Haystack Oncology
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Applying precision technology to improve the detection of residual disease in cancer patients

A next-generation diagnostic test developed by Haystack Oncology detects the presence of minimal residual disease in early-stage cancer patients after curative-intent surgery. By identifying residual disease—which Haystack achieves through accurately measuring circulating tumor DNA in blood—clinicians can make more-informed decisions to help their patients achieve better outcomes.

Numerous advances in precision oncology have led to better outcomes for patients; however, solid tumors still account for an overwhelming number of cancer-related deaths. The majority of these cancers are diagnosed before distant metastases are present (stages I–III), when surgical intervention offers the greatest potential for cure. Unfortunately, many patients undergoing surgery recur as a result of residual disease that persists after surgery. This remaining cancer, referred to as minimal residual disease (MRD), is detectable by the presence of circulating tumor DNA (ctDNA) and can lead to disease relapse if left untreated.

The ability to detect MRD can help physicians make better-informed decisions about their patients' treatment. Per the current standard of care, patients who have a high risk of recurrence as determined by clinicopathologic characteristics are given post-operative or adjuvant chemotherapy. As a result, many patients who are cured by surgery alone still receive adjuvant therapy and suffer its associated risks and costs. To identify patients who are most likely to benefit from adjuvant therapy, detection of ctDNA can be used to indicate the presence of low-level disease, which may be indiscernible by traditional clinical or radiographic assessment. However, detecting MRD requires a test that demonstrates exquisite sensitivity and specificity because ctDNA exists at an extremely low level in this setting.

"To mitigate the risk of recurrent disease, we often overtreat patients with stage II colorectal cancer based on pathological and clinical criteria, and physician choice. The clinical benefit for adjuvant therapy for these patients is modest," said Joshua Cohen, co-founder and chief innovation officer at Haystack Oncology and a clinician researcher at Johns Hopkins University School of Medicine in Baltimore, Maryland. "In stage III patients, the benefit of adjuvant treatment is undisputed, but we still need to know how aggressively these patients should be treated, as many receive a regimen that is not optimal."

Landmark MRD and beyond

MRD testing can better stratify patients by risk of recurrence compared to the current standard of care because ctDNA provides higher-resolution detection of persistent disease in the post-surgical setting. Patients for whom MRD is detected are likely to benefit from additional treatment, while patients for whom MRD is not detected are unlikely to recur, even without additional therapy. Ultimately, ensuring that patients who need therapy receive it, while also reducing overtreatment, greatly improves outcomes and quality of life and can reduce healthcare costs.

Beyond post-surgical evaluation, MRD testing can help inform other critical clinical decision points. For example, ctDNA testing allows clinicians to discern the effectiveness of treatment in real time so they can adapt therapy to match changes in disease burden. For patients who demonstrate no evidence of clinical disease, ctDNA can also be used to monitor molecular

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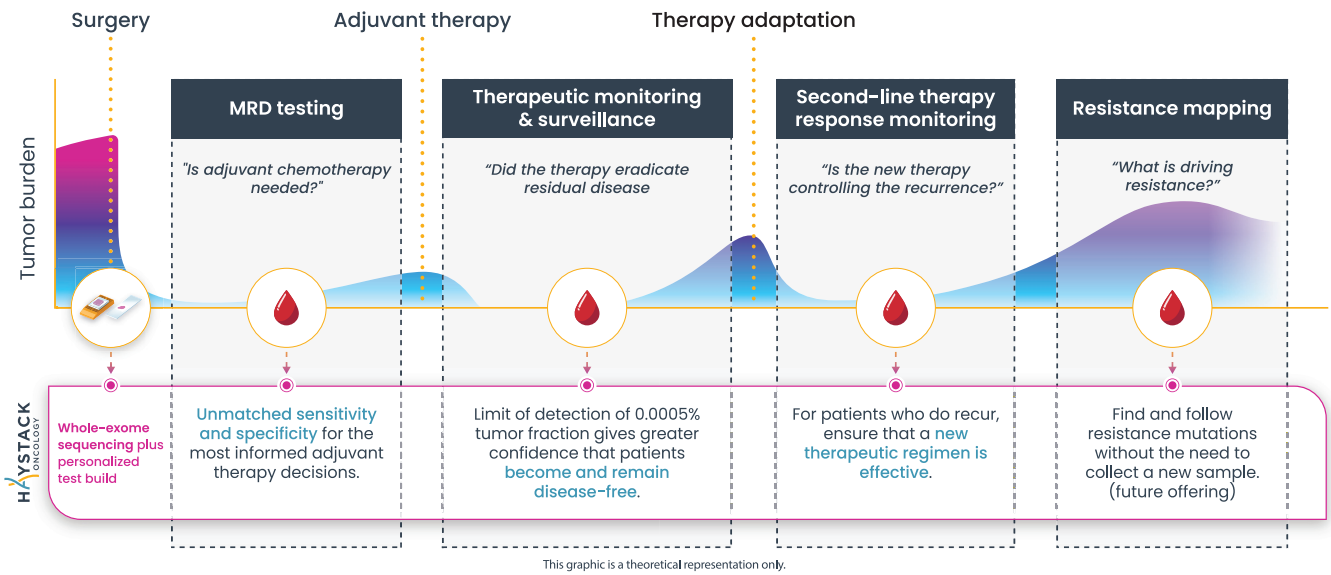

