

# Key Question Posting Document for Role of Liquid Biopsy in Detection and Management of Cancer in the Medicare Population

## Background

Recent technologic advances have allowed for the isolation and analysis of circulating tumor cells, circulating tumor DNA, and extracellular microvesicles, making the promise of a liquid biopsy possible. A liquid biopsy is defined as the analysis of tumor related material in a sample obtained from the peripheral blood. This material includes intact cells, nucleic acids (DNA or RNA), and proteins. Compared to a tissue biopsy; which requires a surgical or image guided procedure to obtain a tissue sample, blood collection by venipuncture is a much less invasive diagnostic approach.

### **Regulatory considerations:**

Liquid biopsies that are manufactured and performed within a single laboratory [a laboratory with a single certificate issued by the Clinical Laboratory Improvement Amendments (CLIA) program] are considered laboratory developed tests (LDTs). These liquid biopsies are not currently regulated by U.S. Food and Drug Administration (FDA). They are overseen by the CLIA program to ensure analytically accurate and reliable test results. The position of the FDA may change in the future as the FDA has issued a discussion paper on LDTs in 2017 but without an enforceable action.<sup>1</sup> The CLIA final rules specify that the actual performance characteristics of LDTs must be comparable to its claimed specifications in the following areas: accuracy, precision, reportable range of results, reference intervals (normal ranges), analytical sensitivity (detection limit), analytical specificity (interferences/cross reactivity) and any other relevant performance characteristics for the particular test/testing system.

On the other hand, in vitro diagnostic liquid biopsy test kits that are manufactured and then commercially sold to multiple labs are regulated by the FDA and must meet premarket approval (PMA) requirements before they can be marketed. Most liquid biopsy tests are currently LDTs.

Pros and cons of having tests regulated by the FDA vs being considered as LDTs have been often discussed. LDTs are argued to be sufficiently validated by experienced lab staff and oversight from CLIA, can be rapidly developed and updated following the most recent advances of clinical research, and provide more access to patients. Proponents of FDA regulation cite higher standards requiring proof of analytic and clinical validity and clinical utility. From evidence synthesis perspective, stratification by this regulatory issue (designated as LDT vs FDA approved) may help in exploring heterogeneity of results.

### **Clinical applications and dilemmas:**

Potential clinical scenarios in which a liquid biopsy can be used include screening in asymptomatic individuals (at high or average risk), a diagnostic tool in patients suspected to have cancer (thus reducing the need for an invasive tissue biopsy), a therapy-guiding tool in patients with established cancer diagnosis (to aid in treatment decisions such as choosing the initial treatment, determining response to a treatment or modifying a treatment) or a prognostic tool in patients with established cancer diagnosis (to establish remission and future prognosis).

In terms of screening (identifying disease in an early stage aiming at reducing cancer mortality and morbidity), one important challenge is that patients with early-stage disease may harbor <1 mutant template molecules per milliliter of plasma, which is beyond the limit of detection of conventional sequencing.<sup>2</sup> Screening also requires studies with a very large sample size because cancer incidence in asymptomatic individuals is generally low; therefore, sensitivity and specificity cannot be reliably estimated.<sup>2</sup> An important dilemma in the realm of screening with a liquid biopsy is that one should not forget that certain conditions, such as autoimmune disorders, are associated with increased concentrations of cell-free DNA.<sup>3</sup> Newer technology may help in distinguishing cancer related DNA from this background DNA.

In the literature, there are a few instances in which a liquid biopsy was used as a diagnostic tool. For example, testing the peripheral blood to detect stage I to IV colorectal cancer.<sup>4</sup> There is great uncertainty about which cancer and what setting is appropriate for this important and pivotal clinical step (establishing the diagnosis). A traditional biopsy will likely be preferred in many situations, particularly because patients may anyway require an excision of a mass and a surgical intervention. However, it is plausible that there are instances in which a liquid biopsy would be preferred over an invasive tissue biopsy such as in poorly accessible tumors and in tumors not routinely biopsied or excised, such as advanced hepatocellular carcinoma, or in patients with a high disease burden (in whom ctDNA may be in quantities sufficient to be a surrogate for tissue sampling).<sup>2</sup> The presence of heterogeneity between the primary tumor and metastasis presents another practical challenge in adequate tissue sampling that may be overcome by ctDNA profiling. This was shown in a case report of a patient with synchronous ductal-lobular carcinoma with bone and liver metastases that displayed intra-tumor genetic heterogeneity at diagnosis.<sup>5</sup>

