functions such as forming clots, interacting with immune cells, and monitoring pathogens. They can also reflect disease progression by altering their pre-mRNA splicing in response to cancer signals, leading to changes in their transcriptome.[51] During their life cycle, platelets acquire tumor-related biomolecules by interacting with cancer cells, ingesting and enriching circulating proteins, nucleic acids, and vesicles. Tumors may recruit immature platelets (megakaryocytes), and in advanced stages, cancer cells can invade bone marrow, releasing younger platelets into the bloodstream.[52] Studies have shown a platelet gene expression profile linked to metastatic lung cancer, with down-regulation of 197 out of 200 genes compared to controls.[53] Because TEPs accumulate tumor-derived signals, they serve as highly sensitive blood-based indicators for early cancer detection, tumor classification, and monitoring disease progression. However, liquid biopsy assays face several important limitations that affect reproducibility and clinical interpretability. Pre-analytical variables—including delayed processing, choice of anticoagulant, and sample storage time—can significantly alter cfDNA fragmentation patterns and cfRNA stability. CTC detection remains technically demanding due to their extremely low abundance, and results often vary substantially across enrichment platforms. These challenges highlight the need for standardized protocols to ensure reliable biomarker quantification.

Table 1 shows the comparison of major circulating biomarkers used in liquid biopsy. Overall, CTCs, ctDNA, exosomes/EVs, cfRNA, and TEPs arise from distinct tumor-related processes, including tumor cell

Table 1
Comparison of major circulating biomarkers used in liquid biopsy.

Biomarker Type	Source/Biological Origin	Molecular Content	Primary Detection Methods	Advantages	Current Limitations	Clinical Utility Examples
Circulating tumor cells (CTCs)[54]	Tumor cells shed into bloodstream during invasion/metastasis	Whole viable cells (DNA, RNA, protein)	CellSearch®, FACS, microfluidics, scRNA-seq	Provides phenotypic & functional information; drug testing possible	Fairly low abundance, technically challenging isolation	Prognosis and treatment monitoring in breast & prostate cancer
Circulating tumor DNA (ctDNA)[55]	Fragmented DNA from tumor cell apoptosis/ necrosis	Somatic mutations, CNVs, methylation	ddPCR, NGS, TEC-Seq, CAPP-Seq	Highly sensitive; MRD monitoring	Low levels in early cancer; false negatives	Relapse detection in CRC (Signatera®, Guardant360®)
Exosomes/EVs[56]	Actively secreted vesicles from live tumor cells	DNA, mRNA, lncRNA, miRNA, proteins, lipids	Ultracentrifugation, SEC, immunocapture, NTA	Stable cargo; reflects active biology	Lack of standardized isolation; heterogeneity	Prostate cancer diagnosis (ExoDx™)
Cell-free RNA (cfRNA)[57]	Passive leakage or active release into blood or fluids	mRNA, miRNA, lncRNA, circRNA	RNA-seq, PCR, EV-RNA assays	Real-time gene expression snapshot	Unstable; requires strict processing	PROGENSA PCA3 for prostate cancer (FDA-approved)
Tumor-educated platelets (TEPs)[58]	RNA-profile changes through tumor interaction	Spliced RNA, proteins	RNA-seq, ELISA, qPCR	High abundance, reflects tumor activity	Biological mechanism not fully defined	With high accuracy in Glioblastoma and CRC discrimination

Abbreviations: 1. EVs: extracellular vesicles 2. CNVs: copy number variations 3. mRNA: messenger RNA 4. lncRNA: long non-coding RNA 5. miRNA: microRNA 6. circRNA: circular RNA 7. FACS: fluorescence activated cell sorting 8. ddPCR: droplet digital polymerase chain reaction 9. NGS: next-generation sequencing 10. TEC-Seq: targeted error-correction sequencing 11. CAPP-Seq: cancer personalized profiling by deep sequencing 12. SEC: size-exclusion chromatography 13. NTA: nanoparticle tracking analysis 14. PCR: polumerase chain reaction 15. EV-RNA: extracellular vesicle RNA 16. ELISA: enzyme-linked immunosorbent assay 17. qPCR: quantitative polumerase chain reaction 18. MRD: minimal residual disease 19. CRC: colorectal cancer. 20. scRNA-seq: single cell RNA sequencing.

shedding, apoptosis, necrosis, vesicle secretion, and platelet–tumor interactions. Each of these circulating analytes provides valuable insight into various aspects of tumor biology, from cellular phenotype and metastatic potential to genomic alterations, epigenetic modifications, and dynamic transcriptional activity. By reflecting tumor burden, molecular evolution, treatment response, and disease progression, they collectively represent key liquid biopsy biomarkers that can be accessed through peripheral blood and other biofluids. In clinical practice, this diversity allows them to support cancer detection, risk stratification, prognosis assessment, and longitudinal monitoring. However, their reliable use requires careful attention to pre-analytical and technical factors, as variability in assay platforms and analytical pipelines can considerably influence biomarker measurements and clinical interpretability.

3. Multi-omic approaches in liquid biopsy

It is essential to note that multi-omic liquid biopsy methods rely on the combined analysis of genetics, epigenomics, transcriptomics, proteomics, and metabolomics to provide a comprehensive molecular picture of cancer. High-throughput methods, such as NGS, bisulfite sequencing, mass spectrometry, and RNA sequencing, capture distinct aspects of tumor biology. NGS detects somatic mutations, copy number variations, and structural variants in ctDNA for tumor genotyping. Bisulfite sequencing (including methylation arrays) identifies cancer-specific DNA methylation patterns in cfDNA, often preceding detectable genetic changes. Mass spectrometry enables proteomics through protein quantification and post-translational modification analysis, as well as metabolomics through small-molecule profiling. RNA sequencing characterizes cfRNA expression profiles, splice variants, and pathway activation. Multi-omic integration links genetic mutations to methylation changes, transcriptional activity, protein expression, and metabolic alterations, substantially enhancing early detection and tumor characterization beyond single-omic approaches (Figure 2).[9,59]

3.1. Genomics

Genomic analysis represents the foundation of liquid biopsy, primarily focusing on the detection and characterization of ctDNA and other cfDNA fragments in bodily fluids. Recent developments have expanded the utility of these techniques beyond detection to encompass comprehensive molecular characterization of tumors without invasive tissue sampling. ctDNA has emerged as a pivotal biomarker in precision oncology, providing genetic information that reflects tumor heterogeneity more comprehensively than traditional tissue biopsies. The detection of tumor-specific genetic alterations, including single nucleotide variants, copy number alterations, and structural rearrangements, offers valuable insights into cancer biology and evolution.[60] For patients with lung cancer, liquid biopsy enables the detection of drug-targetable mutations, such as EGFR variants, particularly beneficial for patients in remote locations or those unable to undergo tissue biopsy.[61]

Despite these advances, genomic liquid biopsy faces several challenges. The limited sensitivity for early-stage cancers remains a significant hurdle, with detection rates often below 50% for stage I cancers using current genomics-based approaches.[62] This limitation stems from the fairly low fraction of tumor-derived DNA in circulation during early disease stages. However, innovations in molecular techniques have significantly improved detection thresholds.

The temporal and spatial advantages of genomic liquid biopsy are noteworthy in particular. Unlike conventional tissue biopsies that capture a single moment in tumor evolution from a specific location, genomic liquid biopsy provides a dynamic snapshot of the tumor's genomic landscape, potentially reflecting contributions from multiple tumor sites.[60] This comprehensive view enables more accurate monitoring of tumor evolution during treatment, allowing for timely detection of resistance mechanisms and informing treatment modifications.