Fig. 1 | Haystack MRD offers superior information across all post-surgical clinical decision points.

recurrence, which has been shown to precede recurrence detected by radiography, often with a significant lead time. This early detection may present a window of opportunity where therapy can be applied when the greatest potential exists to eradicate cancer recurrence (Fig. 1).

"Every diagnostic is rooted in a biomarker. Many of the non-genetic biomarkers used in cancer aren't sensitive or specific enough to accurately find all cases," said Cohen. "Because cancer is genetic at its core, detecting the mutations driving the disease can create a more potent diagnostic."

MRD can also be used to accelerate and streamline the clinical development of novel therapies. For instance, adjuvant trials can be enriched for subjects who are not successfully cured by surgery alone by enrolling only patients who are MRD-positive, rather than enrolling all-comers. This approach not only reduces the size of adjuvant trials but also allows easier visualization of therapeutic efficacy by excluding subjects for whom the disease is no longer present.

"Cancer clinical trials can take years. If we can select the patients who have residual disease, we may be able to conduct smaller trials and get faster results," said Peter Gibbs, professor of medicine at The Walter and Eliza Hall Institute (WEHI) of Medical Research in Melbourne, Australia.

Beyond systemic chemotherapy, MRD testing can help elucidate response to targeted agents such as kinase inhibitors and immuno-oncology therapies including checkpoint inhibitors and personalized cancer vaccines. "As an example, in stage IV melanoma, the test would allow clinicians to gauge patient response to therapy with higher resolution than radiography, and to track the evolution of the tumor over time," said Cohen.

Seeing through the noise

The primary challenge of ctDNA detection is the low level of tumor DNA in circulation compared to normal cell-free DNA from white blood cells. For every molecule of ctDNA, there may be a million or more normal DNA molecules that a test must search through to uncover the cancer-specific signal. As difficult as ctDNA detection can be for late-stage disease, which is known to exhibit relatively high tumor DNA 'shedding' into circulation, one of the most challenging settings for reliable detection is in early-stage patients following curative-intent surgery. Non-metastatic patients are known to have less tumor-derived DNA present in their blood, a scarcity which is underscored after the primary tumor has been removed and there are few, if any, remaining tumor cells. Therefore, a liquid biopsy–based test that is designed for MRD must employ a fundamentally different method than a test that is used for ctDNA detection in late-stage patients.

Haystack's Duo technology is the first next-generation sequencing method that was specifically designed for MRD for solid tumors. By offering significantly reduced technical noise compared to other next-generation sequencing (NGS) methods, even the lowest-level ctDNA signals stand out for reliable detection; in other words, exquisite specificity is what enables Haystack MRD's unmatched sensitivity.

The test is personalized for each patient—tumor tissue is sent to Haystack for sequencing, and a bespoke Duo panel is built for each patient to assess up to 50 mutations that were identified in the tumor tissue. The test is then performed on patients' blood samples to detect and measure ctDNA levels, which indicate the presence and quantity of residual, recurrent, or resistant disease.

