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Liquid profiling for patients with advanced cancer is ready for clinical integration

Samantha O. Hasenleithner · Ellen Heitzer

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Summary Molecular profiling of circulating tumor DNA (ctDNA) to guide treatment decisions has found its way into routine management of patients with advanced cancer. This represents a pivotal advancement in precision oncology, offering a non-invasive and fast-tracked method to detecting clinically relevant biomarkers. With the backing of international oncology guidelines, ctDNA analysis is now a standard approach to consider in molecular diagnostics. Despite the promise of ctDNA in refining treatment strategies through the detection of genomic alterations and treatment-relevant biomarkers with high concordance to tissue biopsies, challenges persist. These include the interpretation of discordances due to tumor heterogeneity, sampling biases, and technical limitations, alongside the differentiation of tumor-derived mutations from clonal hematopoiesis. The current consensus supports the utility of comprehensive genomic profiling (CGP) panels for a broad spectrum of actionable targets, while acknowledging the limitations and advocating for a balanced application of "tissue-first" and "plasma-first" approaches tailored to individual patient scenarios. The essential role of molecular tumor boards (MTBs) is in navigating the complexities of ctDNA data interpretation, thereby ensuring the effective incorporation of liquid biopsy into personalized cancer treatment regimens.

S. O. Hasenleithner · Prof. Mag. Dr. E. Heitzer (✉)
Institute of Human Genetics, Diagnostic & Research Center for Molecular BioMedicine, Medical University of Graz, Neue Stiftingtalstraße 6, 8010 Graz, Austria
ellen.heitzer@medunigraz.at

Prof. Mag. Dr. E. Heitzer
Christian Doppler Laboratory for Liquid Biopsies for Early Detection of Cancer, Medical University of Graz, Graz, Austria

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Introduction

The integration of liquid biopsy into clinical practice marks a significant advancement in the realm of precision oncology, signaling a shift towards more refined and patient-centric approaches in routine cancer care. Clinicians are increasingly performing molecular profiling on circulating tumor DNA (ctDNA) obtained from plasma to non-invasively monitor residual disease [1], guide treatment options [2–4], or to match patients to suitable clinical trials [5], efforts which are underscored by the issuance of comprehensive implementation guidelines from international oncology and molecular pathology consortia such as ASCO/CAP [6, 7], ESMO [8], NCCN [9, 10], and IASLC. Certain indications now even exist that support the "plasma first" use case, meaning that traditional tissue biopsies may be bypassed in favor of a minimally invasive testing strategy. One example would be the testing for *ESR1* mutations in plasma at endocrine resistance in breast cancer to guide addition of selective estrogen receptor degraders. While ctDNA-based assays are redefining clinical pathways, it can be difficult to sift through the growing literature base and clinical trial evidence [11], impeding the adoption of liquid biopsy in everyday cancer management. In this short review, we focus on the advanced cancer setting and offer brief, high-level summaries of the current guidelines, molecular profiling strategies and everyday challenges for incorporating liquid biopsy into real-world precision oncology approaches.

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Concordance between alterations in tumor tissue and ctDNA

Many studies have assessed the concordance of genomic alterations detected in major driver genes and treatment-relevant biomarkers between plasma and tissue and have found consistent sensitivities, ranging approximately between 70–90% across various solid tumors [5, 12–14]. Discordance is mostly related to tumor heterogeneity and temporal dynamics, sampling biases and technical limitations. Tumors are often heterogeneous and the portion of the tumor sampled for tissue analysis may not fully represent the entire genetic landscape of the tumor, whereas ctDNA represents a mixture of DNA shed from various tumor regions. Moreover, tumor genomes evolve over time due to therapeutic pressure and clonal selection. The ctDNA shed into the bloodstream reflects the most recent state of the tumor, whereas tissue samples may have been obtained at an earlier stage of the disease and may not capture these changes. On the other hand, mutations derived from the hematopoietic system, which can lead to clonal expansions, can be picked up in cfDNA. Clonal hematopoiesis-related mutations can introduce specificity concerns in ctDNA analysis. Without careful validation and discrimination strategies, there is a risk of misinterpreting clonal hematopoiesis-related mutations as tumor-derived mutations, leading to false-positive results.

Taken together, tissue and ctDNA analysis each have strengths and limitations, and their findings may be complementary rather than identical. However, a robust detection agreement among key driver events as well as a comparable number of targetable alterations between ctDNA and tissue profiling [15–17] has established the viability of ctDNA-based assays as a substitute for tissue-based testing.

