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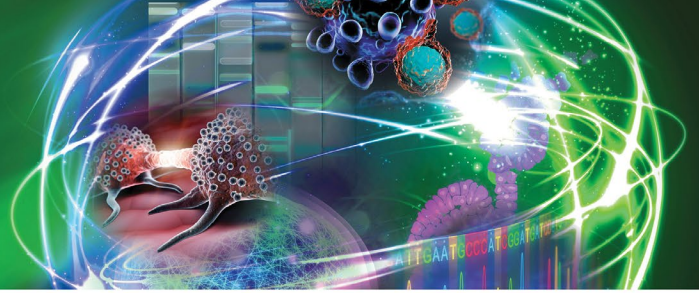

Clinical Validation of a Targeted Methylation-Based Multi-Cancer Early Detection Test

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Disclosure Information

Eric A. Klein

I have the following financial relationships to disclose:

Consultant for: GRAIL, Inc

Speaker's Bureau for: none

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Stockholder in: none

Employee of: Cleveland Clinic

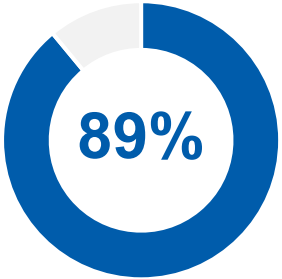
-and-

I will discuss the following investigational use in my presentation:
multi-cancer early detection test (Galleri™)

Benefits of Early Cancer Diagnosis

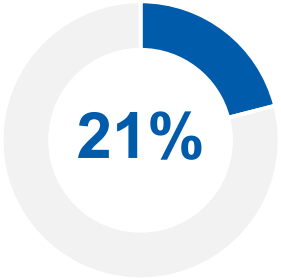
5-year cancer-specific survival (all cancers)[1]

Localized



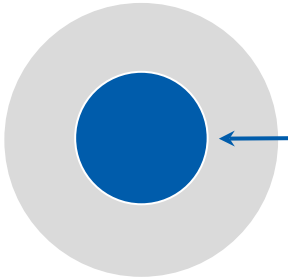
*invasive localized tumors that have **not spread** beyond organ of origin*

Distant Metastases



*invasive cancers that have **metastasized** beyond the organ of origin to other parts of the body*

Stage IV cancer diagnosis[2]



only 18% of diagnoses lead to

45% of cancer-related deaths (within 5 years of diagnosis)

Source: SEER data

MCED, multi-cancer early detection; SEER, Surveillance, Epidemiology, and End Results.

[1]Surveillance, Epidemiology, and End Results (SEER) Program (www.seer.cancer.gov) SEER*Stat Database: Incidence - SEER 18 Regs Research Data, Nov 2018 Sub. Includes persons aged 50-79 diagnosed. Noone AM, Howlader N, Krapcho M, et al. (eds). SEER Cancer Statistics Review, 1975-2015, National Cancer Institute, Bethesda, MD, http://seer.cancer.gov/csr/1975_2015/, based on November 2017 SEER data submission, posted to the SEER website April 2018.

[2]Clarke CA, et al. *Cancer Epidemiol Biomarkers Prev*. 2020;29(5):895-902. DOI: 10.1158/1055-9965.EPI-19-1366.

A Multi-Cancer Early Detection (MCED) Test May Decrease Cancer-Related Mortality

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MCED test as a complement to existing recommended screening

Detect many lethal cancers, including unscreened cancers, using single blood sample collected at point-of-care

Identify signal origin to direct diagnostic workup

High specificity (~99%) could limit false positives and unnecessary workups

Potential for:

- More cancer cases detected through screening
- Improved patient outcomes and survival
- Decreased burden of care

Modeling the Potential Impact of GRAIL MCED Test Added to Usual Care

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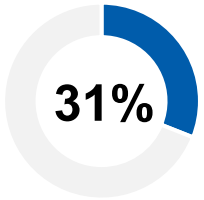
Diagnostic yield[1]



cancers intercepted by a MCED test

Tumor stage shift[1]

-50%

reduction in late-stage (III/IV) cancer incidence

(-78% in intercepted cases)

Mortality reductions[1]

-21%

reduction in 5-year mortality

(-40% in intercepted cases)

Modeling performance of a GRAIL MCED test in a representative population suggests it could reduce overall cancer mortality if added to usual care[a,1]

[a]Based on annual cancer incidence (1187/100,000) in elevated risk population aged 50-79 y (data from US Surveillance, Epidemiology, and End Results [SEER] program). MCED results based upon fast tumor growth model using test version described by Liu et al.[2] Usual care represents real-world cancer diagnostic processes (e.g., screening, incidental detection, symptomatic workup) as captured by SEER, which includes imperfect adherence and limited access to healthcare. MCED, multi-cancer early detection.

