Short Communication

TJ Tumori Journal

Multi-cancer early detection test sensitivity for cancers with and without current population-level screening options

Tumori Journal
1–7

DOI: 10.1177/03008916221133136
journals.sagepub.com/home/tmj

Spencer H Shao[1], Brian Allen[2*], Jessica Clement[3], Gina Chung[4], Jingjing Gao[2], Earl Hubbell[2], Minetta C Liu[5*], Charles Swanton[6,7], WH Wilson Tang[8], Habte Yimer[9] and Mohan Tummala[10]

Abstract

There are four solid tumors with common screening options in the average-risk population aged 21 to 75 years (breast, cervical, colorectal, and, based on personalized risk assessment, prostate), but many cancers lack recommended population screening and are often detected at advanced stages when mortality is high. Blood-based multi-cancer early detection tests have the potential to improve cancer mortality through additional population screening. Reported here is a post-hoc analysis from the third Circulating Cell-free Genome Atlas substudy to examine multi-cancer early detection test performance in solid tumors with and without population screening recommendations and in hematologic malignancies. Participants with cancer in the third Circulating Cell-free Genome Atlas substudy analysis were split into three subgroups: solid screened tumors (breast, cervical, colorectal, prostate), solid unscreened tumors, and hematologic malignancies. In this post hoc analysis, sensitivity is reported for each subgroup across all ages and those aged ⩾50 years overall, by cancer, and by clinical cancer stage. Aggregate sensitivity in the solid screened, solid unscreened, and hematologic malignancy subgroups was 34%, 66%, and 55% across all cancer stages, respectively; restricting to participants aged ⩾50 years showed similar aggregate sensitivity. Aggregate sensitivity was 27%, 53%, and 60% across stages I to III, respectively. Within the solid unscreened subgroup, aggregate sensitivity was >75% in 8/18 cancers (44%) and >50% in 13/18 (72%). This multi-cancer early detection test detected cancer signals at high (>75%) sensitivity for multiple cancers without existing population screening recommendations, suggesting its potential to complement recommended screening programs.

Clinical trial identifier: NCT02889978.

Keywords

Multi-cancer early detection, early detection, cancer screening, liquid biopsy, precision medicine, epidemiology and prevention

Date received: 11 May 2022; revised: 23 September 2022; accepted: 26 September 2022

[1]Compass Oncology, Portland, OR, USA
[2]GRAIL, LLC, a subsidiary of Illumina Inc., Menlo Park, CA, USA†
[3]Hartford Healthcare, Hartford, CT, USA
[4]The Christ Hospital Health Network, Cincinnati, OH, USA
[5]Mayo Clinic, Rochester, MN, USA
[6]The Francis Crick Institute, London, UK
[7]University College London Cancer Institute, London, UK
[8]Cleveland Clinic, Cleveland, OH, USA
[9]Texas Oncology/USON, Tyler, USA
[10]Mercy Springfield, Springfield, MO, USA

*At the time of the study
†Currently held separate from Illumina Inc. under the terms of the Interim Measures Order of the European Commission dated 29 October 2021

Corresponding author:
Spencer H Shao, Compass Oncology, 265 N Broadway, Portland, OR 97227, USA.
Email: Spencer.Shao@compassoncology.com

Introduction

In recent decades, reductions in cancer mortality have likely been driven by reductions in smoking, improvements in treatment, and earlier detection through screening.[1] For the average-risk population aged 21–75 years, there are four solid tumors with common screening options.[2–5] The United States Preventive Services Task Force (USPSTF) recommends breast, cervical, and colorectal cancer screening based on age, and prostate cancer based on personalized risk assessment. Lung cancer screening is only recommended by the USPSTF for high-risk individuals with a smoking history.[6] Less than 13% of the 55–80-year-old population met lung cancer screening criteria in 2017.[7] There is an unmet need for cancer population screening for average-risk individuals beyond just four cancers.

Most cancer deaths occur from those that are not addressed by current screening guidelines.[1] Furthermore, individuals who are recommended for cancer screenings have a greater risk of receiving a different cancer diagnosis than the one being screened.[8] Screening for more cancers may address this unmet need.