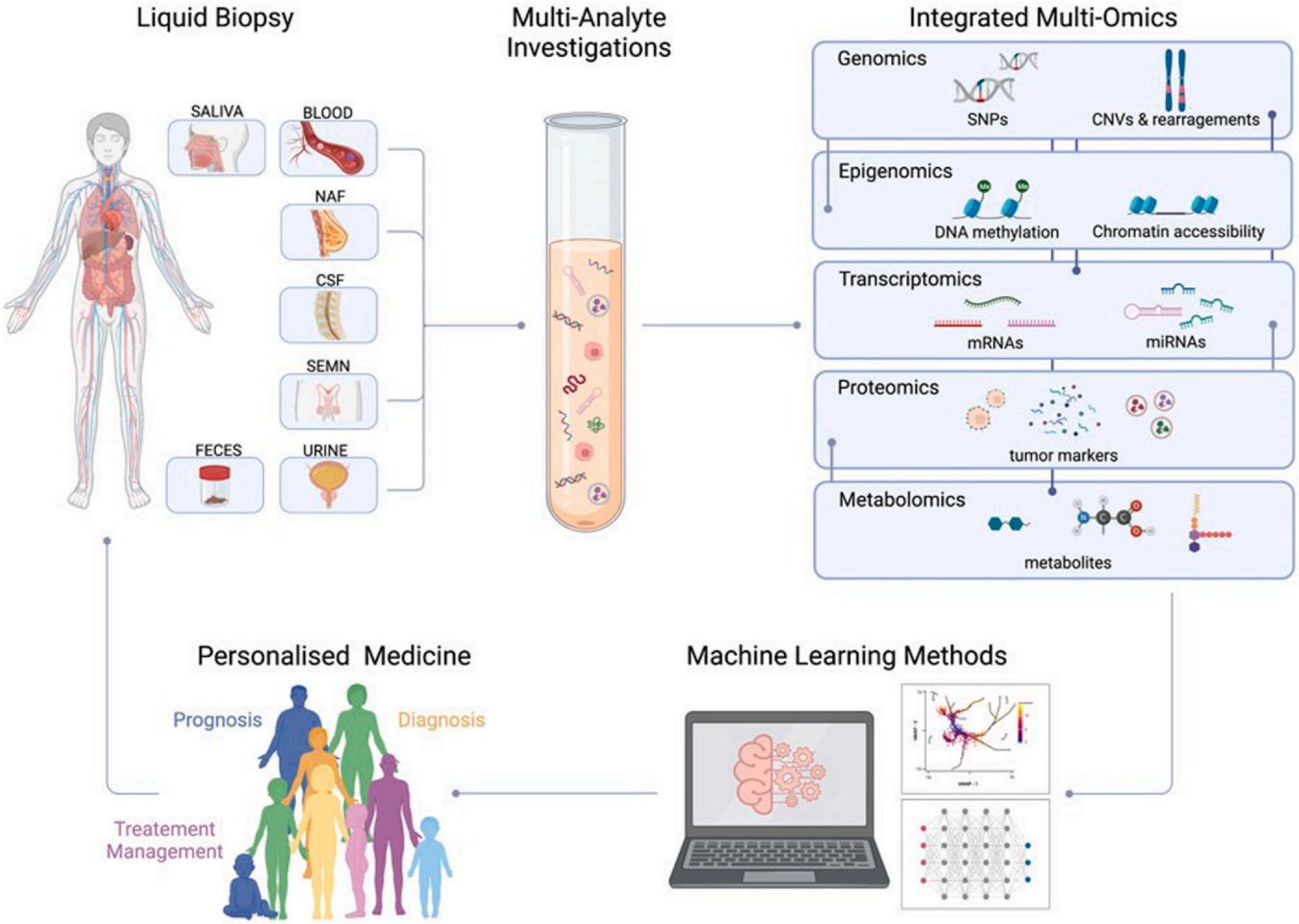


Figure 2. Schematic of liquid biopsy for clinical decision-making. Liquid biopsy collected from different body fluids can be profiled at multiple-omics levels and combined with computational analysis to inform diagnosis, prognosis and treatment decisions. Figure 2 was obtained with permission from Di Sario et al.[9] Abbreviations: 1. NAF: nipple aspirate fluid 2. CSF: cerebrospinal fluid 3. SEMN: seminal fluid 4. SNPs: single nucleotide polymorphisms 5. CNVs: copy number variations.

Future directions include the development of ultrasensitive detection methods, integration with other omic platforms, and implementation of AI for data interpretation. As these technologies continue to evolve, genomic liquid biopsy is expected to play an increasingly vital role in the paradigm of precision oncology.

3.2. Epigenomics

Epigenomic analysis in liquid biopsy has emerged as a powerful complement to genomic approaches, offering unique insights into cancer biology through the examination of epigenetic modifications in circulating nucleic acids. Epigenetic alterations, which affect gene expression without changing the underlying DNA sequence, provide an additional layer of information that can significantly enhance the sensitivity and specificity of cancer detection. DNA methylation, a key epigenetic modification, has gained significant attention in liquid biopsy applications. Recent studies have demonstrated that cancer-specific DNA methylation patterns can be detected in cfDNA extracted from bodily fluids, providing a promising avenue for early cancer diagnosis.[63] These methylation signatures often precede genetic mutations in cancer development, potentially allowing for earlier detection than genomic approaches alone.

The advantages of methylation-based liquid biopsy are particularly evident in multi-cancer early detection (MCED) efforts. A recent blood test developed to identify over 50 different cancer types based on methylation signatures in cfDNA has shown promising results.[62] However, sensitivity for early-stage cancers remains a challenge, with detection rates below 50% for stage I cancers, highlighting the need for continued refinement and potential integration with other omic approaches. Beyond DNA methylation, other epigenetic modifications such as histone modifications, chromatin accessibility, and nucleosome positioning in cfDNA have shown potential as cancer biomarkers. These features can provide information about the TOO for detected cancer signals, aiding in cancer localization following a positive screening result.

Technological advances have considerably enhanced the capability to detect and analyze epigenetic modifications in liquid biopsy samples. Methods such as bisulfite sequencing, methylation-specific PCR, and array-based approaches have improved the sensitivity and specificity of epigenetic biomarker detection, enabling more accurate cancer classification and monitoring.

Despite these promising developments, challenges remain in standardizing methodologies, bioinformatic analysis, and result interpretation across different laboratories. Additionally, distinguishing cancer-specific epigenetic alterations from age-related or environmental changes presents a significant challenge that requires sophisticated computational approaches and reference datasets.

3.3. Transcriptomics

Transcriptomic analysis in liquid biopsy examines RNA-based biomarkers circulating in bodily fluids, offering unique insights into gene expression patterns and cancer biology. Unlike DNA-based approaches

that primarily detect genetic alterations, transcriptomic liquid biopsy provides a dynamic view of cellular activity and biological processes, capturing the functional state of tumors in real-time. cfRNA, including mRNA, miRNA, lncRNA, and other RNA species, constitutes the primary focus of transcriptomic liquid biopsy. These RNA molecules are released into circulation through various mechanisms, including cellular apoptosis, necrosis, and active secretion via EVs such as exosomes.[9,59] The diverse landscape of circulating RNA offers a rich source of potential cancer biomarkers that reflect not only the presence of cancer but also its biological behavior and response to therapy.

Recent advances in liquid transcriptomics have demonstrated remarkable utility in cancer diagnosis and classification. A breakthrough reported in early 2025 combined cfRNA analysis with AI to achieve high diagnostic accuracy across multiple cancer types, including solid tumors (area under the curve [AUC] = 0.86), myeloid neoplasms (AUC = 0.86), and lymphoid neoplasms (AUC = 0.79).[64] This approach has the potential to reduce the need for traditional tissue biopsies, as noted by the International Society of Liquid Biopsy.

Technical innovations have significantly enhanced the capability to detect and analyze circulating RNA. High-throughput sequencing technologies, digital PCR, and microarray platforms have improved the sensitivity and specificity of RNA detection in liquid biopsy samples. Additionally, novel RNA stabilization methods have addressed the challenges associated with the inherent instability of RNA molecules, enabling more reliable analysis of transcriptomic biomarkers.

Beyond cancer detection, transcriptomic liquid biopsy offers useful information into tumor heterogeneity, metastatic potential, and treatment response. RNA expression patterns can reveal the activation of specific pathways associated with cancer progression and drug resistance, potentially guiding treatment decisions and monitoring. Furthermore, splice variants and fusion transcripts detectable in liquid biopsy samples can indicate specific therapeutic vulnerabilities, informing targeted treatment approaches.

3.4. Proteomics

Proteomic analysis in liquid biopsy examines circulating proteins and peptides in bodily fluids, providing direct clues into the functional state of tumors and their microenvironment. Unlike nucleic acid-based approaches that infer protein activity, proteomic liquid biopsy directly measures the end products of gene expression, capturing post-translational modifications and protein-protein interactions that play crucial roles in cancer biology. Recent advances in proteomics technologies have substantially enhanced the capability to detect and analyze low-abundance proteins in liquid biopsy samples. A groundbreaking study published in early 2024 developed a novel proteome-based multicancer screening test capable of detecting early-stage cancers, yielding high accuracy.[62] Using proximity extension assay technology, researchers measured over 3000 proteins in plasma samples from 440 individuals with 18 different early-stage solid tumors.

Beyond cancer detection, proteomic liquid biopsy offers valuable insights into tumor biology and treatment response. A recent study employed a liquid biopsy-based proteomic assay to identify CTCs in metastatic prostate cancer patients, demonstrating the ability to detect druggable protein targets on CTCs.[65] This approach has the potential to guide precision oncology by matching patients with targeted therapies based on the actual expression of protein drug targets, rather than relying solely on genetic alterations.

The clinical applications of proteomic liquid biopsy extend beyond solid tumors to various cancer types and even to non-malignant conditions. For instance, proteomic analysis of aqueous humor showed promise in predicting treatment response in ophthalmological conditions, demonstrating the broader utility of this approach.[66] In a pilot study, proteins identified through liquid biopsy of aqueous humor helped determine whether the predominant pathogenic event was primarily angiogenic or inflammatory in nature, potentially guiding more rational and cost-effective treatment selection.

Interestingly, research has shown that many of the most useful protein biomarkers for early cancer detection are those present in low concentrations in the plasma proteome,[62] highlighting the importance of sensitive detection methods and the potential of proteomics to detect cancer at early stages when genetic alterations may be below the detection threshold of current technologies.