[1]Hubbell E, et al. *Cancer Epidemiol Biomarkers Prev*. December 16 2020. DOI: 10.1158/1055-9965.EPI-20-1134. [2]Liu MC, et al. *Ann Oncol*. 2020;31(6):745-759. DOI: 10.1016/j.annonc.2020.02.011.

The Circulating Cell-Free Genome Atlas Study (CCGA Study; NCT02889978)

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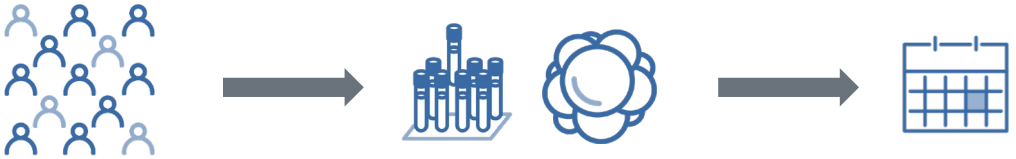
Prospective, multicenter, case-control, observational study

Study Goals

- Develop and validate a blood-based MCED test analyzing plasma cell-free DNA (cfDNA)
- Detect cancer signals across multiple cancer types & simultaneously predict their signal origin

Study Design



15,254 participants
with/without cancer

Fully Enrolled (142 sites)

Blood samples
all participants

Tissue samples
cancer only

Follow-up for 5 years
(vital status, cancer status)

cfDNA, cell-free DNA; MCED, multi-cancer early detection test.

CCGA: Divided Into 3 Pre-Specified Substudies

Discovery[1,2]

- Methylation Patterns
- Mutations
- Chromosome Alterations

Whole Genome Methylation
(identified as method for further development)

Training: n=1785
Validation: n=1015

Assay Refinement and Classifier Development[3]

- Methylation Patterns
- Machine-learning classifier to differentiate cancer vs non-cancer and predict signal origin

Targeted Methylation
(identified key informative methylation regions; training and validation of the selected and updated targeted methylation classifier)

Training: n=3133
Validation: n=1354

Further Assay Refinement and Clinical Validation

- Methylation Patterns
- **Further refined for use** as screening tool
- **Clinical validation** supporting product launch

n=5309 participants
(cancer=3237; non-cancer=2069)

[1]Klein E, et al. *J Clin Oncol*. 2018;36(15) suppl. DOI: 10.1200/JCO.2018.36.15_suppl.12021.
[2]Razavi P, et al. Poster presented at: American Society for Clinical Oncology 2017 Annual Meeting; June 2-7, 2017; Chicago, IL. Abstract 11526. [3]Liu MC, et al. *Ann Oncol*. 2020;31(6):745-759. DOI: 10.1016/j.annonc.2020.02.011.

CCGA: Substudy 3

Key objectives were to evaluate test performance:

Primary	• for cancer signal detection (specificity, overall sensitivity, sensitivity by cancer class, sensitivity by clinical stage) • for signal origin prediction (accuracy)
(Key) Secondary	• for cancer signal detection in a pre-specified group of 12 cancers responsible for two-thirds of cancer deaths[a,1] • in cancers with and without common screening options • by age group

Methods

- cfDNA from evaluable samples was analyzed using a targeted methylation bisulfite sequencing assay and machine learning techniques
- The classifier was trained to target a specificity of 99.4% and locked before analysis

CCGA 3

Further Assay Refinement and Clinical Validation

Methylation Patterns

Further refined for use as screening tool

Clinical validation supporting product launch

MCED test results for the n=4077 confirmed status analysis set (cancer=2823; non-cancer=1254)