In terms of liquid biopsy helping with management and treatment choice, an example would be identifying non-small cell lung cancer that acquired resistance to EGFR tyrosine kinase inhibitors.<sup>6</sup> As for establishing prognosis, an example is using liquid biopsy to identify oncogenic drivers in lung cancer; which can predict survival.<sup>7</sup>

One important challenge in the literature of liquid biopsy is that it has been derived from nonrandomized studies, in which confounding and selection bias may occur. Statistical techniques such as multivariable adjustment or propensity score analysis may not adequately account for known or unknown confounders.

The limited and variable sensitivity of liquid biopsies may reflect the biology that some cancers do not shed sufficient amounts of DNA into the bloodstream; which is impacted by factors such as the extent of the disease in terms of stage and number of metastatic sites.<sup>8</sup> Another challenge that faces oncologists is that in patients receiving cytotoxic chemotherapy, leukocyte and erythrocyte apoptosis is expected. Hence, an increase in DNA fragments in the plasma could be from the death of these cells rather than tumor cells. Furthermore, it remains unclear whether ctDNA released from cancer cells is due to their death from therapy or due to the fact that they are resistant to therapy. Lastly, certain conditions, such as autoimmune disorders, are associated with increased concentrations of cell-free DNA; which can affect the accuracy of a liquid biopsy.<sup>3</sup> Thus, the analytic validity may differ in various subpopulations of patients.

### **Validity of a medical test:**

For a medical test such as the liquid biopsy to be used in practice, several conditions are required.<sup>9</sup> The test needs to have sufficient 1) pre-analytic validity (i.e., the test needs to conform to technical specifications that relate to the collection, handling and storage of the specimen); 2) analytic validity (i.e., the test needs to measure the biomarker in an accurate manner concordant with a gold or reference standard), 3) clinical validity (i.e., the test needs to have diagnostic accuracy in classifying the target population) and 4) clinical utility (i.e., the test needs to demonstrate improvement in patients' management and outcomes).

Therefore, a systematic review of the utility of liquid biopsy should collect data on these domains of validity and consider them as markers of methodological quality and possible covariates that can explain heterogeneity.

### **Draft Key Questions**

KQ 1: In adults at risk for lung, prostate, breast, ovarian, or colorectal cancer who are undergoing a liquid biopsy (CTCs and ctDNA) as a screening test:

KQ1a: What are the pre-analytic, analytic and clinical validity of a liquid biopsy?

KQ1b: What is the clinical utility of a liquid biopsy?

KQ 2: In adults suspected to have lung, prostate, breast, ovarian, or colorectal cancer who are undergoing a liquid biopsy (CTCs and ctDNA) to establish a diagnosis:

KQ2a: What are the pre-analytic, analytic and clinical validity of a liquid biopsy?

KQ2b: What is the clinical utility of a liquid biopsy?