3.5. Metabolomics

Metabolomic analysis in liquid biopsy examines small molecule metabolites in bodily fluids, providing a functional readout of cellular processes and offering unique insights into cancer metabolism. As the end products of cellular processes, metabolites reflect the integration of genetic, epigenetic, and environmental influences, potentially capturing early metabolic alterations that precede genetic and protein changes in cancer development. The metabolic reprogramming observed in cancer cells has long been recognized as a hallmark of malignancy. Recent advances in metabolomic liquid biopsy leverage these cancer-specific metabolic signatures for diagnostic and prognostic applications. A comprehensive review by Wang et al.[67] highlighted the utility of metabolic markers from various biological fluids in clinical cancer studies, emphasizing their potential for cancer screening, diagnosis, differentiation of cancer typing, grading, staging, and prediction of therapeutic response and prognosis.

The diversity of sample types represents a key advantage of metabolomic liquid biopsy. While blood remains the most studied biofluid, recent research has expanded to include urine, sputum, CSF, and pleural effusion, each offering unique metabolic insights.[68] This diversity allows for optimized biomarker discovery based on the pathophysiological characteristics of different cancer types, potentially enhancing sensitivity and specificity through targeted approaches.

Technical innovations have significantly enhanced the capability to detect and analyze metabolites in liquid biopsy samples. Nuclear magnetic resonance spectroscopy and mass spectrometry coupled with various separation techniques have improved the sensitivity, specificity, and throughput of metabolite detection. These advances have enabled the comprehensive profiling of the cancer metabolome, revealing cancer-specific alterations in glycolysis, the tricarboxylic acid cycle, lipid metabolism, amino acid metabolism, and nucleotide synthesis. Beyond cancer detection, metabolomic liquid biopsy offers valuable insights into tumor biology, treatment response, and prognosis. Several studies have identified metabolic markers for predicting therapy efficacy in various cancer types. For example, specific metabolites, such as isocitric acid and citric acid, have shown potential in predicting responses to neoadjuvant chemoradiotherapy. Similarly, a five-metabolite panel containing lactic acid, 2-hydroxyglutaric acid, and succinic acid demonstrated high sensitivity (88.9%) and specificity (88.0%) in predicting recurrence of renal cell carcinoma after surgery.[67]

Despite these promising developments, challenges remain in the standardization of sample collection, processing, and analysis protocols. Additionally, distinguishing cancer-specific metabolic signatures from normal physiological variations, dietary influences, and comorbidities presents a significant challenge, requiring sophisticated bioinformatic approaches and reference datasets.

3.6. Advantages of multi-omic integration

The integration of multiple omic approaches in liquid biopsy represents a paradigm shift in cancer diagnostics, offering synergistic benefits that surpass the limitations of single-modality analyses. This comprehensive strategy harnesses the complementary strengths of genomics, epigenomics, transcriptomics, proteomics, and metabolomics

Table 2
Performance comparison of multi-omic integration pipelines across major clinical trials.

Pipeline	Omics Combination	AI Method	Cohort Size	Sensitivity	Specificity	Cancer Types
CancerSEEK[69]	Genomics + Proteomics	Logistic Regression	1,005	70%	99%	8 solid tumors
MCED methylation-based[70]	Epigenomics + Fragmentomics	Transformer neural network	4,077	41.40%	98.50%	Pan-cancer
Fragmentome-based cfDNA[71]	Fragmentomics + Epigenomics	CNN	796	94%	0.90 AUC	7 cancers

Abbreviations: 1. MCED: multi-cancer early detection 2. cfDNA: cell-free DNA 3. CNN: convolutional neural network 4. SVM: support vector machine 5. AUC: area under the curve.

to provide a multidimensional view of cancer biology, significantly enhancing diagnostic accuracy, prognostic assessment (Table 2), and therapeutic guidance. One of the primary advantages of multi-omic integration is the substantial improvement in sensitivity and specificity for early cancer detection. Each omic layer provides unique insights into cancer biology, capturing different aspects of the disease process. For instance, while genomic approaches excel at identifying specific mutations, they may miss cancers with low mutational burden. Epigenomic analyses can detect cancer-specific methylation patterns that often precede genetic alterations, while proteomic and metabolomic approaches directly measure the functional consequences of these changes. A recent study demonstrated that integrating different tumor-derived omics data from body fluids for cancer detection and monitoring significantly outperformed single-modality analyses alone,[59] highlighting the synergistic potential of multi-omic integration.

Multi-omic integration also addresses the challenge of tumor heterogeneity more effectively than single-omic approaches. Cancer represents a heterogeneous collection of diseases with diverse genetic, epigenetic, transcriptomic, proteomic, and metabolomic profiles. By simultaneously examining multiple omic layers, integrated approaches can capture this complexity, providing a more comprehensive characterization of the tumor.[9] This comprehensive view is especially valuable for monitoring treatment response and resistance, as different omic signatures may reveal distinct resistance mechanisms and guide subsequent therapeutic strategies.

The complementary nature of different omic platforms enhances the biological interpretability of liquid biopsy results. For example, integrating genomic alterations with proteomic data can verify whether mutations actually result in functional protein changes, distinguishing driver from passenger mutations. Similarly, combining transcriptomic and metabolomic data can elucidate the functional consequences of gene expression changes, providing insights into activated pathways and potential therapeutic vulnerabilities.

Technological advances have significantly facilitated the integration of multi-omic data in liquid biopsy. High-throughput sequencing, mass spectrometry, and advanced computational methods enable simultaneous profiling of multiple omic layers from limited sample volumes. Furthermore, AI, ML algorithms in particular, have enhanced the interpretation of complex multi-omic data, improving the accuracy of cancer detection, classification, and prognostication. Looking ahead, the future of liquid biopsy lies in the seamless integration of multiple omic layers, potentially complemented by imaging technologies and clinical data.[72] This holistic approach promises to transform cancer diagnostics and management, enhancing early detection, enabling more precise prognostication, and guiding personalized therapeutic strategies, ultimately improving patient outcomes in the era of precision oncology.

Despite the advantages of multi-omic integration, cross-platform variability (Table 3) introduces substantial challenges. Differences in sequencing depth, proteomic quantification methods, and metabolite extraction workflows can cause inconsistent biomarker signatures across studies. Additionally, tumor heterogeneity may result in discordant signals between genomic, epigenomic, and proteomic layers, complicating downstream AI-based integration.

4. AI in liquid biopsy analysis

4.1. ML algorithms for biomarker detection

ML has emerged as a critical component in processing and analyzing the complex data generated from liquid biopsies, enabling the discovery and validation of cancer biomarkers with unprecedented sensitivity and specificity (Table 4). Traditional analysis methods are increasingly inadequate for capturing the high-resolution complex relationships between biomarkers and cancer subtypes, necessitating advanced computational approaches to extract meaningful information from liquid biopsy data.[18]

A wide range of ML algorithms have been successfully implemented for biomarker detection in liquid biopsies. These include support vector machines (SVMs), decision trees, random forests, and neural networks, each offering unique advantages for specific applications in cancer detection. Supervised learning approaches are particularly valuable when biomarkers are collected to characterize subject groups with known labels (such as cancer vs. non-cancer), while unsupervised learning techniques help investigate data structure or discover groups of similar examples within datasets without prior labeling. The power of these algorithms lies in their ability to evaluate the effects of multiple biomarkers simultaneously and discover higher-order interactions that might be missed by conventional statistical methods. When applied to liquid biopsy data, ML algorithms have consistently outperformed the sensitivity and specificity of individual markers across numerous cancer applications.[74] For instance, in the comprehensive evaluation of various ML approaches for cancer detection from liquid biopsies, researchers

Table 3
Technical challenges matrix comparing single-omic and multi-omic liquid biopsy technologies.

Technology	Analytical Sensitivity	Pre-analytical Variability	Sample Volume (mL)	Processing Time (hrs)	Clinical Validation
Single-omic ctDNA[73]	< 0.1% VAF	ddPCR/NGS	10	4–6	Phase II trials
Single-omic proteomics[70]	ng/mL range	Proximity extension	5	2–3	Phase I trials
Multi-omic CancerSEEK[69]	Composite score	Logistic regression	4	6–8	Phase III
Multi-omic methylation-based MCED[70]	Methylation + Fragmentation	Transformer neural network	4	24	Phase III
Multi-omic fragmentome-based cfDNA[71]	Fragmentome CNN	Deep learning	10	12	Prospective

Abbreviations: 1. ctDNA: circulating tumor DNA 2. MCED: multi-cancer early detection 3. cfDNA: cell-free DNA 4. VAF: variant allele frequency 5. CNN: convolutional neural network 6. ddPCR: droplet digital polymerase chain reaction 7. NGS: next-generation sequencing.

Table 4
Benchmark of artificial intelligence algorithms for liquid biopsy analysis and clinical validation metrics.