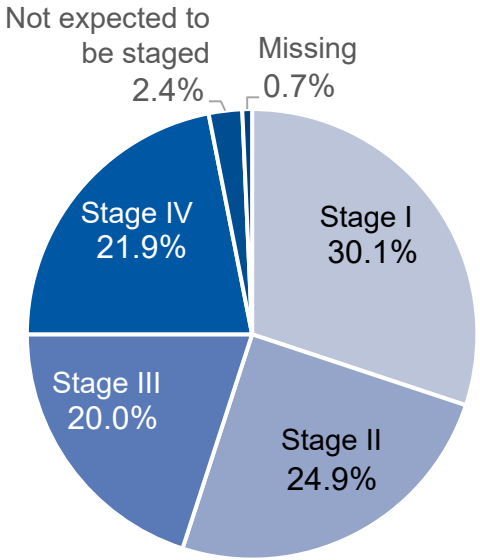
[a]Anus, bladder, colon/rectum, esophagus, head and neck, liver/bile duct, lung, lymphoma, ovary, pancreas, plasma cell neoplasm, and stomach.
[1]American Cancer Society. Cancer Facts & Figures 2021. Atlanta: American Cancer Society; 2021. https://www.cancer.org/content/dam/cancer-org/research/cancer-facts-and-statistics/annual-cancer-facts-and-figures/2021/cancer-facts-and-figures-2021.pdf

Demographics and Baseline Characteristics

Demographics		Cancer (n=2823)	Non-cancer (n=1254)	Total (N=4077)
Age	Mean (SD)	**62.6** (11.8)	**56.2** (12.6)	60.6 (12.4)
	≥50, n (%)	2438 (**86.4**)	850 (**67.8**)	3288 (80.6)
Sex, n (%)	Female	1394 (**49.4**)	864 (**68.9**)	2258 (55.4)
Race/ Ethnicity, n (%)	White[a]	2316 (82.0)	996 (79.4)	3312 (81.2)
	Black[a]	193 (6.8)	85 (6.8)	278 (6.8)
	Hispanic	192 (6.8)	103 (8.2)	295 (7.2)
	Other[b]	122 (4.3)	70 (5.6)	192 (4.7)
Smoking Status, n (%)[c]	Never	1322 (46.8)	719 (57.3)	2041 (50.1)
	Former	1080 (**38.3**)	402 (**32.1**)	1482 (36.4)
	Current	378 (**13.4**)	126 (**10.0**)	504 (12.4)
Family Cancer History, n (%)[d]	Yes	2254 (79.8)	1014 (80.9)	3268 (80.2)
	No	493 (17.5)	210 (16.7)	703 (17.2)
BMI, n (%)[e]	Under/Normal	785 (27.8)	330 (26.3)	1115 (27.3)
	Overweight	908 (32.2)	376 (30.0)	1284 (31.5)
	Obese	1124 (39.8)	543 (43.3)	1667 (40.9)

BMI, body mass index.
[a]Non-Hispanic. [b]Other includes Asian, Native Hawaiian or Pacific Islander; American Indian or Alaska Native; or Other. [c]Smoking status was missing for 50 participants.
[d]Cancer history was not known in 106 participants. [e]BMI category was missing for 11 participants.

Cancer Clinical Stage Distribution



Cancer Signal Detection: Specificity, Overall Sensitivity, and Signal Origin Prediction

	Cancer (n=2823)	Non-cancer (n=1254)	Total (n=4077)
Test Positive	1453	6	1459
Test Negative	1370	1248	2618

Specificity:
99.5%
(95% CI: 99.0–99.8%)

↓

0.5%
false-positive rate

Sensitivity:
51.5%
(95% CI: 49.6–53.3%)

↓

Signal origin prediction accuracy[a]: 88.7%
(95% CI: 87.0–90.2%)

[a]Accuracy was defined as the number of correctly predicted top 1 signal origin (n=1273) divided by all cancer participants with a positive cancer signal, excluding participants with unknown primary (n=1435). CI, two-sided 95% Wilson confidence intervals.