Multi-cancer early detection (MCED) testing has the potential to address this need and reduce cancer mortality. An MCED test (Galleri®) (GRAIL, LLC, a subsidiary of Illumina, Inc., Menlo Park, CA, USA) was validated in the third and final Circulating Cell-free Genome Atlas (CCGA) substudy.[9] This test uses cell-free DNA (cfDNA) targeted methylation signals to detect a shared cancer signal and predict cancer signal of origin with a specificity of 99.5% (false-positive rate, 0.5%) across multiple cancer types that collectively account for more than two-thirds of annual US cancer deaths.[9]

Clinical use necessitates understanding how the MCED test may complement existing screening paradigms for the average-risk population. We performed a post-hoc analysis from the third and final CCGA substudy[9] of the MCED test performance for three subgroups: solid tumors with and without common screening options for the average-risk population, and hematologic malignancies. Of note, sample sizes for individual cancers were small, limiting statistical comparisons. Hematologic malignancies were analyzed separately because diagnostic investigation for hematologic malignancies is substantially different than that for most solid tumors.

Methods

The CCGA study (NCT02889978) is an observational, multi-center (142 sites across North America), case-control study that was analyzed in three prespecified substudies, as previously described (see online Supplementary Methods).[9,10]

The third substudy of CCGA validated a targeted methylation-based MCED screening test using cfDNA sequencing for population use[9] and is the basis for the post-hoc analysis reported here (cancers only). The primary analysis of the third CCGA substudy included participants with and without cancer. Cancer participants (≥20 years) included in CCGA were those who were either enrolled with a confirmed cancer diagnosis at enrollment, or enrolled with high cancer suspicion and subsequently confirmed with a cancer diagnosis through a biopsy or surgical resection within the enrollment window. Diagnoses may have been prompted by clinical symptoms, routine screening, or incidental detection of a different cancer during screening. Potential cancer participants were excluded if they had received treatment for cancer including chemotherapy or radiotherapy, definitive local therapy, or more extensive surgery than that required to confirm diagnosis before blood draw.

Blood sample collection and processing was performed as previously described[9] to generate targeted methylation data per genomic region from cfDNA for each sample (see online Supplementary Methods). Samples were randomized across processing batches, operators, and reagent lots to reduce the potential for bias. During processing, up to 75 ng plasma cfDNA underwent customized bisulfite conversion as a dual indexed sequencing library, and was enriched using hybridization capture for 150 bp paired-end sequencing (Illumina NovaSeq).

Clinical data were abstracted from medical records, pathology reports, and radiology reports and entered into a validated electronic data capture system. Clinical stage was assigned by the treating physician/a certified cancer registry professional according to the AJCC Staging Manual (7th/8th edition).[9] Cancers without staging classification in the manual were analyzed without staging information. Cancers were classified as previously published cancer classes (see online Supplementary Methods).[9]

A custom machine learning model was used to recognize methylation patterns in cfDNA for each sample as indicative of a cancer signal or no cancer signal. Samples from the third CCGA substudy were used for independent validation of the classifier.[9]

A post-hoc analysis of cancer signal detection results from the third substudy of CCGA was performed to understand the performance of the MCED test in solid tumors with recommendations for average-risk population screening, solid tumors without recommendations for population screening, and in hematologic malignancies (all of which have no recommended screening). The subgroup of solid tumors with population screening ('solid screened') included breast, cervical, colorectal, and prostate cancers (lung cancer was excluded since screening is exclusively recommended for high-risk individuals). The subgroup of solid tumors without recommended population screening ('solid unscreened') included all carcinomas, sarcomas, and melanomas, excluding solid screened tumors. The subgroup of hematologic malignancies ('heme malignancy') included myeloid neoplasms, lymphoid leukemias, lymphomas, and plasma cell neoplasms, representing a subset of cancers with a wide spectrum of severity and aggressiveness, many of which are not stageable.

Cancer signal detection performance by the MCED test was measured by sensitivity. Sensitivity was calculated in each subgroup as the percent of cancer participants in whom a cancer signal was detected. Sensitivity within each subgroup was calculated across all ages and those aged ⩾50 years overall, by cancer, and by clinical cancer stage. Sensitivity in the solid screened subgroup was additionally calculated for those participants whose cancer was found through screening versus clinical presentation.