Genomic profiling of ctDNA in patients with advanced cancer for treatment selection

Selecting the right testing approach

Currently, the standard clinical application of genomic profiling of ctDNA is for treatment selection in the advanced cancer setting, meaning detecting alterations that can be matched to targeted therapies or identifying alterations that would be a contraindication for a particular therapy. As with tissue analysis, several scenarios may warrant the limited analysis of a single gene [18–20] or using a cancer hotspot panel ([4, 21]; Table 1). However, the general direction of the treatment selection setting is moving toward employing larger comprehensive genomic profiling (CGP) panels to maximize the detection of therapeutic targets and

Table 1 Comparisons and justifications of current molecular testing approaches

Test	Example use case	Rationale for approach	Benefit	Challenges/limitations
Single-gene testing	Determining neoRAS WT	Anti-EGFR rechallenge in mCRC	Quick Cost-effective Scalable	Interpretation of a negative result without knowing the tumor content in the sample
	Emergence of *ESR1* resistance-related mutations in ER+/HER2- negative breast cancer	SERD treatment to counteract endocrine resistance due to *ESR1* mutations	Quick Cost-effective Scalable	Interpretation of a negative result without knowing the tumor content in the sample Resistance mutations are often subclonal
	Detection of *PIK3CA* activating mutations in HR+/HER2 advanced or metastatic breast cancer	Identification of HR+/HER2– advanced breast cancer who had received endocrine therapy and who may benefit from alpelisib + fulvestrant	Quick Cost-effective Scalable	Interpretation of negative results
Hotspot panels (amplicon)	Lung cancer-specific panel	Availability of many targeted treatments for NSCLC, simultaneous testing of most relevant actionable targets	Quick Cost-effective Simple to interpret	Detection of somatic copy number alterations (SCNAs) may not be reliable Relevant biomarkers such as fusions, MSI, TMB for certain indications are not assessed
Genomic profiling using gene panels	Gene panels (50–150 genes) for treatment selection in patients who have exhausted all standard lines of therapy	Treatment selection in patients who have exhausted all standard lines of therapy	Enables detection of all 4 classes of genomic alterations Enables an aneuploidy-based estimation of tumor fraction Higher probability of detecting actionable alterations	Relevant biomarkers such as fusions not included MSI, TMB cannot be inferred from smaller panels Variant interpretation is more complex Distinction of tumor-derived variants, germline variants, and variants derived from clonal hematopoiesis Often only covers relevant genes for specific tumor entities
	CGP panel (> 500 genes) for treatment selection in patients who have exhausted all standard lines of therapy	Treatment selection in patients who have exhausted all standard lines of therapy	Enables detection of all 4 classes of genomic alterations Enables an aneuploidy-based estimation of the tumor fraction Pan-cancer suited Maximization of actionable insight Inclusion of complex biomarkers like MSI and bTMB	Most expensive Only cost-effective with a high throughput Variant interpretation is more complex Distinction of tumor-derived variants, germline variants, and variants derived from clonal hematopoiesis High likelihood of detecting multiple co-existing alterations Requires MTB discussions