Sensitivity of Cancer Signal Detection by Clinical Stage

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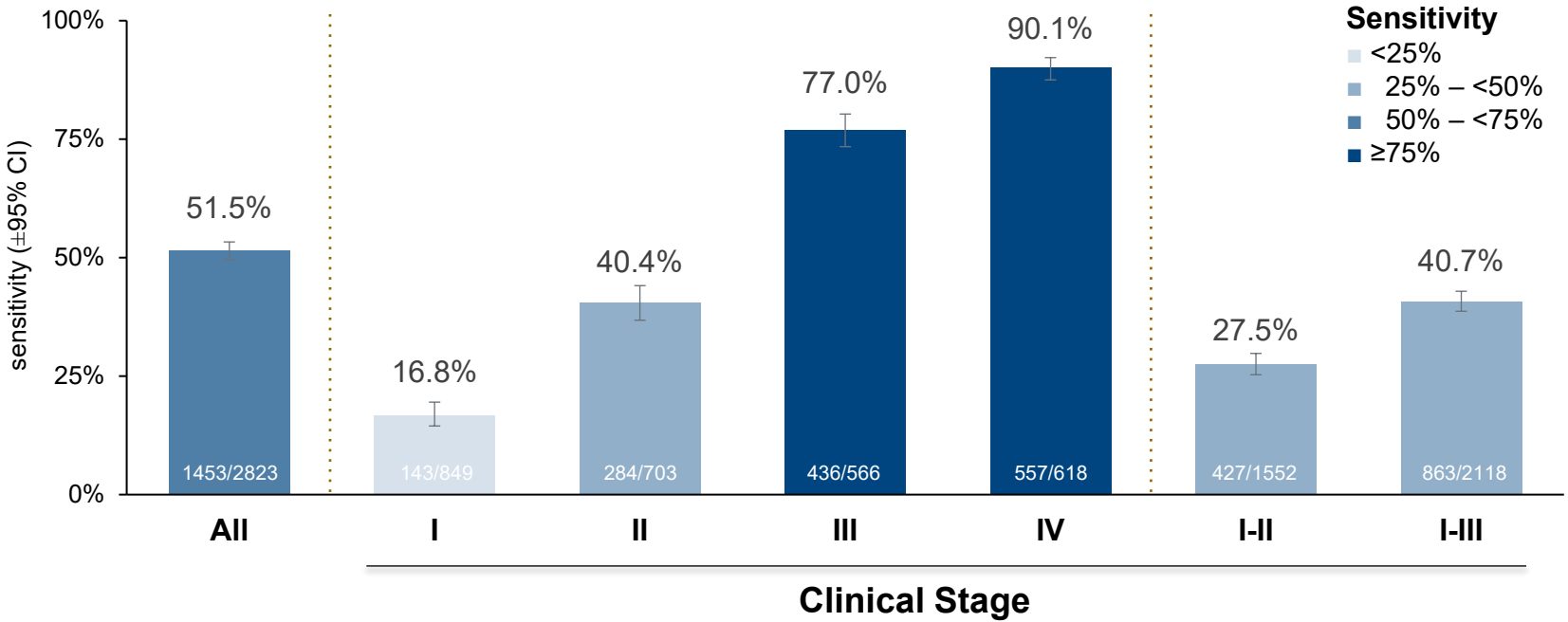
Sensitivity increased with increasing clinical stage



"All" comprises all cancer stages, including missing stage and cancer classes that do not have staging per American Joint Committee on Cancer (AJCC) staging manual.
CI, confidence interval.

Sensitivity of Cancer Signal Detection in 12 Pre-Specified Cancers Responsible for Two-Thirds of Cancer Deaths