Results

A total of 2794 participants with cancer from the third and final CCGA substudy were included in this post-hoc analysis; 1175 participants were included in the solid screened subgroup, 1336 in the solid unscreened subgroup, and 283 in the heme malignancy subgroup.

Aggregate sensitivity across all stages and all ages was 34% (95% CI: 31%, 37%) in the solid screened subgroup, 66% (95% CI: 63%, 68%) in the solid unscreened subgroup, and 55% (95% CI: 49%, 61%) in the heme malignancy subgroup (including cancers that were not expected to be staged). Restricting to participants aged ⩾50 years showed similar aggregate sensitivity (30% [95% CI: 27%, 33%], 66% [95% CI: 64%, 69%], and 54% [95% CI: 48%, 60%], respectively). Across stages I–III for all ages, aggregate sensitivity was similar or lower (27% [95% CI: 25%, 30%], 53% [95% CI: 49%, 56%], 60% [95% CI: 53%, 67%], respectively).

Sensitivity differed by specific cancers within each subgroup (Figure 1A). Within the solid screened subgroup, sensitivity was >75% for cervical and colorectal cancers and <40% for prostate and breast cancers across all stages. Within the solid unscreened subgroup (18 cancers), sensitivity of cancer signal detection across all stages was >75% in eight (44%) cancers and >50% in 13 (72%) cancers. Cancer-specific sensitivity in the heme malignancy subgroup (including cancers that were not expected to be staged) ranged from 20%–100% across all stages.

In the solid unscreened subgroup, sensitivity across stages I–III was >75% in five (28%) cancers and >50% in nine (50%) cancers (Figure 1B). In the solid screened and heme malignancy subgroups, cancer-specific sensitivity across stages I–III was similar to sensitivities across all stages.

Sensitivity generally increased by stage within the solid screened and solid unscreened subgroups and was similar across stages II–IV for the heme malignancy subgroup (Figure 2A). Within the solid screened tumors, prostate cancer had a sensitivity of 6% (95% CI: 4%, 8%) across stages I–III, but stage IV prostate cancer had a sensitivity of 83% (95% CI: 66%, 93%). Breast cancer had a sensitivity of 31% (95% CI: 27%, 35%) across all stages, but had a sensitivity of 86% (95% CI: 74%, 92%) and 91% (95% CI: 72%, 98%) in stages III–IV, respectively. In cervical and colorectal cancers, cancer signals were readily detected across all stages (Figure 2B). Details on sensitivity of cancer signal detection by cancer and stage can be found in online Supplementary Table 1.

Participants diagnosed with cancer were enrolled in CCGA following clinical presentation or screening. Important clinical differences and outcomes exist between cancers that present clinically versus those detected by screening.[11] For stages I–III in the solid screened subgroup, this MCED test had a numerically higher sensitivity to detect cancer signals from cancers diagnosed through clinical presentation than through screening (Figure 3).

Discussion

We examined MCED test performance across cancer subgroups, providing evidence that it may serve as a useful screening tool for the average-risk population. The aggregate sensitivity of cancer signal detection was 66% in the solid unscreened subgroup and >75% for many unscreened cancers, indicating that the MCED test may expand the volume of screen-detectable cancers beyond those captured by USPSTF recommendations. Further, cancer signals were detected from unscreened cancers that contribute to significant cancer-related mortality.[12] Importantly, MCED test performance in participants aged ⩾50 years was similar to performance for all ages across all subgroups. A negative MCED test result may not rule out commonly screened cancers, especially early stage; patients should be encouraged to continue recommended single-cancer screenings. MCED may be useful alongside existing screening, not as a replacement.

In the solid screened subgroup, the MCED test could detect cancer signals with relatively high sensitivity (>80%) for stages II–IV colorectal and cervical cancers and for later stages of prostate (stage IV) and breast cancers (stages III–IV). Cancer signal detection rates in prostate (stages I–III) and breast cancers (stage I) were numerically lower than colorectal and cervical cancers in those stages. Notably, more than four times more participants with prostate and breast cancers were enrolled at stages I–II than III–IV (colorectal and cervical cancers were more evenly represented across stages). Thus, as expected, the aggregate sensitivity in the solid screened subgroup was skewed toward the sensitivity of early-stage breast and prostate cancers.