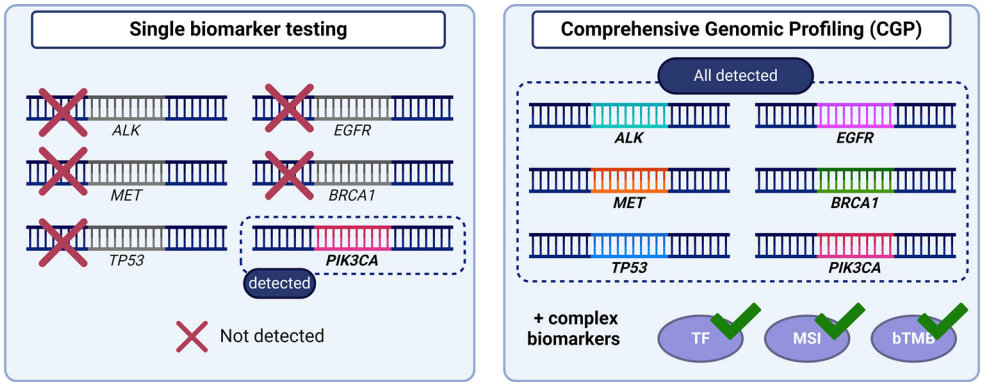

Fig. 1 Single biomarker testing vs. CGP of DNA. Outside of the academic hospital setting, many clinics still predominantly employ single marker testing in their cancer diagnostic workflows. This routine approach is often cancer type-specific and it is able to identify alterations from a pre-specified gene of interest, but the technology overlooks potential existing mutations in other genes (*left*). Comprehensive genomic profiling (CGP) is gaining traction as a pan-cancer approach to detecting all four classes of alterations across hundreds of clinically relevant cancer-associated genes (*right*). CGP extends beyond the limited hotspot mutations and includes insertions and deletions, copy number alterations and fusions all from a single sample and test. In addition, large gene panels can also measure complex biomarkers, such as tumor fraction (TF), microsatellite instability (MSI) and blood tumor mutational burden (bTMB). The goal of CGP via NGS is to maximize the detection of therapeutically relevant and targetable genomic alterations that can be used to direct selection of suitable individualized treatment options

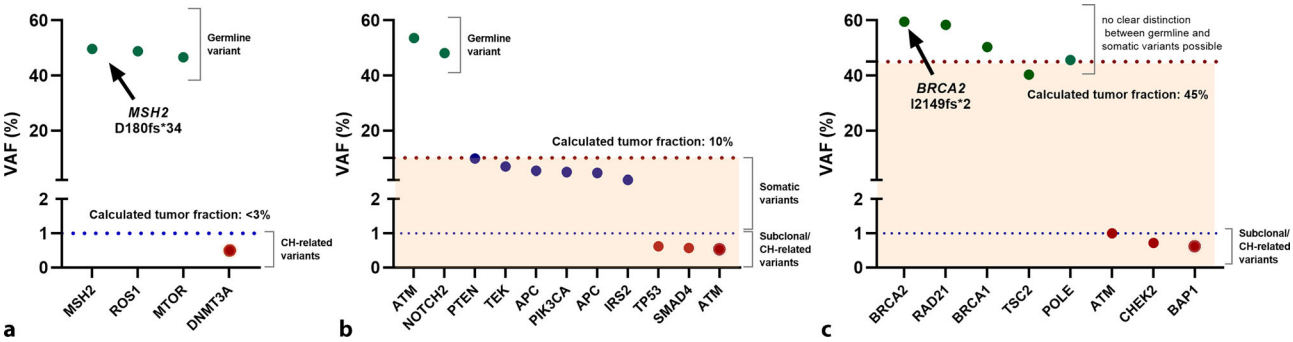

Fig. 2 Potential outcomes of liquid profiling using CGP. **a** Samples with low tumor fraction (TF). Mutations in the range of a variant allele frequency (VAF) of 40–60% indicate germline origin. Pathogenic *MSH2* mutation is a clear indication for genetic counseling regarding Lynch syndrome. **b** Sample with intermediate tumor fractions. While mutations in the range of the estimated TF are likely to be tumor-derived, higher VAFs are more likely associated with germline variants. Variants with substantially lower VAFs than the estimated TF can be of subclonal or hematopoietic origin. **c** Samples with a high TF. VAFs in the range of the estimated TF cannot distinguish between tumor-derived and germline variants. Germline testing and genetic counseling is indicated

to harvest the additional, valuable information such as mutations, copy numbers, fusions, tumor fraction in plasma (Fig. 1; [22, 23]). In fact, several ctDNA-based CGP tests have already received FDA approval/clearance for select indications (Table 2). Putting algorithmically estimated levels of plasma tumor fraction (TF) into context with the detected variant allele frequencies (VAF) of detected mutations can help with the interpretation of detected mutations and indicate whether they are derived from the germline, from subclones or even from the hematopoietic system (Fig. 2). For CGP panels, TF is usually estimated based on tumor aneuploidy measured as deviations in coverage across the genome. When TFs are below 5–10%, such an estimate is no longer informative, while SNVs can still reliably be detected down to 0.1% [16]. If the panel is large enough, also complex biomarkers such as microsatellite instability (MSI) status and blood tumor mutational burden (bTMB), which have significant relevance for immunotherapies, can be inferred [24]. The added insight that can only be obtained from CGP approaches is critical to downstream interpretation of ctDNA results and provides the best comprehensive overview of the patient's sample (Table 1).

Selecting the right analyte: tissue first, plasma first, or both in parallel?