"By taking a tumor sample and sequencing it, we know the 'ground-truth' mutations present for that individual patient's cancer. This significantly increases our ability to find those known mutations in the blood, at the lowest levels," said Cohen.

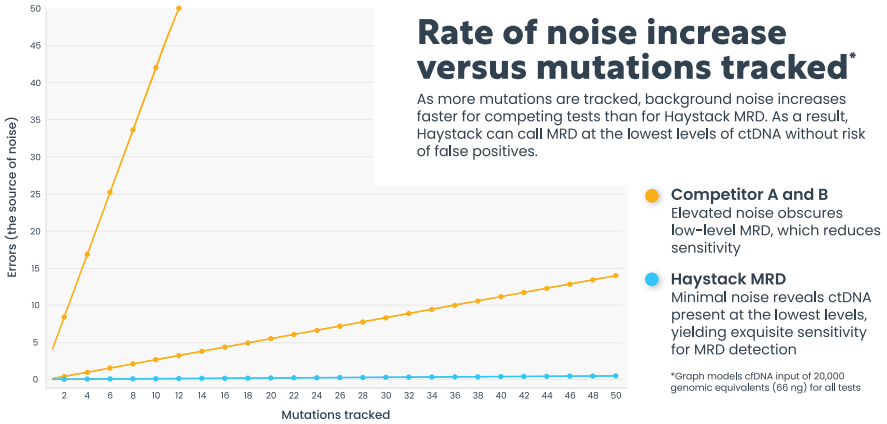

Fig. 2 | Haystack Duo technology reduces background noise for better ctDNA detection in the MRD setting.

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Personalized MRD tests are designed to detect multiple tumor-informed mutations, with each mutation tracked representing another chance to detect ctDNA. However, each additional position sequenced can also introduce background noise that increasingly obscures accurate ctDNA detection. This is why the primary purpose of Haystack Duo is to eliminate background noise—it ensures that the ctDNA signal increases faster than noise as additional mutations are tracked, resulting in improved sensitivity for MRD. In contrast, other tests that track 16 or even 1,800 mutations struggle to sift through elevated noise and must employ complex, proprietary methods to try to discern low-level ctDNA signals from background noise (Fig. 2). High background noise limits the sensitivity of other MRD tests; as the false-negative rate increases, so does the risk that patients may not receive appropriate treatment.

The first prospective interventional trial for solid-tumor MRD

The landmark DYNAMIC clinical trial for patients with stage II colon cancer was conducted at WEHI in Australia and Johns Hopkins Kimmel Cancer Center in the US; this represented the first prospective, randomized interventional MRD trial to generate practice-changing results. In the trial, the findings of which were reported at the American Society of Clinical Oncology (ASCO) meeting in 2022, post-surgery treatment decisions in the experimental arm were informed by ctDNA results, where ctDNA-positive (that is, MRD-positive) patients received adjuvant therapy and ctDNA-negative patients did not. Outcomes were compared to the results of a standard-of-care arm in which patients were managed according to traditional clinicopathological features.

Compared to the standard-of-care arm, patients whose treatment was ctDNA-informed were 50% less likely to receive adjuvant therapy, with no negative impact on 2-year recurrence-free survival. Overall, DYNAMIC reported that MRD was detected in 15.3% of stage II colorectal cancer (CRC) patients, versus 7.6% in the recently published observational CIRCULATE study, which used a first-generation MRD method. ctDNA was detected in DYNAMIC using a next-generation sequencing method developed by researchers at Johns Hopkins and demonstrated a distinct clinical benefit for guiding adjuvant chemotherapy. "This was a paradigm-changing clinical trial. It showed that physicians could reduce adjuvant-chemotherapy use without compromising survival," said Gibbs. Haystack's Duo technology is based on the method developed by Cohen and others, led by Bert Vogelstein at Johns Hopkins University, and represents an evolution of the MRD detection method used in the DYNAMIC study. Regarding Haystack's technology, Gibbs stated, "Duo is the culmination of decades of pioneering work in ctDNA and the only MRD [technology] to have been validated in randomized controlled trials."

CONTACT
Jared Snider, Director of Marketing
Haystack Oncology
Baltimore, MD, USA
Tel: +1-410-348-1918
Email: media@haystackoncology.com