The relatively lower sensitivity observed in early-stage breast and prostate cancers suggests that the sensitivity of this MCED test may be less than currently recommended screening for breast and prostate cancers. Notably, single-cancer screening tests value sensitivity over specificity to maximize the number of cancers detected. However, MCED tests are able to detect more cancers with high specificity and moderate sensitivity given the aggregate prevalence of multiple cancers.[13] It is interesting to also consider this result in the context of evidence from the second CCGA substudy, which indicated that the MCED test may preferentially detect cancer signals from clinically significant cancers.[14] Concordantly, evidence has shown that current

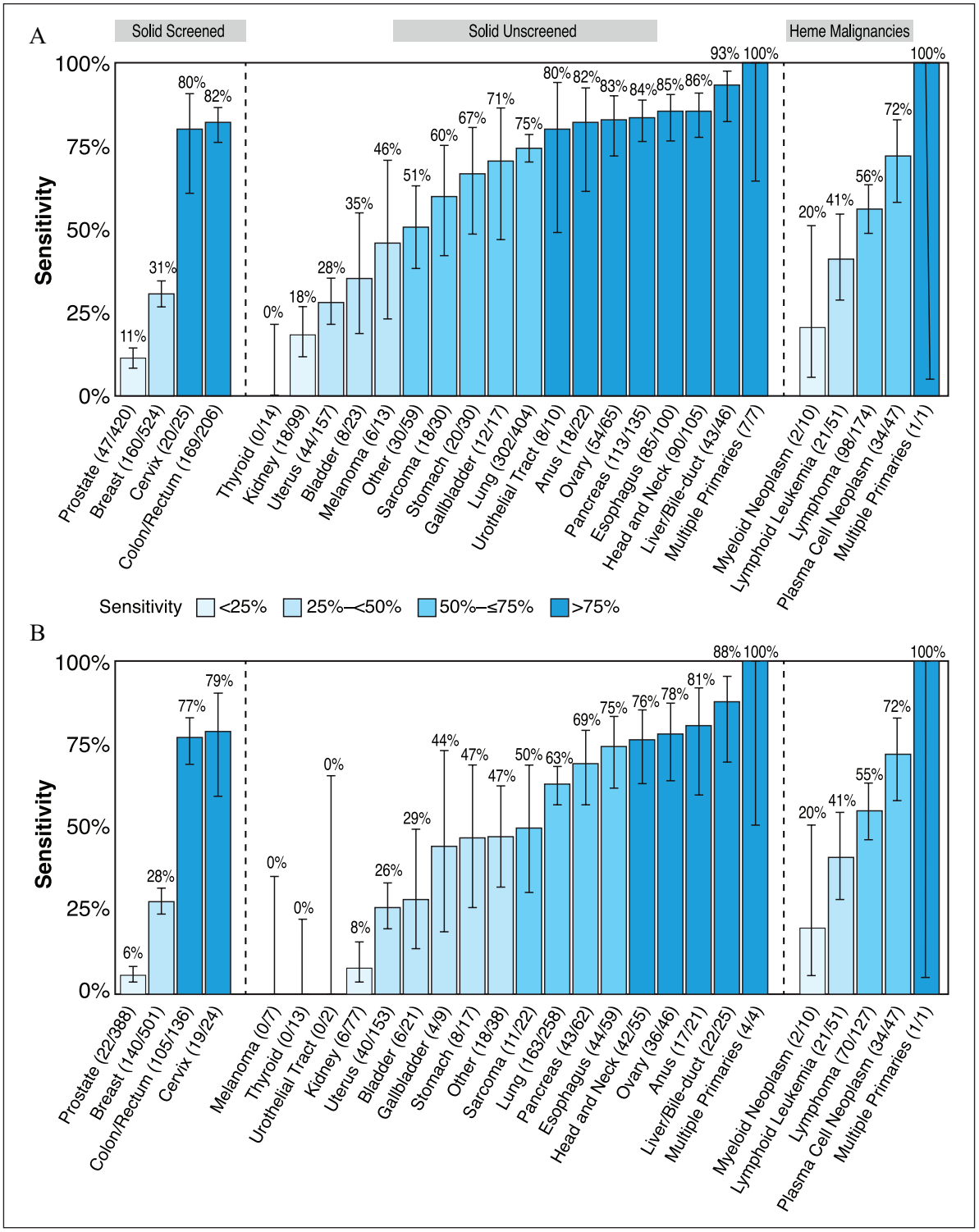

Figure 1. Sensitivity by cancer and subgroup. (A) Cancers across all stages within each subgroup are plotted against sensitivity. (B) Cancers across stages I–III within each subgroup are plotted against sensitivity. Cancers within each subgroup are plotted for solid screened tumors on the left, solid unscreened tumors in the middle, and hematologic malignancies on the right. The heme malignancy subgroup included cancers that were not expected to be assigned a stage. Note that urothelial tract cancers include renal, pelvis, ureter, and urethra cancers. "Other" cancers (n) included adrenal (1), ampulla of vater (1), brain (6), choriocarcinoma (1), mesothelioma (7), non-melanoma non-basal cell carcinoma/squamous cell carcinoma skin cancer (2), unspecified (10), penis (1), small intestine (13), testis (6), thymus (2), vagina (2), and vulva (7). Error bars represent 95% Wilson confidence intervals, and data values above the error bars indicate the sensitivity.