AI Algorithm	Input Modality	Validation	AUC	TOO Accuracy	Sample Size	Clinical Stage
Transformers[64]	cfRNA	5-fold CV	0.86	85%	2,500	Prospective
CNN + LSTM[71]	cfDNA fragmentation	Prospective	0.94	91%	385	Phase II
SVM Ensemble[59]	Multi-omic	External validation	0.85	88%	1,500	Retrospective
Random Forest[73]	ctDNA VAF + CNV	Nested CV	0.88	82%	1,082	Prospective
Deep Learning[58]	TEP RNA	Prospective	0.93	84%	1,028	Phase II

Abbreviations: 1. AUC: area under the curve 2. TOO: tissue-of-origin 3. CNN: convolutional neural network 4. LSTM: long short-term memory 5. SVM: support vector machine 6. cfRNA: cell-free RNA 7. cfDNA: cell-free DNA 8. ctDNA: circulating tumor DNA 9. VAF: variant allele frequency 10. CNV: copy number variation 11. TEP: tumor-educated platelet 12. CV: cross validation.

found that these algorithms could identify subtle patterns in biomarker data that correlate with disease states, even when individual markers showed minimal differences between groups. A noteworthy advancement in ML-based biomarker detection is the development of "fragmentomics" approaches, which analyze the fragmentation patterns of cfDNA circulating in the bloodstream. Cancer cells and normal cells break down differently, resulting in distinct cfDNA fragmentation signatures that can be identified through ML algorithms.[75] This approach allows for the detection of cancer using markedly smaller blood samples than traditional diagnostic tools, potentially enabling more widespread screening programs.

The application of ML to liquid biopsy extends beyond genomic data to other biomarker types. In CTC detection, ML-powered tomographic phase imaging flow cytometry systems can provide high-throughput 3D phase-contrast tomograms of individual cells. Using hierarchical ML decision-makers working on features calculated from the 3D tomograms of cells' refractive index, researchers have achieved identification of tumor cells with success rates higher than 97%, and with accuracy over 97% in discriminating between different cancer cell types.[76]

Recent developments in this field have concentrated on enhancing the sensitivity and specificity of biomarker detection through advanced feature extraction and selection methods. By carefully selecting the most informative features from liquid biopsy data, ML algorithms can reduce noise and improve classification performance.[74] Additionally, techniques such as ensemble learning, which combines multiple ML models, have shown promise in raising the robustness of biomarker detection, particularly in heterogeneous cancer types.

For proper implementation of ML in biomarker detection, rigorous validation procedures are essential. This includes cross-validation techniques to ensure that the algorithms generalize well to new data and are not overfitting to training samples.[18] Furthermore, external validation using independent datasets is crucial for establishing the clinical utility of ML-based biomarker detection methods.

4.2. AI-driven data integration and interpretation

The integration and interpretation of multi-omic liquid biopsy data represent one of the most significant challenges and opportunities in precision oncology. AI has emerged as a critical tool for synthesizing the vast and heterogeneous datasets derived from different biomarker classes, enabling a holistic understanding of cancer biology that surpasses the limitations of single-modality analyses. AI systems facilitate multimodal analysis by integrating genomic, epigenomic, transcriptomic, proteomic, and metabolomic features into unified predictive frameworks, thereby promoting both diagnostic accuracy and biological interpretability.[77]

A wide array of ML and deep learning (DL) architectures have been applied to multi-omic liquid biopsy integration. Traditional algorithms such as SVMs, random forests, and elastic-net regression remain valuable for high-dimensional but relatively small datasets due to their robustness, embedded feature-selection mechanisms, and interpretability. In contrast, DL models—including convolutional neural networks (CNNs), autoencoders, graph neural networks, and multimodal transformers—are better suited for capturing nonlinear relationships and hierarchical patterns across omic layers. CNNs are often used for structured ctDNA fragmentation signatures and methylation patterns, while graph neural networks model molecular interactions between features across omic layers. Multimodal transformers have gained traction because they can learn cross-modal attention weights, highlighting how information from one omic type influences another.

Handling feature dimensionality is another critical methodological step. High-dimensional multi-omic datasets often contain tens of thousands of features, requiring algorithms such as least absolute shrinkage and selection operator, mutual-information ranking, recursive feature elimination, or unsupervised dimensionality reduction (e.g., principal component analysis, Uniform Manifold Approximation and Projection) to prevent overfitting. DL frameworks can automatically extract salient features, but regularization strategies (dropout, early stopping, weight decay) remain necessary for generalizability.

AI-driven multi-omic integration is further strengthened through robust strategies for dealing with data missingness and imbalance, both common in liquid biopsy studies. Missing values arise from incomplete sequencing coverage or failed biomarker assays; imputation strategies such as k-nearest neighbors, matrix factorization, denoising autoencoders, or generative adversarial networks (generative adversarial network -based imputation) are increasingly used to reconstruct absent data. Class imbalance—particularly the scarcity of early-stage cancer cases relative to controls—can result in biased models. Techniques such as synthetic minority oversampling technique, class-weighted loss functions, and focal loss have proven effective in improving sensitivity for minority classes.

Multimodal fusion approaches continue to illustrate the value of AI-enhanced interpretation. A recent study combining EV long RNA with CT imaging achieved AUC values of 91.9% for adenocarcinoma classification and 94.8% for benign–malignant discrimination, outperforming both single-modality models and human radiologists.[78] These findings highlight the capacity of AI to integrate heterogeneous biomarker sources into clinically meaningful predictions.

Despite their power, AI-driven data-integration frameworks face challenges, including missing data, platform heterogeneity, batch effects, and computational burden. Advanced approaches such as transfer learning, domain adaptation, and batch-effect correction using adversarial networks are being applied to harmonize data across studies and enhance model generalizability. Such strategies aim to ensure that multi-omic liquid biopsy models remain robust across diverse populations, sequencing platforms, and clinical settings.[79]

4.3. Predictive modeling for early cancer detection

Predictive modeling using AI has revolutionized the potential for early cancer detection through liquid biopsies, addressing one of the most critical challenges in oncology: identifying cancer at its most treatable stages. By leveraging complex algorithms to analyze the multidimensional data obtained from liquid biopsies, AI-powered predictive models can identify patterns associated with cancer development before clinical symptoms manifest. The impact of early detection on patient outcomes cannot be overstated. As demonstrated in a groundbreaking study from Mayo Clinic, an AI model successfully

identified pancreatic cancer in CT scans approximately 475 days before clinical diagnosis, which yielded an accuracy of 92%.[80] This remarkable lead time highlights the transformative potential of AI-enhanced detection methods, especially for aggressive cancers with typically late diagnosis. Similar approaches applied to liquid biopsy data show comparable promise for detecting molecular signatures of cancer, significantly earlier than conventional diagnostic methods.

AI-based predictive models for liquid biopsies typically follow a structured development process. First, they are trained on large datasets of biomarker profiles from both cancer patients and healthy individuals. These models learn to recognize subtle patterns in circulating biomarkers that distinguish early-stage cancer from normal physiological variations. Once trained, the models can be applied to new patient samples to predict cancer risk or presence with increased accuracy.

MCED represents a considerable advancement in this field, with AI playing a crucial role in analyzing the tens of thousands of analytical targets typically included in MCED tools. The interpretation of complicated patterns composed of multiple components would be exceedingly difficult without AI approaches, making it a "must-use" technology in analyzing MCED data.[77] These models can simultaneously screen multiple cancer types from a single blood sample, potentially transforming population-level cancer screening programs.

The versatility of AI-powered predictive modeling extends across various cancer types and biomarker classes. For instance, BioMark Diagnostics developed a liquid biopsy platform that leverages metabolomics and ML to predict estrogen receptor status in breast cancer patients, with an accuracy of 93%. This non-invasive test demonstrates how AI can extract clinically relevant information beyond simple cancer detection, which may guide treatment decisions from the earliest stages of disease. Similarly, researchers have developed a blood test that uses AI to detect cancer-related genetic changes and protein biomarkers for early detection of ovarian cancer, a disease typically diagnosed at advanced stages.[81] By combining AI-driven detection of cancer-associated patterns of DNA fragments with protein biomarkers (CA-125 and HE4), this approach improved screening accuracy and helped distinguish cancerous tumors from benign growths.

A particularly promising direction is the integration of liquid biopsy data with other clinical information. AI models can combine molecular biomarkers with patient demographics, medical history, and imaging data to create comprehensive risk profiles with greater predictive power than any single data source. This holistic approach accounts for the multifactorial nature of cancer development and enables more personalized screening recommendations.

Despite these advances, challenges remain in translating AI-powered predictive models into clinical practice. Issues of model interpretability, validation across diverse populations, regulatory approval, and integration into existing clinical workflows must be addressed. Additionally, distinguishing true cancer signals from biological noise requires sophisticated algorithms and rigorous validation to minimize false positives that could lead to unnecessary interventions.[82]

A major limitation of AI-based models is their dependence on high-quality, diverse training datasets. Most existing models are derived from small or demographically narrow cohorts, which increases the risk of overfitting and biases in prediction. Errors in early cancer detection—whether false positives leading to unnecessary testing or false negatives resulting in delayed diagnosis—pose substantial clinical risks. Moreover, the opaque nature of many DL architectures remains a barrier to clinical adoption.