An increasing body of evidence supporting the usage, advantages and limitations of plasma-based CGP for guiding treatment decisions has been instrumental in updating clinical guidelines for routine ctDNA testing. This has led to terminologies and concepts such as "tissue-first", "plasma-first" or matched tissue and

Table 2 List of FDA-cleared or FDA-approved companion diagnostic devices (*in vitro*) for liquid biopsy testing from plasma [32]

Diagnostic Name (Manufacturer)	Indication—Sample Type	Drug Trade Name (Generic) NDA/BLA	Biomarker(s)	Biomarker(s) (Details)	PMA /510(k)/513(f)(2)/HDE (Approval/ Clearance/Grant Date)
Agilent Resolution ctDx FIRST assay (Resolution Bioscience, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Krazati (adagrasib) NDA 216340	*KRAS*	*KRAS* G12C	P210040 (12/12/2022)
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tagrisso (osimertinib) NDA 208065	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P120019/S018 (04/18/2018)
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Tissue or Plasma	Iressa (gefitinib) NDA 206995	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P120019/S019 (08/22/0218)
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Tissue or Plasma	Iressa (gefitinib) NDA 206995	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P120019/S031 (10/27/2020)Group Labeling
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Tissue or Plasma	Tarceva (erlotinib) NDA 021743	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P120019/S031 (10/27/2020)Group Labeling
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Tissue or Plasma	Gilotrif (afatinib) NDA 201292	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P120019/S031 (10/27/2020)Group Labeling
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Tissue or Plasma	Tagrisso (osimertinib) NDA 208065	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P120019/S031 (10/27/2020)Group Labeling
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tagrisso (osimertinib) NDA 208065	*EGFR (HER1)*	T790M	P150044 (09/28/2016)
Cobas EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tarceva (erlotinib) NDA 021743	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P150047 (06/01/2016)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Exkivity (mobocertinib) NDA 215310	*EGFR (HER1)*	Exon 20 insertion mutations	P190032/S005 (05/03/2023)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Iressa (gefitinib) NDA 206995	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P190032 (08/26/2020)P190032/S008 (12/19/2022)Group Labeling
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tagrisso (osimertinib) NDA 208065	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P190032 (08/26/2020)P190032/S008 (12/19/2022)Group Labeling
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tarceva (erlotinib) NDA 021743	*EGFR (HER1)*	Exon 19 deletion or exon 21 L858R substitution mutation	P190032 (08/26/2020)P190032/S008 (12/19/2022)Group Labeling
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	BRAFTOVI (encorafenib) NDA210496 in combination with MEKTOVI (binimetinib) NDA210498	*BRAF*	V600E	P190032/S011 (10/11/2023)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Metastatic Castrate Resistant Prostate Cancer (mCRPC)—Plasma	Rubraca (rucaparib) NDA 209115	*BRCA 1 and BRCA 2*	*BRCA 1* and *BRCA 2* alterations	P190032 (08/26/2020)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tabrecta (capmatinib) NDA 213591	*MET*	*MET* single nucleotide variants and indels that lead to *MET* exon 14 skipping	P190032/S001 (07/15/2021)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Rozlytrek (entrectinib) NDA 212725	*ROS1*	*ROS1* fusions	P190032/S004 (12/22/2022)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Solid Tumors—Plasma	Rozlytrek (entrectinib) NDA 212725	*NTRK1, NTRK2, and NTRK3 fusions*	*NTRK1*/2/3 fusions	P190032/S004 (12/22/2022)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Alecensa (alectinib) NDA 208434	*ALK*	*ALK* rearrangements	P200006 (10/26/2020)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Breast Cancer—Plasma	Piqray (alpelisib) NDA 212526	*PIK3CA*	C420R, E542K, E545A, E545D [1635G>T only], E545G, E545K, Q546E, Q546R, H1047L, H1047R, and H1047Y	P200006 (10/26/2020)

Table 2 (Continued)