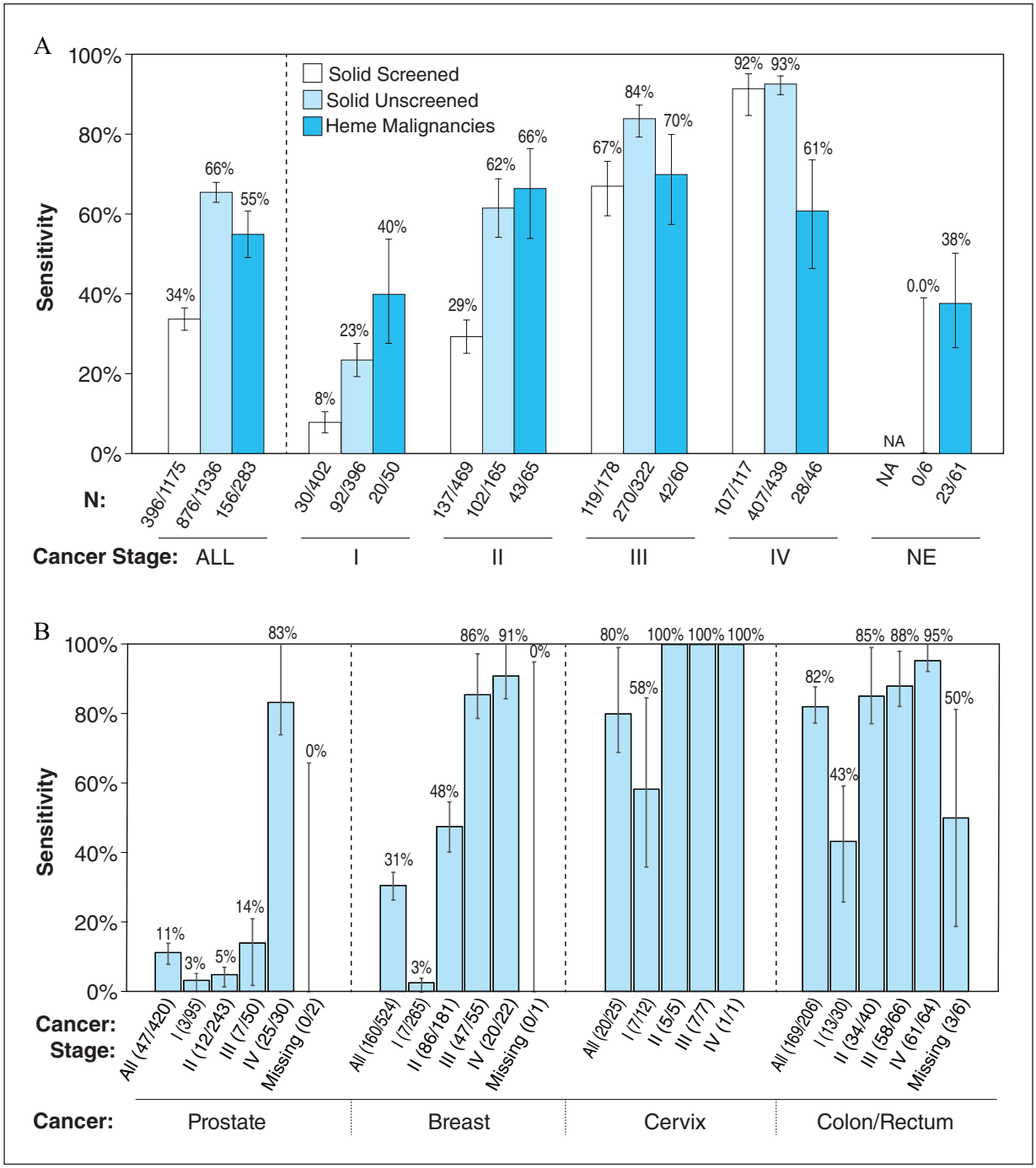

Figure 2. Sensitivity by cancer clinical stage and subgroup. (A) Cancer clinical stage for each subgroup is plotted against sensitivity. Stages I–IV are plotted individually to the right. (B) Sensitivity for solid screened tumors by cancer and clinical stage. Error bars represent 95% Wilson confidence intervals, and data values above the error bars indicate the sensitivity. Missing indicates unavailable stage information. NE, not expected to be staged.

screening paradigms for prostate and breast cancers can prompt overdiagnosis.[15,16] The lower sensitivity in early-stage prostate and breast cancers observed may be affected by cancers found through current screening that are less biologically active or have molecular features associated with less aggressive disease, and so less detectable by MCED testing. This possibility is consistent with the numerically higher early-stage sensitivity observed for solid screened tumors that were enrolled through clinical presentation versus screening. While sensitivity was lower for breast and prostate cancers, research has suggested that MCED testing at preventive care exams may complement existing breast and prostate cancer screening, given that the ease of a blood test may promote adherence.[17]