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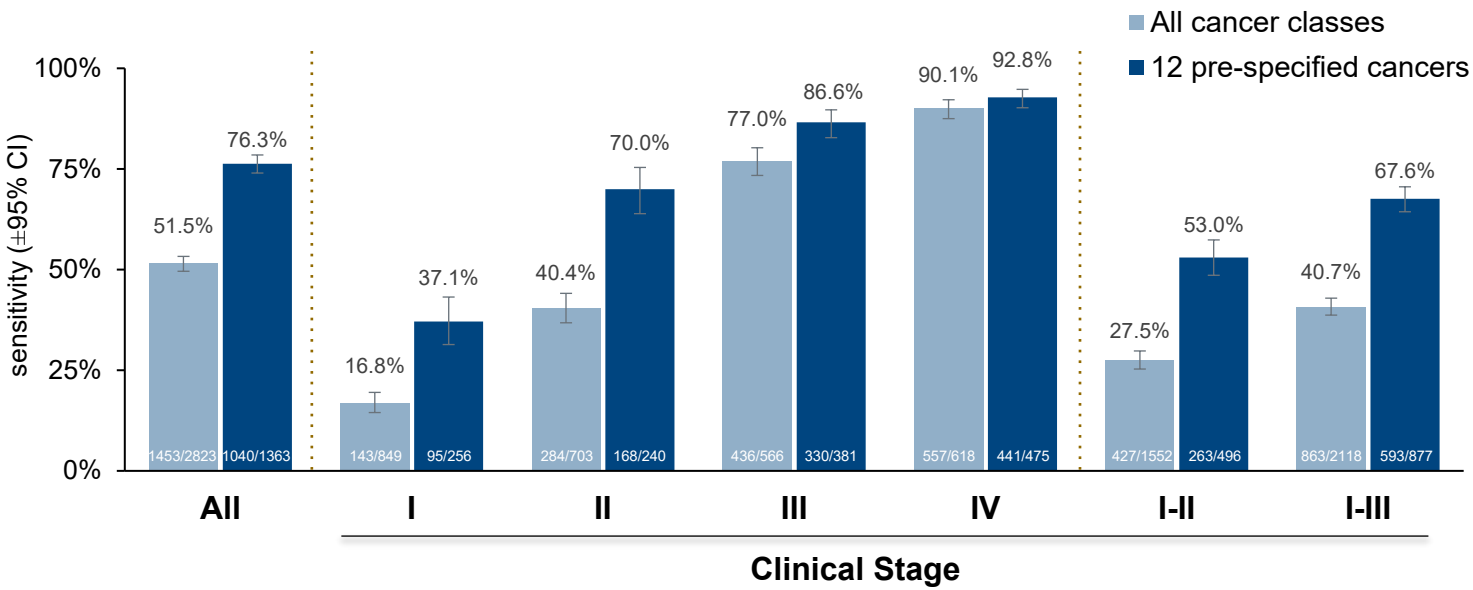
Sensitivity was higher in these 12 cancers vs overall, particularly in early-stage



12 cancers that account for 62% of US cancer deaths[1]

- Anus
- Bladder
- Colon/rectum
- Esophagus
- Head and neck
- Liver/bile duct
- Lung
- Lymphoma
- Ovary
- Pancreas
- Plasma cell neoplasm
- Stomach

"All" comprises all cancer stages, including missing stage and cancer classes that do not have staging per American Joint Committee on Cancer (AJCC) staging manual.
CI, confidence interval.
[1]American Cancer Society. Cancer Facts & Figures 2021. Atlanta: American Cancer Society; 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

Sensitivity of Cancer Signal Detection by Cancer Class

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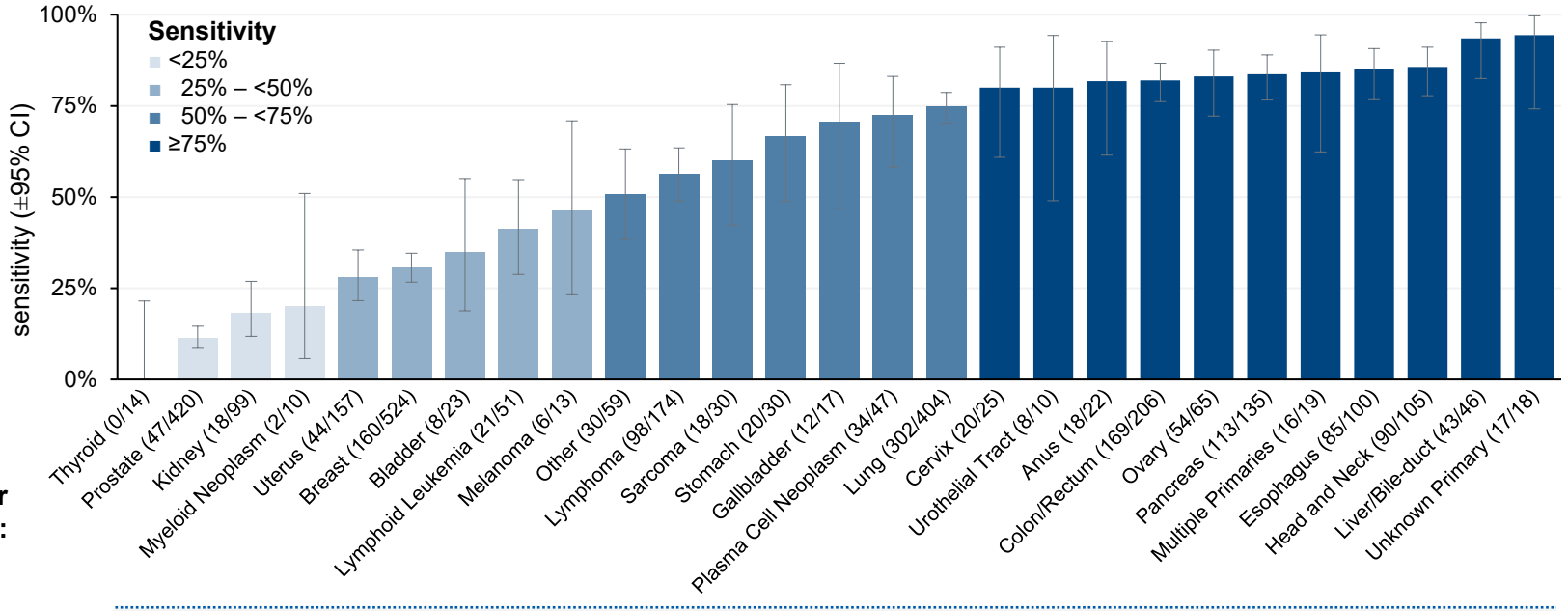