Diagnostic Name (Manufacturer)	Indication—Sample Type	Drug Trade Name (Generic) NDA/BLA	Biomarker(s)	Biomarker(s) (Details)	PMA /510(k)/513(f)(2)/HDE (Approval/ Clearance/Grant Date)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Metastatic Castrate Resistant Prostate Cancer (mCRPC)—Plasma	Lynparza (olaparib) NDA 208558	*BRCA 1, BRCA 2 and ATM*	*BRCA 1*, *BRCA 2*, and *ATM* alterations	P200006 (10/26/2020)
FoundationOne Liquid CDx (Foundation Medicine, Inc.)	Metastatic Colorectal Cancer (mCRC)—Plasma	BRAFTOVI (encorafenib) NDA 210496 in combination with cetuximab BLA 125084	*BRAF*	*BRAF* V600E alteration	P190032/S010 (06/08/2023)
Guardant360 CDx (Guardant Health, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Tagrisso (osimertinib) NDA 208065	*EGFR (HER1)*	*EGFR* exon 19 deletions, *EGFR* exon 21 L858R, and T790M	P200010 (08/07/2020)
Guardant360 CDx (Guardant Health, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Rybrevant (amivantamb) BLA 761210	*EGFR (HER1)*	*EGFR* exon 20 insertions	P200010/S001 (05/21/2021)
Guardant360 CDx (Guardant Health, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	Lumakras (sotorasib) NDA 214665	*KRAS*	G12C	P200010/S002 (05/28/2021)
Guardant360 CDx (Guardant Health, Inc.)	Non-Small Cell Lung Cancer (NSCLC)—Plasma	ENHERTU (fam-trastuzumab deruxtecan-nxki) BLA 761139	*ERBB2*	*ERBB2* Activating Mutations (SNVs And Exon 20 Insertions)	P200010/S008 (08/11/2022)
Guardant360 CDx (Guardant Health, Inc.)	Breast Cancer—Plasma	Orserdu (elacestrant) NDA 217639	*ESR1*	*ESR1* missense mutations between codons 310 and 547	P200010/S010 (01/27/2023)
Therascreen PIK3CA RGQ PCR Kit (QIAGEN GmbH)	Breast Cancer—Tissue or Plasma	Piqray (alpelisib) NDA 212526	*PIK3CA*	C420R, E542K, E545A, E545D [1635G>T only], E545G, E545K, Q546E, Q546R, H1047L, H1047R, and H1047Y	P190001 (05/24/2019)P190004 (05/24/2019)

liquid profiling, indicating in which clinical scenario it makes sense to perform initial testing on biopsy material, cfDNA from plasma, or both, respectively. Because each approach has its obvious advantages and disadvantages, it is difficult to design a universal testing strategy for patients with advanced cancer and, in the past, recommendations have often conflicted among authors of clinical guidelines [10, 25–28] Recently, several of these professional multidisciplinary expert panels have reconvened to provide a general but thorough framework for assay and analyte selection for advanced cancer genotyping which are, for reasons of brevity, summarized here [8, 9]. Generally, it is recognized that ctDNA assays have demonstrated utility in the identification of actionable alterations to inform targeted treatment and may be used routinely for the management of advanced cancer patients, but the assay limitations must be considered. The "tissue-first" approach remains the gold standard for the majority of patients, especially as ctDNA assays are often limited in detecting important events of therapeutic relevance, such as fusions and SCNAs. As such, most guidelines stress the importance of the "tissue-first" approach and generally recommend "plasma-first" for most tumor types when tissue material is unavailable or inadequate [8]. However, a "plasma-first" approach may be performed when a quicker turnaround time is critical for a clinical decision, like in aggressive tumors such as advanced NSCLC [8]. Additionally, there are several scenarios in which ctDNA testing is preferred to standard tissue profiling, such as for the detection of *ESR1* mutations in breast cancer and the detection of resistance-related mutations in NSCLC patients who previously received tyrosine kinase inhibitor (TKI) therapy [8]. However, negative ctDNA results can have different interpretations depending on the clinical context and the specific characteristics of the patient and the tumor. While it may suggest the absence of detectable tumor DNA in the bloodstream or a favorable response to treatment, it does not definitively rule out the presence of a mutations, in particular when the sample harbors low tumor content. Therefore, expert guidelines advise reflex tumor testing when non-informative results are obtained. There is also accumulating evidence that performing molecular profiling on both tissue and plasma in parallel significantly enhances the detection of actionable alterations, thus increasing the chance of being able to match patients to targeted therapies [4, 15]. In addition, joint tissue and liquid testing provides valuable complementary, technical and biological information that enables a more holistic interpretation and evaluation of the molecular results.