5. Next-generation early cancer detection technologies

5.1. Multi-analyte blood tests

Multi-analyte blood tests represent an emerging approach to cancer screening, offering the potential to detect multiple cancer types simultaneously through a minimally invasive blood draw. These assays analyze diverse circulating biomarkers and may expand early-detection options for cancers that currently lack effective screening tools. Among these platforms, CancerSEEK—developed at the Johns Hopkins Kimmel Cancer Center—illustrates the promise of multimodal biomarker analysis. The test integrates two complementary components: cfDNA mutation detection using a 61-amplicon panel targeting 16 cancer-associated genes, and measurement of eight protein biomarkers (CA-125, CEA, CA 19–9, prolactin, hepatocyte growth factor, osteopontin, myeloperoxidase, and TIMP-1). This dual strategy aims to address a key limitation of relying solely on ctDNA, as many early-stage tumors shed insufficient DNA to be detected reliably. In an initial case-control study of 1005 patients with nonmetastatic, clinically detected cancers—spanning eight cancer types, including ovary, liver, stomach, pancreas, and esophagus—CancerSEEK demonstrated encouraging results. The assay detected a median of 70% of these cancers, achieving sensitivities of 69%–98% for five types with no widely recommended population screening methods. Specificity exceeded 99%, with only 7 of 812 healthy controls testing positive. The test also localized cancers to a limited set of anatomical sites in a median of 83% of individuals.[83] These results highlight early feasibility but derive from controlled studies rather than real-world screening cohorts.

The DETECT-A study provided the first prospective interventional evaluation of CancerSEEK in an average-risk population.[19] When paired with confirmatory PET-CT imaging, the test identified several cancers not captured by standard screening. Follow-up at 4.4 years showed that 50% of CancerSEEK-detected cancers were alive and cancer-free, and early-stage (I–II) cases showed favorable outcomes. While promising, this study was not designed to assess population-level mortality reduction, and CancerSEEK remains in the investigational stage pending larger trials.

Other multi-analyte circulating assays have also emerged. For example, GRAIL's Galleri test analyzes cfDNA methylation patterns to detect over 50 cancer types and infer TOO. Exact Sciences' multi-analyte MCED classifier combines DNA methylation with protein biomarkers. In the ASCEND-2 study, the test achieved 99.0% specificity but exhibited stage-dependent sensitivity, ranging from 23.6% for early-stage (I–II) disease to 84.6% for stage IV cancers.[84]

Despite these advances, sensitivity for early-stage tumors remains a major limitation of current MCED technologies. A multi-analyte test reported at ESMO 2024 detected only 15.4% of stage I cancers, indicating that most early tumors would be missed.[85] Likewise, CancerSEEK demonstrated 43% sensitivity for stage I cancers compared to substantially higher detection rates at later stages.[83] These findings emphasize that while multi-analyte assays are a promising avenue, they are not yet validated replacements for established screening programs and require further optimization, expanded prospective trials, and integration with complementary modalities such as AI-based risk modeling and imaging.

5.2. AI-enhanced imaging and liquid biopsy combinations

The integration of AI with both imaging technologies and liquid biopsy represents a powerful synergistic approach to early cancer detection. By combining the spatial resolution of imaging with the molecular information from liquid biopsies, AI-enhanced multimodal strategies overcome the limitations of either method alone, significantly improving diagnostic accuracy and patient outcomes. A groundbreaking development in this field comes from researchers at Johns Hopkins Kimmel Cancer Center, who developed an AI-powered liquid biopsy that analyzes DNA fragmentation patterns in blood samples to identify individuals with lung cancer. In a prospective study involving approximately 1000 participants who met criteria for traditional LDCT screening, this blood test demonstrated remarkable capability in identifying those most likely to have lung cancer, potentially transforming the screening paradigm.[86] The researchers noted, "We have a simple blood test that could be done in a doctor's office that would tell patients

whether they have potential signs of lung cancer and should get a follow-up CT scan".[86]

This approach addresses a critical gap in current lung cancer screening programs. Despite evidence that yearly LDCT screening in high-risk patients can detect lung cancers early and reduce mortality, only 6%–10% of eligible individuals undergo screening. The AI-enhanced blood test could function as an initial screening tool, identifying those who would benefit most from follow-up imaging, thereby improving screening adherence and efficiency.[87]

Similar multimodal approaches have shown promise in other cancer types. Research exploring multimodal fusion for the differential diagnosis of early-stage lung adenocarcinoma (tumor sizes <2 cm) combined liquid biopsy biomarkers, specifically EV long RNA, with CT attributes.

For ovarian cancer, a difficult-to-detect malignancy typically diagnosed at advanced stages, researchers have developed a blood test combining AI-powered analysis of DNA fragmentation patterns with protein biomarkers (CA-125 and HE4).[88] The integration of these approaches improved screening accuracy and helped distinguish cancerous tumors from benign growth. As the senior author noted, "The combination of AI, cfDNA fragmentomes and a pair of protein biomarkers in a simple blood test improved detection of ovarian cancer even in patients with early-stage disease,"[88] offering potential for widespread, accessible screening for this lethal cancer.

The advantages of these combined approaches extend beyond enhanced sensitivity and specificity. In neuro-oncology, where tissue biopsies carry significant risks, the integration of advanced imaging techniques, liquid biopsies, and AI algorithms provides a comprehensive diagnostic approach. AI-driven algorithms, CNNs in particular, integrate data from multiple modalities to improve diagnostic accuracy, reduce interobserver variability, and accelerate clinical decision-making for central nervous system tumors.[89]

The technical foundation for these multimodal approaches lies in the complementary nature of the data they provide. Imaging techniques offer spatial information and physical characteristics of suspicious lesions, while liquid biopsies provide molecular profiles indicating the presence of cancer. AI algorithms serve as the bridge between these data types, identifying complex patterns and correlations that would be impossible for human observers to detect. This synergy enables earlier detection, more accurate characterization, and better treatment planning, potentially transforming cancer care paradigms across multiple malignancies.

5.3. Novel biomarker discovery through AI

AI is revolutionizing biomarker discovery for early cancer detection, enabling researchers to identify complex patterns in multimodal data that would remain hidden using conventional analytical approaches. By applying sophisticated algorithms to vast datasets, AI can uncover novel biomarkers and biomarker signatures that enhance the sensitivity and specificity of liquid biopsy-based cancer screening. The fundamental advantage of AI in biomarker discovery lies in its ability to analyze and integrate diverse data types simultaneously. Traditional approaches to biomarker identification often rely on hypothesis-driven investigations of single molecules or pathways. In contrast, AI-based models can uncover innovative and surprising connections within high-dimensional datasets, identifying patterns that common statistical methods would easily miss. As noted in recent research, DL models demonstrate expertise in decoding complex information from tumor biopsies, blood tests, and medical images to identify biomarkers associated with patient outcomes such as survival rates and medication responses.[90]

This capability has led to significant advancements in detecting ctDNA, a key component of liquid biopsies. Researchers have developed an AI-powered tool called MRD-EDGE that shows unprecedented sensitivity in predicting cancer recurrence.[91] In one notable case, this method detected colorectal cancer recurrence in five patients without any false negatives. The researchers achieved what they described as "remarkable signal-to-noise enhancement,"[91] enabling them to detect cancer recurrence months or even years before standard clinical methods. This approach relies on whole-genome sequencing of DNA in blood samples, gathering much more "signal" than targeted approaches focusing on small sets of cancer-associated mutations, resulting in enhanced sensitive detection of tumor DNA.

The application of AI extends beyond genomic biomarkers to other molecular entities. BioMark Diagnostics has developed a liquid biopsy platform that leverages metabolomics and ML to identify metabolic biomarkers. In a study of pulmonary neuroendocrine tumors, their approach revealed that these cancers reprogram their metabolism, producing distinctive metabolic signatures.[92] A logistic regression model constructed with just eight metabolites discriminated lung neuroendocrine tumors from controls with impressive performance (AUC $= 0.91$).

The integration of proteomics with AI has also yielded promising results. Researchers combined liquid biopsy proteomics with single-cell transcriptomics and AI to trace the cellular origin of 5953 proteins detected in ocular fluid.[66] This approach identified hundreds of cell-specific protein markers and revealed unexpected findings, such as retinal degeneration in Parkinson's disease. The researchers developed AI models to assess individual cellular aging a that many eye diseases undergo accelerated molecular aging of specific cell types. While focused on ocular disease, this methodology demonstrates how AI can identify novel protein biomarkers from liquid biopsies, with potential applications to cancer detection.