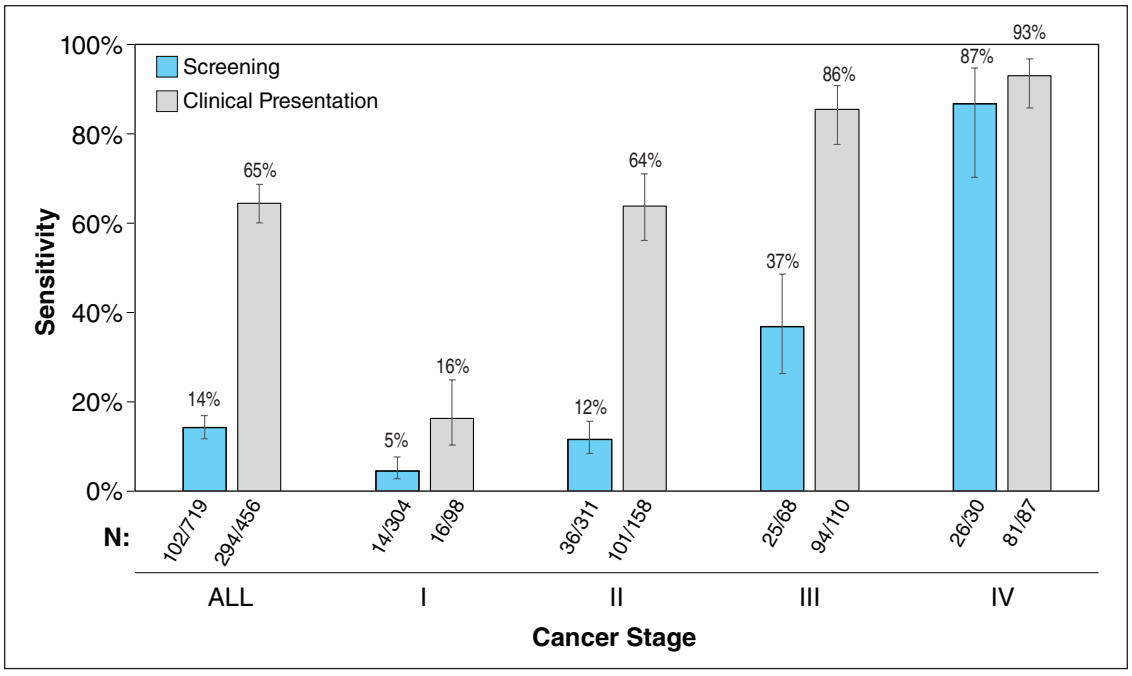

Figure 3. Solid screened tumors diagnosed through screening or clinical presentation. MCED test sensitivity by stage for solid screened tumor cases that were diagnosed through screening (light blue) or clinical presentation (gray). Error bars represent 95% Wilson confidence intervals, and data values above the error bars indicate the sensitivity.

Although sensitivity in the solid screened and unscreened subgroups increased with stage, sensitivity in the heme malignancy subgroup (excluding those not expected to be staged) was variable across stages. Notably, the hematologic malignancies examined in this study are heterogeneous with respect to aggressiveness,[18,19] which likely influences analyzable ctDNA; indeed, the fraction of ctDNA relative to cfDNA (circulating tumor fraction) is a major determinant of cancer signal detection and prognosis.[14,20] Higher tumor fractions are generally observed in late-stage cancers with higher tumor burden.[14] Additionally, the hematologic malignancies studied encompass a spectrum of disease severity, making it difficult to interpret the variable sensitivities. The range of sensitivities across hematologic malignancies warrants further study to confirm performance in these cancers.

The CCGA study reported here was a case-control study and was not designed to determine whether the MCED test can detect cancer cases missed by population screening in the intended use, all-risk population. This post-hoc analysis was limited by small sample sizes for individual cancers and was not powered to draw statistical conclusions. However, this analysis provides insights into the ability of MCED tests to complement current recommended screening. Specifically, the MCED test detected a shared cancer signal across many cancers that currently have no screening recommendations. These data support MCED testing as a potential promising tool to expand early cancer detection, and continued research is warranted to confirm the feasibility of test implementation and complementarity to existing population screening.