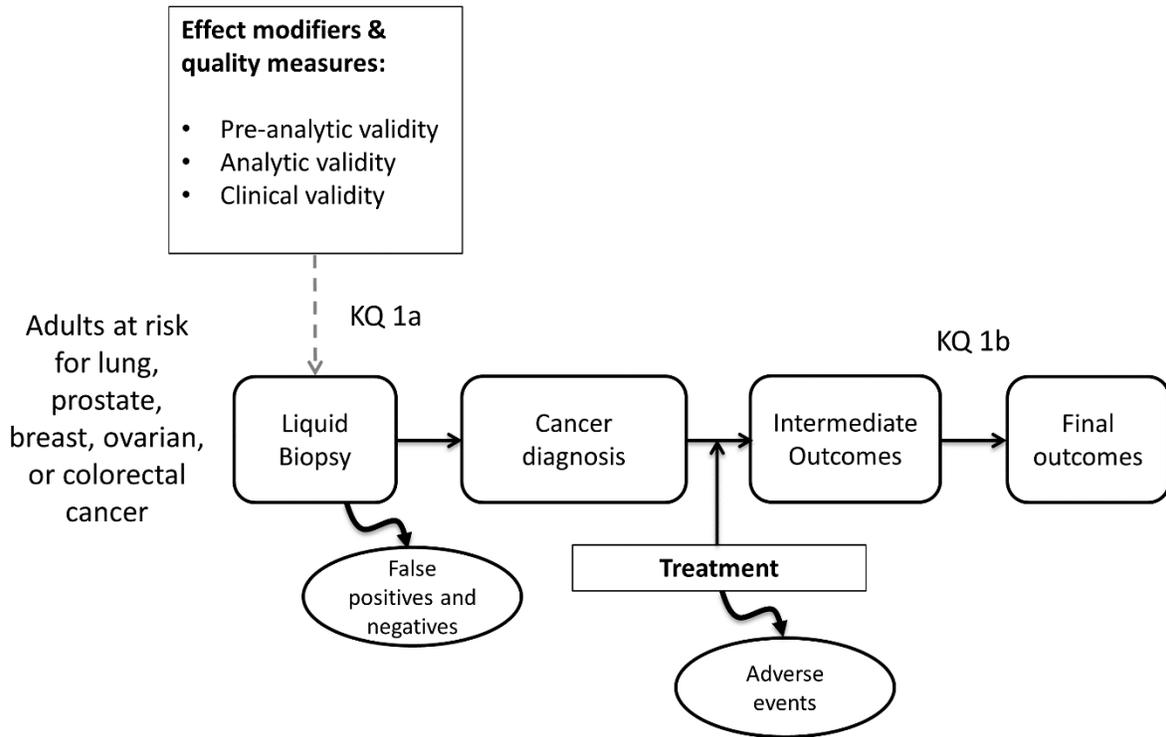
KQ 3: In adults with established diagnosis of lung, prostate, breast, ovarian, or colorectal cancer who are undergoing a liquid biopsy (CTCs and ctDNA) to guide therapeutic decisions:

KQ3a: What are the pre-analytic, analytic and clinical validity of a liquid biopsy?

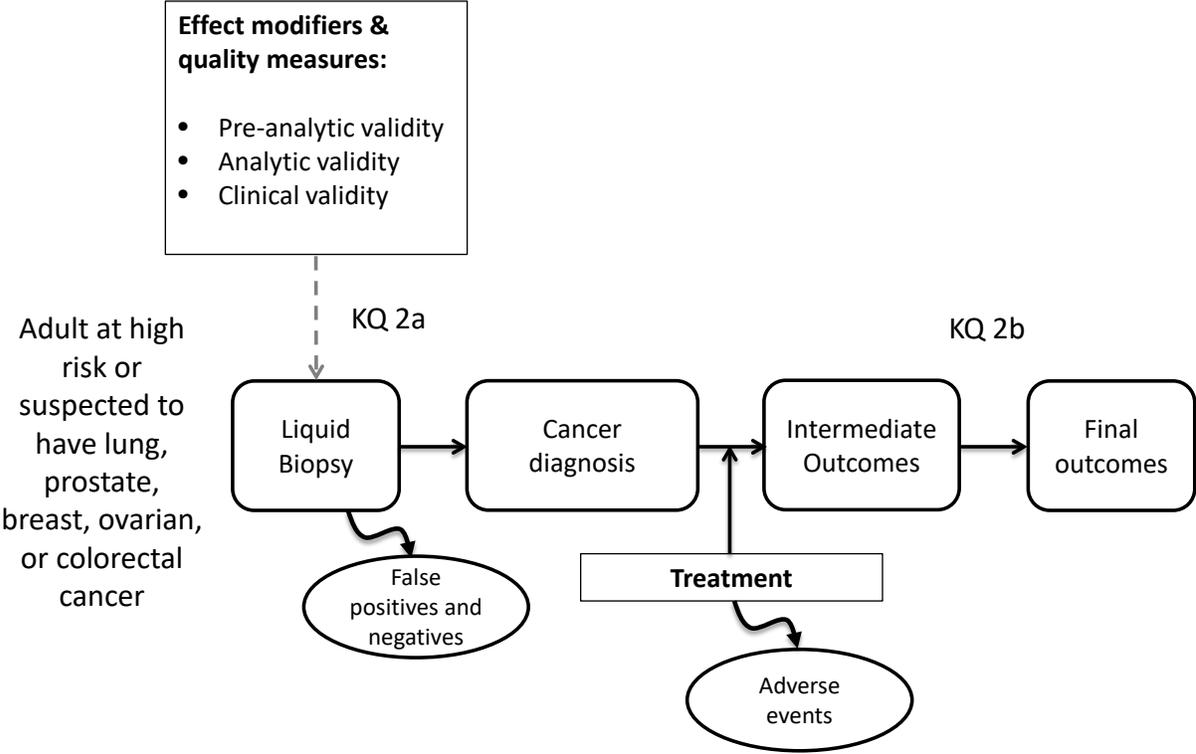
KQ3b: What is the clinical utility of a liquid biopsy?