A promising direction is the integration of multiple omic layers through AI. Current projects are exploring how AI tools can integrate multidimensional profiles, genomic, transcriptomic, miRNA analysis, and proteomic data, to develop multi-omic signatures for predicting cancer outcomes and treatment responses. For triple-negative breast cancer, researchers are measuring ctDNA, mRNA, miRNA, and proteins in both tumor and plasma through liquid biopsies, with AI algorithms integrating these diverse data types to develop novel prognostic signatures.[93]

The emergence of "fragmentomics," which analyzes the fragmentation patterns of cfDNA in the bloodstream, represents another AI-powered approach to biomarker discovery. Cancer cells and normal cells break down differently, resulting in distinct cfDNA fragmentation signatures that can be identified through ML algorithms. This approach allows for cancer detection using significantly smaller blood samples than traditional diagnostic tools, potentially enabling more widespread screening programs.[94]

Despite these advances, challenges remain in biomarker discovery using AI, including the need for large, high-quality datasets, model interpretability, and ensuring diversity to avoid exacerbating health disparities. Addressing these issues will be crucial to realizing the full potential of AI-discovered biomarkers in revolutionizing early cancer detection and improving patient outcomes.

Although next-generation technologies show promise, their translation into routine screening is hindered by cost and logistical barriers. Multi-analyte assays require high sequencing depth, specialized consumables, and advanced computational pipelines, resulting in high per-test costs. Integration with AI further adds expenses related to data storage, curation, and cybersecurity. Without scalable and cost-effective workflow optimization, widespread population-level implementation remains challenging.

6. Challenges and future directions

Despite rapid progress in liquid biopsy and AI, multi-omic liquid biopsy assays integrated with computational models are still confined to exploratory research rather than routine oncology practice. Several practical and methodological barriers slow this translation. One major issue is the lack of standardization across the analytical pipeline, from

pre-analytical handling of blood and other body fluids to library preparation, sequencing depth, and downstream bioinformatic workflows. Small differences in processing, storage, or batch correction can markedly change cfDNA, cfRNA, and protein readouts, making it difficult to compare results across platforms and studies. Harmonized operating procedures, reference materials, and external quality assessment schemes are therefore essential to ensure reproducible measurements and reliable cross-study benchmarking.[95]

On the computational side, most current AI models are trained on fragmented, heterogeneous, and relatively small datasets. Missing values, noisy labels, and pronounced class imbalance are common, especially in early-stage disease and rare tumor subtypes. Many multi-omic classifiers have been developed in single-center or demographically narrow cohorts, which increases the risk of overfitting and limits model performance in underrepresented populations. Approaches such as federated learning, privacy-preserving data sharing, source-free domain adaptation, and stronger calibration/interpretability tools may partly address these issues, but they also introduce new technical and logistical demands. In parallel, robust data security standards are required to protect sensitive genomic and multi-omic profiles, as multi-institutional consortia and large biobanks become the main source of training data.[96]

Clinical integration represents another critical challenge. Existing MCED tests based on cfDNA methylation or fragmentomics still show modest sensitivity for stage I tumors and usually require confirmatory imaging, additional invasive procedures, and careful work-up of incidental findings. For multi-omic liquid biopsy panels, evidence is even more limited. Large, prospective trials powered for hard clinical endpoints such as cancer-specific mortality and treatment modification are needed to demonstrate true clinical utility rather than diagnostic accuracy alone. Regulatory and reimbursement pathways must also keep pace with rapidly evolving assays and adaptive AI models, taking into account analytical validity, clinical benefit, cost-effectiveness, and equity of access. Without coherent frameworks, even technically strong tests may remain confined to academic centers or high-income settings.[97]

Ethical and societal considerations further shape the future of multi-omic liquid biopsy and AI. Highly sensitive screening may increase detection of indolent or slowly progressive lesions, raising the risk of overdiagnosis, overtreatment, and anxiety in asymptomatic individuals. Both false positives and false negatives can have considerable psychological, financial, and clinical consequences, especially when complex AI outputs are poorly understood by patients and clinicians. Clear communication strategies, shared decision-making tools, and transparent reporting of test limitations are therefore needed to support responsible use of these technologies and to maintain public trust.[98]

Looking ahead, the most promising opportunities may lie in multimodal integration rather than single-platform solutions. Combining liquid biopsy-derived cfDNA, cfRNA, and protein signatures with radiomics, pathology, electronic health record data, and wearable-sensor information could promote robust risk stratification, early relapse detection, and dynamic treatment adaptation. However, these ambitions will only be realized if technical standardization, data governance, algorithmic fairness, rigorous clinical validation, and health-system readiness advance in parallel. Addressing these challenges in a coordinated way will determine whether multi-omic liquid biopsy and AI remain powerful research tools or evolve into practical components of everyday cancer screening, diagnosis, and longitudinal care.[99]

7. Conclusions

The integration of multi-omic liquid biopsies and AI represents a paradigm shift in cancer management with implications across the cancer care continuum. This convergence can transform how cancer is detected, monitored, and treated, improving outcomes through earlier intervention and more precise strategies. Liquid biopsy technologies offer a noninvasive alternative to tissue biopsies, enabling continuous monitoring of tumor dynamics and real-time detection of resistance or recurrence. Such ongoing assessment addresses the evolving nature of cancer and provides clinicians with a comprehensive view of tumor genetics across fluidic environments, crucial for evaluating treatment, identifying resistance, and making timely adjustments. This real-time monitoring has profound implications for clinical practice, potentially reducing repeated invasive procedures while guiding decisions more effectively. Economically, the global liquid biopsy market is projected to reach $7.7 billion by 2031, reflecting its clinical utility and adoption across oncology.

Perhaps most significantly, liquid biopsies may reduce unnecessary treatments while maintaining outcomes. The evolution of these tools toward personalized early detection is being driven by multi-omic approaches and AI, which together enable precise cancer signature identification at the earliest stages. Companies are already developing intelligent screening platforms and AI-enhanced assays that illustrate how multimodal biomarker analysis can refine early detection and treatment guidance, moving the field toward accessible and individualized cancer screening.

However, several real-world challenges must be addressed to translate these advances into routine clinical practice. Multi-omic liquid biopsy assays face complex regulatory pathways, as they require rigorous analytical validity, clinical validity, and clinical utility evidence to meet FDA and Clinical Laboratory Improvement Amendments certification standards. The incorporation of AI introduces additional scrutiny regarding algorithmic transparency, reproducibility, and mitigation of demographic and technical biases. Ethical considerations also play a central role: highly sensitive assays risk overdiagnosis of indolent lesions, and false-positive results can lead to patient anxiety, unnecessary downstream procedures, and increased healthcare burden. Conversely, false negatives may provide false reassurance, underscoring the importance of responsible risk communication and equitable deployment of AI-based tools.

Finally, economic feasibility remains a significant determinant of widespread adoption. Multi-omic sequencing, high-throughput data analysis, and long-term computational infrastructure are substantially more expensive than many traditional screening methods. Reimbursement frameworks and insurance coverage remain limited for early detection applications, and robust cost-effectiveness analyses comparing liquid biopsy–based screening to current population-level programs are urgently needed. Addressing these regulatory, ethical, and economic barriers will be key for achieving the ultimate vision: noninvasive, accurate, and personalized cancer detection implemented at scale and capable of transforming cancer outcomes worldwide.

CRediT authorship contribution statement

Esmaeil Mahmoudi: Writing – original draft, Conceptualization. **Mona Ebrahimi:** Data curation. **Elham Bahramian:** Writing – review & editing, Supervision.

Ethical statement

The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are Authors' contributions appropriately investigated and resolved.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