Acknowledgements

The authors would like to thank all the individuals who participated in the CCGA study, as well as all study staff. The authors would also like to thank Stephannie Shih, an employee of GRAIL, LLC, a subsidiary of Illumina, Inc., for significant contributions to the data analysis.

Medical writing support was provided by Alexis Fedorchak, Sarah Prins, and Grace Wang (GRAIL, LLC, a subsidiary of Illumina, Inc., Menlo Park, CA), editorial and graphics assistance was provided by Erin Spohr from ENGAGE Labs, LLC (Oak Ridge, NJ) and Kristi Whitfield, PhD, from PosterDocs (Oakland, CA), and funded by GRAIL, LLC, a subsidiary of Illumina, Inc. (Menlo Park, CA).

Author contributions

All the authors met the criteria for authorship set forth by the International Committee of Medical Journal Editors, and were involved in the conception, preparation and approval of the manuscript.

Data sharing

De-identified data and code to reproduce the figures in this paper are available upon request.

Declaration of conflicting interests

The author(s) declared the following potential conflicts of interest with respect to the research, authorship, and/or publication of this article: Spencer H Shao, Jessica Clement, Gina Chung, WH Wilson Tang, Habte Yimer, and Mohan Tummala have no conflicts to disclose. Brian Allen and Jingjing Gao were employees of GRAIL, LLC, a subsidiary of Illumina, Inc., with equity in Illumina, Inc. Earl Hubbell is an employee of GRAIL, LLC, a subsidiary of Illumina, Inc., with equity in Illumina, Inc. Minetta C Liu is an uncompensated consultant for GRAIL, LLC, a subsidiary of Illumina, Inc. Charles Swanton holds stock in Illumina, Inc, Epic Biosciences, Apogen Biotech; receives grants from Pfizer, AstraZeneca; receives honoraria or consultant fees from Roche Ventana, Celgene, Pfizer, Novartis, Genentech, Bristol Myers Squibb; and is a co-founder of Achilles Therapeutics.