Cancer class:

AJCC cancer type[1]:

Cancer signals were detected across >50 AJCC cancer types, with a test positive rate across AJCC cancer types of **51.0%** (95% CI: 49.1%–52.8%)

AJCC, American Joint Committee on Cancer; CI, confidence interval.

[1]Amin MB, et al. *CA Cancer J Clin*. 2017;67(2):93-99. DOI: 10.3322/caac.21388.

Sensitivity of Cancer Signal Detection in Cancers With and Without Common Screening

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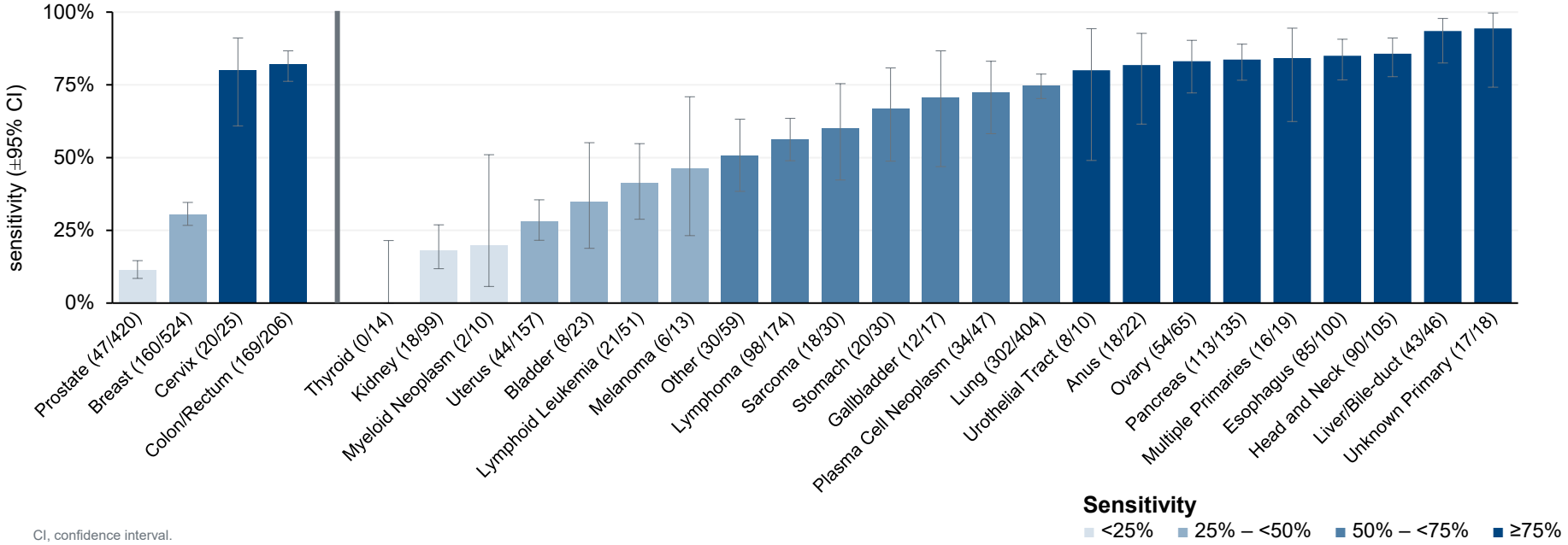
With Common Screening Options: 33.7%
(95% CI: 31.1–36.5%)