1. Connal S, Cameron JM, Sala A, et al. Liquid biopsies: the future of cancer early detection. *J Transl Med.* 2023;21(1):118.
2. Tobore TO. On the need for the development of a cancer early detection, diagnostic, prognosis, and treatment response system. *Future Sci OA.* 2020;6(2):FSO439.
3. Chen M, Zhao H. Next-generation sequencing in liquid biopsy: cancer screening and early detection. *Hum Genom.* 2019;13(1):34.
4. Iragorri N, de Oliveira C, Fitzgerald N, Essue B. The out-of-pocket cost burden of cancer care—a systematic literature review. *Curr Oncol.* 2021;28(2):1216–1248.
5. Late-stage cancer detection in the USA is costing lives. *Lancet.* 2010;376(9756):1873.
6. Smit DJ, Pantel K. Circulating tumor cells as liquid biopsy markers in cancer patients. *Mol Asp Med.* 2024;96:101258.
7. Crowley E, Di Nicolantonio F, Loupakis F, Bardelli A. Liquid biopsy: monitoring cancer-genetics in the blood. *Nat Rev Clin Oncol.* 2013;10(8):472–484.
8. Labib M, Mohamadi RM, Poudineh M, et al. Single-cell mRNA cytometry via sequence-specific nanoparticle clustering and trapping. *Nat Chem.* 2018;10(5):489–495.
9. Di Sario G, Rossella V, Famulari ES, et al. Enhancing clinical potential of liquid biopsy through a multi-omic approach: a systematic review. *Front Genet.* 2023;14:1152470.
10. Kim ES, Hirsh V, Mok T, et al. Gefitinib versus docetaxel in previously treated non-small-cell lung cancer (INTEREST): a randomised phase III trial. *Lancet.* 2008;372(9652):1809–1818.
11. Quach N, Goodman MF, Shibata D. In vitro mutation artifacts after formalin fixation and error prone translesion synthesis during PCR. *BMC Clin Pathol.* 2004;4(1):1.
12. Bedard PL, Hansen AR, Ratain MJ, Siu LL. Tumour heterogeneity in the clinic. *Nature.* 2013;501(7467):355–364.
13. Gazdar. Af. Activating and resistance mutations of EGFR in non-small-cell lung cancer: role in clinical response to EGFR tyrosine kinase inhibitors. *Oncogene.* 2009;28(1):S24–S31.
14. Inukai M, Toyooka S, Ito S, et al. Presence of epidermal growth factor receptor gene T790M mutation as a minor clone in non–small cell lung cancer. *Cancer Res.* 2006;66(16):7854–7858.
15. Yazdi ZF, Roshannezhad S, Sharif S, Abbaszadegan MR. Recent progress in prompt molecular detection of liquid biopsy using Cas enzymes: innovative approaches for cancer diagnosis and analysis. *J Transl Med.* 2024;22(1):1173.
16. Vaidyanathan R, Soon RH, Zhang P, Jiang K, Lim CT. Cancer diagnosis: from tumor to liquid biopsy and beyond. *Lab Chip.* 2019;19(1):11–34.
17. Foser S, Maiese K, Digumarthy SR, Puig-Butille JA, Rebhan C. Looking to the future of early detection in cancer: liquid biopsies, imaging, and artificial intelligence. *Clin Chem.* 2024;70(1):27–32.
18. Liu L, Chen X, Petinrin OO, et al. Machine learning protocols in early cancer detection based on liquid biopsy: a survey. *Life.* 2021;11(7):638.
19. Buchanan AH, Lennon AM, Choudhry OA, et al. Multiyear clinical outcomes of cancers diagnosed following detection by a blood-based multicancer early detection test. *Cancer Prev Res (Phila).* 2024;17(8):349–353.
20. Brito-Rocha T, Constâncio V, Henrique R, Jerónimo C. Shifting the cancer screening paradigm: the rising potential of blood-based multi-cancer early detection tests. *Cells.* 2023;12(6):935.
21. Gentile F, Malara N. Artificial intelligence for cancer screening and surveillance. *ESMO Real World Data Digit Oncol.* 2024;5:100046.
22. Cherukuri SP, Kaur A, Goyal B, et al. Artificial intelligence-enhanced liquid biopsy and radiomics in early-stage lung cancer detection: a precision oncology paradigm. *Cancers (Basel).* 2025;17(19):3165.
23. Di Sario G, Rossella V, Famulari ES, et al. Enhancing clinical potential of liquid biopsy through a multi-omic approach: a systematic review. *Front Genet.* 2023;14:1152470.
24. Cheng CH, Shi SS. Artificial intelligence in cancer: applications, challenges, and future perspectives. *Mol Cancer.* 2025;24(1):274.
25. Zhu E, Muneer A, Zhang J, et al. Progress and challenges of artificial intelligence in lung cancer clinical translation. *NPJ Precis Oncol.* 2025;9(1):210.
26. Rockall AG, Chiu SM, Aboagye EO, et al. Artificial intelligence in women's cancers: innovation and challenges in clinical translation. *Lancet Digit Health.* 2025;7(10):100940.
27. Siravegna G, Marsoni S, Siena S, Bardelli A. Integrating liquid biopsies into the management of cancer. *Nat Rev Clin Oncol.* 2017;14(9):531–548.
28. Palmirotta R, Lovero D, Cafforio P, et al. Liquid biopsy of cancer: a multimodal diagnostic tool in clinical oncology. *Ther Adv Med Oncol.* 2018;10:1758835918794630.
29. Zhang W, Xia W, Lv Z, Ni C, Xin Y, Yang L. Liquid biopsy for cancer: circulating tumor cells, circulating free DNA or exosomes? *Cell Physiol Biochem.* 2017;41(2):755–768.
30. cobas EGFR Mutation Test v2. U.S. Food and Drug Administration. June 02, 2016. Accessed August 18, 2025. https://www.fda.gov/drugs/resources-information-approved-drugs/cobas-egfr-mutation-test-v2.
31. Brown P. The cobas® EGFR mutation test v2 assay. *Future Oncol.* 2016;12(4):451–452.
32. Greig SL. Osimertinib: first global approval. *Drugs.* 2016;76:263–273.
33. Bondy ML, Scheurer ME, Malmer B, et al. Brain tumor epidemiology: consensus from the brain tumor epidemiology consortium. *Cancer.* 2008;113(S7):1953–1968.
34. Batool SM, Yekula A, Khanna P, et al. The liquid biopsy consortium: challenges and opportunities for early cancer detection and monitoring. *Cell Rep Med.* 2023;4(10).
35. De Mattos-Arruda L, Mayor R, Ng CKY, et al. Cerebrospinal fluid-derived circulating tumour DNA better represents the genomic alterations of brain tumours than plasma. *Nat Commun.* 2015;6(1):8839.
36. Miller AM, Shah RH, Pentsova EI, et al. Tracking tumour evolution in glioma through liquid biopsies of cerebrospinal fluid. *Nature.* 2019;565(7741):654–658.
37. Michela B. Liquid biopsy: a family of possible diagnostic tools. *Diagnostics.* 2021;11(8):1391.
38. Pantel K, Alix-Panabières C. Circulating tumour cells in cancer patients: challenges and perspectives. *Trends Mol Med.* 2010;16(9):398–406.
39. Gorges TM, Pantel K. Circulating tumor cells as therapy-related biomarkers in cancer patients. *Cancer Immunol Immunother.* 2013;62(5):931–939.
40. Cristofanilli M, Hayes DF, Budd GT, et al. Circulating tumor cells: a novel prognostic factor for newly diagnosed metastatic breast cancer. *J Clin Oncol.* 2005;23(7):1420–1430.
41. Abbosh C, Birkbak NJ, Wilson GA, et al. Phylogenetic ctDNA analysis depicts early-stage lung cancer evolution. *Nature.* 2017;545(7655):446–451.
42. Diehl F, Schmidt K, Choti MA, et al. Circulating mutant DNA to assess tumor dynamics. *Nat Med.* 2008;14(9):985–990.
43. Wan JCM, Massie C, Garcia-Corbacho J, et al. Liquid biopsies come of age: towards implementation of circulating tumour DNA. *Nat Rev Cancer.* 2017;17(4):223–238.
44. Hindson BJ, Ness KD, Masquelier DA, et al. High-throughput droplet digital PCR system for absolute quantitation of DNA copy number. *Anal Chem.* 2011;83(22):8604–8610.
45. Kalluri R. The biology and function of exosomes in cancer. *J Clin Invest.* 2016;126(4):1208–1215.
46. Thakur BK, Zhang H, Becker A, et al. Double-stranded DNA in exosomes: a novel biomarker in cancer detection. *Cell Res.* 2014;24(6):766–769.
47. Madhavan B, Yue S, Galli U, et al. Combined evaluation of a panel of protein and miRNA serum-exosome biomarkers for pancreatic cancer diagnosis increases sensitivity and specificity. *Int J Cancer.* 2015;136(11):2616–2627.
48. Allen RM, Zhao S, Ramirez Solano MA, et al. Bioinformatic analysis of endogenous and exogenous small RNAs on lipoproteins. *J Extra Vesicles.* 2018;7(1):1506198.
49. Ratajczak J, Miekus K, Kucia M, et al. Embryonic stem cell-derived microvesicles reprogram hematopoietic progenitors: evidence for horizontal transfer of mRNA and protein delivery. *Leukemia.* 2006;20(5):847–856.
50. Xing S, Zhu Y, You Y, et al. Cell-free RNA for the liquid biopsy of gastrointestinal cancer. *Wiley Inter Rev RNA.* 2023;14(5):e1791.
51. Sol N, Wurdinger T. Platelet RNA signatures for the detection of cancer. *Cancer Metastas Rev.* 2017;36:263–272.
52. Dovizio M, Ballerini P, Fullone R, Tacconelli S, Contursi A, Patrignani P. Multifaceted functions of platelets in cancer: from tumorigenesis to liquid biopsy tool and drug delivery system. *Int J Mol Sci.* 2020;21(24):9585.
53. Calverley DC, Phang TL, Choudhury QG, et al. Significant downregulation of platelet gene expression in metastatic lung cancer. *Clin Transl Sci.* 2010;3(5):227–232.
54. Wang X, Wang L, Lin H, et al. Research progress of CTC, ctDNA, and EVs in cancer liquid biopsy. *Front Oncol.* 2024;14:1303335.
55. Parums DV. A review of circulating tumor DNA (ctDNA) and the liquid biopsy in cancer diagnosis, screening, and monitoring treatment response. *Med Sci Monit.* 2025;31.
56. Yu D, Li Y, Wang M, et al. Exosomes as a new frontier of cancer liquid biopsy. *Mol Cancer.* 2022;21(1):56.
57. Jelski W, Okrasinska S, Rutkowska W, Mroczko B. Circulating molecular biomarkers for the diagnosis and monitoring of NSCLC-a review. *Int J Mol Sci.* 2025;26(21):10278.
58. Antunes-Ferreira M, Koppers-Lalic D, Würdinger T. Circulating platelets as liquid biopsy sources for cancer detection. *Mol Oncol.* 2021;15(6):1727–1743.
59. Chen G, Zhang J, Fu Q, Taly V, Tan F. Integrative analysis of multi-omics data for liquid biopsy. *Br J Cancer.* 2022;128(4):505.
60. Bao Y, Zhang D, Guo H, Ma W. Beyond blood: advancing the frontiers of liquid biopsy in oncology and personalized medicine. *Cancer Sci.* 2024;115(4):1060.
61. Pérez-Callejo D, Romero A, Provencio M, Torrente M. Liquid biopsy based biomarkers in non-small cell lung cancer for diagnosis and treatment monitoring. *Transl Lung Cancer Res.* 2016;5(5):455.
62. Budnik B, Amirkhani H, Forouzanfar MH, Afshin A. Novel proteomics-based plasma test for early detection of multiple cancers in the general population. *BMJ Oncol.* 2024;3(1):73.
63. Loomans-Kropp HA. The utility of liquid biopsy-based methylation biomarkers for colorectal cancer detection. *Front Oncol.* 2024;14:1351514.
64. Albitar M, Charifa A, Agersborg S, Pecora A, Ip A, Goy A. Expanding the clinical utility of liquid biopsy by using liquid transcriptome and artificial intelligence. *J Liq Biopsy.* 2024;6:100270.
65. Horrmann A, Arafa A, Kamalanathan K, et al. A liquid biopsy-based proteomic assay to identify circulating tumor cell (CTC)–derived druggable targets in cancer patients. *J Clin Oncol.* 2025;43(5_suppl):258.
66. Wolf J, Franco JA, Yip R, et al. Liquid biopsy proteomics in ophthalmology. *J Proteome Res.* 2024;23(2):511–522.
67. Wang W, Zhen S, Ping Y, Wang L, Zhang Y. Metabolomic biomarkers in liquid biopsy: accurate cancer diagnosis and prognosis monitoring. *Front Oncol.* 2024;14:1331215.
68. Bao Y, Zhang D, Guo H, Ma W. Beyond blood: advancing the frontiers of liquid biopsy in oncology and personalized medicine. *Cancer Sci.* 2024;115(4):1060.
69. Connal S, Cameron JM, Sala A, et al. Liquid biopsies: the future of cancer early detection. *J Transl Med.* 2023;21(1):118.
70. Klein EA, Richards D, Cohn A, et al. Clinical validation of a targeted methylation-based multi-cancer early detection test using an independent validation set. *Ann Oncol.* 2021;32(9):1167–1177.
71. Mathios D, Johansen JS, Cristiano S, et al. Detection and characterization of lung cancer using cell-free DNA fragmentomes. *Nat Commun.* 2021;12(1):5060.
72. Foser S, Maiese K, Digumarthy SR, Puig-Butille JA, Rebhan C. Looking to the future of early detection in cancer: liquid biopsies, imaging, and artificial intelligence. *Clin Chem.* 2024;70(1):27–32.
73. Phallen J, Sausen M, Adleff V, et al. Direct detection of early-stage cancers using circulating tumor DNA. *Sci Transl Med.* 2017;9(403).