Ethics approval

All participants provided written informed consent. The study was approved by an Institutional Review Board or independent ethics committee at each participating site and conducted per the International Conference on Harmonization for Good Clinical Practice guidelines and the Declaration of Helsinki.

Funding

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work was supported by GRAIL, LLC, a subsidiary of Illumina, Inc.

ORCID iDs

Spencer H. Shao https://orcid.org/0000-0002-8390-3824
Earl Hubbell https://orcid.org/0000-0001-7301-3759

Supplemental material

Supplemental material for this article is available online.

References

1. Siegel RL, Miller KD, Fuchs HE, et al. Cancer Statistics, 2021. *CA Cancer J Clin* 2021; 71: 7–33.
2. Siu AL and US Preventive Services Task Force. Screening for breast cancer: US Preventive Services Task Force Recommendation Statement. *Ann Intern Med* 2016; 164: 279–296.
3. Davidson KW, Barry MJ, Mangione CM, et al. Screening for colorectal cancer: US Preventive Services Task Force recommendation statement. *JAMA* 2021; 325: 1965–1977.
4. Grossman DC, Curry SJ, Owens DK, et al. Screening for prostate cancer: US Preventive Services Task Force recommendation statement. *Jama* 2018; 319: 1901–1913.
5. Curry SJ, Krist AH, Owens DK, et al. Screening for cervical cancer: US Preventive Services Task Force recommendation statement. *Jama* 2018; 320: 674–686.
6. US Preventive Services Task Force, Krist AH, Davidson KW, et al. Screening for lung cancer: US Preventive Services Task Force Recommendation Statement. *JAMA* 2021; 325: 962.
7. Richards TB, Soman A, Thomas CC, et al. Screening for lung cancer—10 states, 2017. *Morb Mortal Wkly Rep* 2020; 69: 201.
8. Clarke CA, Hubbell E and Ofman JJ. Multi-cancer early detection: A new paradigm for reducing cancer-specific and all-cause mortality. *Cancer Cell* 2021; 39: 447–448.
9. Klein E, 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: 1167–1177.
10. Liu MC, Oxnard GR, Klein EA, et al. Sensitive and specific multi-cancer detection and localization using methylation signatures in cell-free DNA. *Ann Oncol* 2020; 31: 745–759.
11. Kim J, Lee S, Bae S, et al. Comparison between screen-detected and symptomatic breast cancers according to molecular subtypes. *Breast Cancer Res Treat* 2012; 131: 527–540.
12. American Cancer Society. Cancer Facts & Figures (2021), https://www.cancer.org/content/dam/cancer-org/research/cancer-facts-and-statistics/annual-cancer-facts-and-figures/2021/cancer-facts-and-figures-2021.pdf. (accessed 10 March 2021).
13. Braunstein GD and Ofman JJ. Criteria for evaluating multi-cancer early detection tests. *touchREVIEWS Oncol Haematol* 2021; 17: 3–6.
14. Chen X, Dong Z, Hubbell E, et al. Prognostic significance of blood-based multi-cancer detection in plasma cell-free DNA. *Clin Cancer Res* 2021; 27: 4221-4229.
15. Fenton JJ, Weyrich MS, Durbin S, et al. Prostate-specific antigen–based screening for prostate cancer: evidence report and systematic review for the US Preventive Services Task Force. *Jama* 2018; 319: 1914–1931.
16. Nelson HD, O'Meara ES, Kerlikowske K, et al. Factors associated with rates of false-positive and false-negative results from digital mammography screening: an analysis of registry data. *Ann Intern Med* 2016; 164: 226–235.
17. Hathaway C, Paetsch P, Li Y, et al. Association of breast cancer screening behaviors with stage at breast cancer diagnosis and potential for additive multi-cancer detection via liquid biopsy screening: a claims-based study. *Front Oncol* 2021; 11: 688455.
18. Arber DA, Orazi A, Hasserjian R, et al. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. *Blood* 2016; 127: 2391–2405.
19. Swerdlow SH, Campo E, Pileri SA, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. *Blood J Am Soc Hematol* 2016; 127: 2375–2390.
20. Bredno J, Lipson J, Venn O, et al. Clinical correlates of circulating cell-free DNA tumor fraction. *PLOS ONE* 2021; 16: e0256436.