The analytic framework for these 3 KQs is depicted in Figure 1.

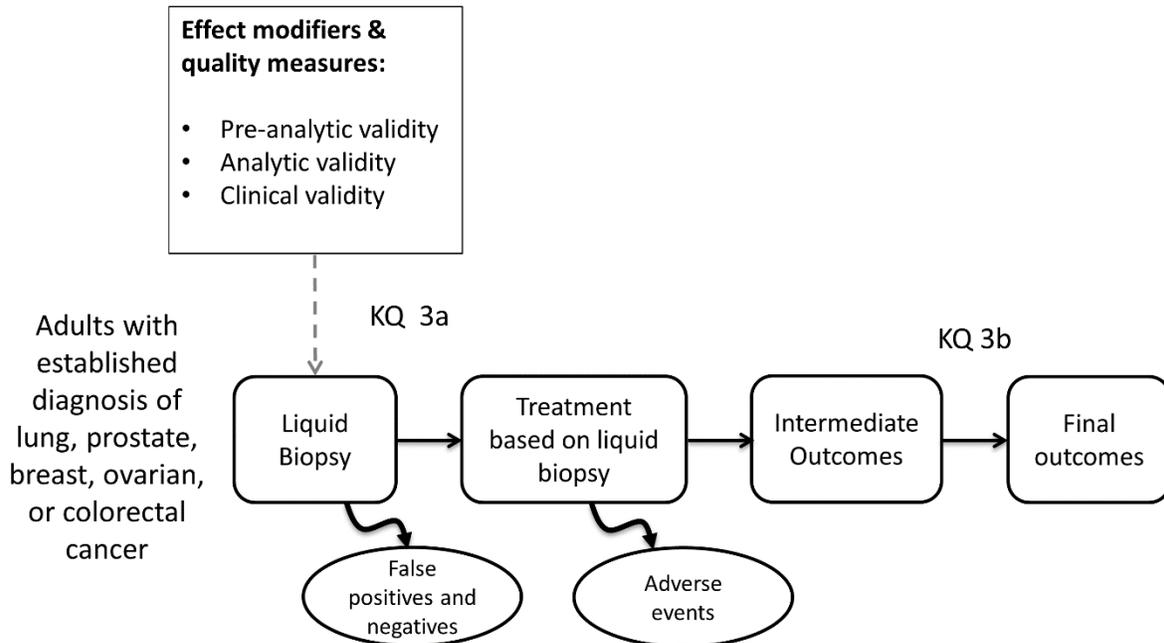
**Figure 1 A. Liquid biopsy used in a screening paradigm**



**Figure 1 B. Liquid biopsy used in a diagnosis paradigm**



**Figure 1 C. Liquid biopsy used in a therapeutic paradigm**



**PICOTS**

Population(s)

- Adult patients (18 years and older) with focus on the Medicare population
- Patients at risk (KQ1), suspected to have (KQ2), or have an established diagnosis (KQ3), of:
  - lung cancer
  - prostate cancer
  - breast cancer
  - ovarian cancer
  - colorectal cancer

Intervention (test)

- Liquid biopsy based on circulating tumor cells (CTCs) and circulating tumor DNA (ctDNA)

Comparators

- Tissue biopsy
- Management without tissue or liquid biopsy

## Outcomes

- Intermediate outcomes
  - Sensitivity, specificity, inter- and intra-laboratory reproducibility (domains of analytic and clinical validity)
  - Lymph node status, depth of invasion, distant metastases, pathological complete response, change in circulating tumor cell, and need for additional tissue biopsy tests (domains of clinical utility)
- Final outcomes
  - Overall survival, disease free survival, treatment response recurrence, and quality of life (domains of clinical utility)

## Timing

- Any duration of follow-up

## Settings

- Oncology clinical settings (outpatient and hospital settings)

## Subgroup analyses/possible effect modifiers

- Designated as LDT vs. FDA approved test
- Patient characteristics such as autoimmune disorders
- Different assays
- Adequacy of pre-analytic validity

## **References**

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