Table 3 Select issues and open challenges in interpretation of ctDNA testing results

Issue	Explanation	Solution	Open challenges
When is a liquid biopsy result a true negative? How do you determine this?	Incomplete sensitivity of ctDNA assays poses a risk for false-negative results. In certain scenarios, it may be difficult to differentiate between a non-informative result, i.e. a true negative, or if a variant was undetected because of assay resolution limitations, i.e. false negative	In cases of non-informative results, reflex tissue testing can confirm true negatives. In addition, measuring tumor fraction of the sample is central to determining if sufficient ctDNA levels are present to provide informative results	ESMO guidelines: "*Interpretation of a sample as 'truly negative' for fusion variants, or copy number variations, using ctDNA remains difficult. Although assays for detection of tumor fraction are in development, they are still experimental, and not available for routine clinical practice.*"
Potential germline variants may be detected through liquid CGP. How do you infer potential germline variants and when is there an indication for germline follow-up testing?	Although ctDNA profiling primarily targets somatic mutations, it can also incidentally detect potential germline variants, a factor of which both clinicians and patients should be aware of prior to CGP testing. Detection of potential germline variants necessitates a careful discussion of patient history and a subsequent diagnostic workup	If a variant is present at a high VAF in the absence of extensive tumor shedding in the blood, it may suggest a germline origin. This is particularly relevant when the VAF is around 50%, suggesting that the variant may be present in every cell (as is typical for germline variants). Some variants detected might be in genes commonly associated with germline mutations. Particular caution must be taken when interpreting pathogenic variants in high penetrance cancer susceptibility genes (such as *BRCA 1, BRCA 2, PALB2*). If these mutations are known to be common in hereditary cancers and have been documented in germline databases, they might be flagged as potential germline variants. A patient's personal or family history that suggests a hereditary cancer syndrome is an indication for further germline testing. Validated germline testing from blood or saliva should be carried out to confirm germline or somatic nature	Before pursuing germline testing, it is essential to obtain informed consent and provide genetic counseling to discuss the implications of the results for the patient and their family. Germline testing raises considerations about privacy, insurance discrimination, and family dynamics, which need careful handling
How do you determine if a variant is CHIP-associated or tumor-specific?	A significant challenge in employing ctDNA-based CGP arises from the absence of standardized approaches for pinpointing the origins of the variants detected in plasma, which includes mutations related to clonal hematopoiesis (CH). Because CH-related variants are not tumor-specific, it is of utmost important to first determine the variant's origin in order to avoid incorrect treatment matches	Always sequence matched PBMCs at a comparable depth of ctDNA to filter out CH-related mutations	Because an additional sample must be sequenced in parallel to cfDNA, additional costs are incurred, which limits practical application in the clinical setting
		Set a threshold, i.e. any variant with a VAF ≥ 0.5% is tumor-specific	The selected threshold may be arbitrary and may still result in the inclusion of CH-related variants or exclusion of tumor-specific variants

Interpretation of liquid biopsy data poses challenges for integration into routine clinical care

Much emphasis is put on the challenges of the technical implementation of liquid biopsy in the clinic. However, the complexity of downstream interpretation of molecular testing results from ctDNA is often greatly underestimated (Table 3). The increasing broad coverage and high accuracy of liquid CGP panels has propelled the potential of this technology for guiding treatment decisions, but it comes with everyday challenges in interpreting genomic variance from hundreds of genes and alteration types. This information can only be processed accurately, efficiently and with high confidence within the framework of a clinical team that covers diverse medical disciplines, a construct referred to as the molecular tumor board (MTB) [29–31]. MTBs bring together specialists in oncology, pathology, genetics, molecular biology, bioinformatics, patient care and clinical trials. They collaborate to tailor personalized treatment choices for patients, taking into account genomic alterations within their tumors and other relevant clinical factors and data. MTBs may also decide which patients to test, which analytes to assess and which molecular assays to employ. This helps provide clinicians with essential diagnostic, prognostic and actionable insights, enabling them to integrate molecular findings into optimized and individualized care plans for their patients. In routine MTB settings, results from ctDNA testing pose several interpretation challenges that require careful discussion among panel members before reaching treatment or further diagnostic workup recommendations.

Concluding remarks

The accumulating evidence supporting the use of liquid biopsy in routine oncology has paved the way for regulatory approval of several ctDNA-based tests, which has in turn driven an increase in clinical adoption in the advanced disease setting. Currently, one of the main challenges for those starting with liquid biopsy is determining which test best aligns with the clinical question at hand, how many biomarkers should be tested, whether to partner with an academic laboratory or outsource testing to an industry provider, and how to convert NGS readouts into evidence-based treatment decisions, particularly when the results are not as straightforward as with standard tissue testing. While the introduction of CGP has improved the probability of detecting a biomarker-based indication, the additional information provided

in liquid biopsy medical reports has posed interpretation challenges that interfere with streamlined treatment decision-making, thus necessitating the interdisciplinary collaboration among medical professionals in the form of an MTB.

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Conflict of interest S.O. Hasenleithner and E. Heitzer declare that they have no competing interests.

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