74. Ko J, Baldassano SN, Loh PL, Kording K, Litt B, Issadore D. Machine learning to detect signatures of disease in liquid biopsies - A user's guide. *Lab Chip.* 2018;18(3):395.
75. Douville C, Lahouel K, Kuo A, et al. Machine learning to detect the SINEs of cancer. *Sci Transl Med.* 2024;16(731).
76. Pirone D, Montella A, Sirico DG, et al. Label-free liquid biopsy through the identification of tumor cells by machine learning-powered tomographic phase imaging flow cytometry. *Sci Rep.* 2023;13(1):6042.
77. Wang HY, Lin WY, Zhou C, Yang ZA, Kalpana S, Lebowitz MS. Integrating artificial intelligence for advancing multiple-cancer early detection via serum biomarkers: a narrative review. *Cancers.* 2024;16(5):862.
78. Zhang Y, Sun B, Yu Y, et al. Multimodal fusion of liquid biopsy and CT enhances differential diagnosis of early-stage lung adenocarcinoma. *NPJ Precis Oncol.* 2024;8(1):50.
79. Acosta JN, Falcone GJ, Rajpurkar P, Topol EJ. Multimodal biomedical AI. *Nat Med.* 2022;28(9):1773–1784.
80. Korfiatis P, Suman G, Patnam NG, et al. Automated artificial intelligence model trained on a large data set can detect pancreas cancer on diagnostic computed tomography scans as well as visually occult preinvasive cancer on prediagnostic computed tomography scans. *Gastroenterology.* 2023;165(6):1533–1546.e4.
81. Medina JE, Annapragada AV, Lof P, et al. Early detection of ovarian cancer using cell-free DNA fragmentomes and protein biomarkers. *Cancer Discov.* 2025;15(1):105–118.
82. Ünal AA, Yazarkan Y, Sönmez G, et al. Revolutionizing lung cancer care: the multifaceted approach of artificial intelligence, liquid biopsies, and circulating tumor DNA in screening, diagnosis, and prognosis. *Turkish Med Stud J.* 2024;11(2):32–39.
83. Cohen JD, Li L, Wang Y, et al. Detection and localization of surgically resectable cancers with a multi-analyte blood test. *Science.* 2018;359(6378):926–930.
84. Gainullin V, Hwang HJ, Hogstrom L, et al. Presented at European society of medical oncology congress. *Ann Oncol.* 2022;33(1):12–49.
85. Thompson CL, Baskin ML, Arem H, Gurusamy U. The promise and challenges of multi-cancer early detection assays for reducing cancer disparities. *Front Oncol.* 2024;14:1305843.
86. Mazzone PJ, Bach PB, Carey J, et al. Clinical validation of a cell-free DNA fragmentome assay for augmentation of lung cancer early detection. *Cancer Discov.* 2024;14(11):2224–2242.
87. Gandhi Z, Gurram P, Amgai B, et al. Artificial intelligence and lung cancer: impact on improving patient outcomes. *Cancers.* 2023;15(21):5236.
88. Li G, Zhang Y, Li K, et al. Transformer-based AI technology improves early ovarian cancer diagnosis using cfDNA methylation markers. *Cell Rep Med.* 2024;5(8):101666.
89. Rafanan J, Ghani N, Kazemeini S, Nadeem-Tariq A, Shih R, Vida TA. Modernizing Neuro-Oncology: The Impact of Imaging, Liquid Biopsies, and AI on Diagnosis and Treatment. *Int J Mol Sci.* 2025;26(3):917.
90. Alum EU. AI-driven biomarker discovery: enhancing precision in cancer diagnosis and prognosis. *Discov Oncol.* 2025;16(1):313.
91. Caputo V, Ciardiello F, Corte CM, Della Martini G, Troiani T, Napolitano S. Diagnostic value of liquid biopsy in the era of precision medicine: 10 years of clinical evidence in cancer. *Explor Target Antitumor Ther.* 2023;4(1):102.
92. Sen T, Dotsu Y, Corbett V, et al. Pulmonary neuroendocrine neoplasms: the molecular landscape, therapeutic challenges, and diagnosis and management strategies. *Lancet Oncol.* 2025;26(1):e13–e33.
93. Coffey K, Aukland B, Amir T, Sevilimedu V, Saphier NB, Mango VL. Artificial intelligence decision support for triple-negative breast cancers on ultrasound. *J Breast Imaging.* 2024;6(1):33–44.
94. Eledkawy A, Hamza T, El-Metwally S. Precision cancer classification using liquid biopsy and advanced machine learning techniques. *Sci Rep.* 2024;14(1):5841.
95. Crisafulli G. Liquid biopsy and challenge of assay heterogeneity for minimal residual disease assessment in colon cancer treatment. *Genes (Basel).* 2025;16(1):71.
96. Hsu CY, Askar S, Alshkarchy SS, et al. AI-driven multi-omics integration in precision oncology: bridging the data deluge to clinical decisions. *Clin Exp Med.* 2025;26(1):29.
97. Schroll MM, Quinn E, Pritchard D, et al. Perspectives on clinical adoption barriers to blood-based multi-cancer early detection tests across stakeholders. *J Pers Med.* 2024;14(6):593.
98. Sempere LF. Ethical considerations and implications of multi-cancer early detection screening: reliability, access and cost to test and treat. *Camb Q Health Ethics.* 2025;34(3):489–498.
99. Imai M, Nakamura Y, Yoshino T. Transforming cancer screening: the potential of multi-cancer early detection (MCED) technologies. *Int J Clin Oncol.* 2025;30(2):180–193.