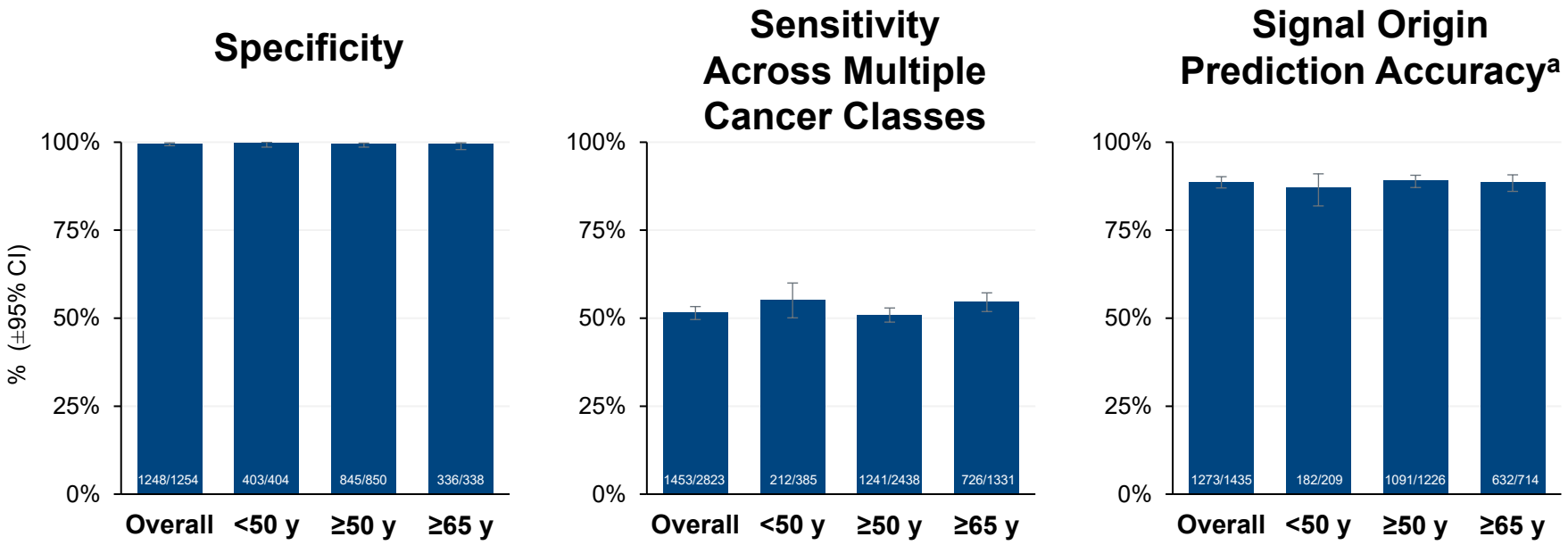
Without Common Screening Options: 63.8%
(95% CI: 61.4–66.1%)



CI, confidence interval.

Specificity, Sensitivity, and Signal Origin Prediction by Age Group

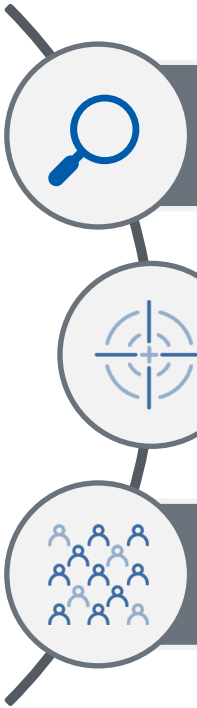
Cancer signal detection was similar across age groups



[a]Accuracy was defined as the number of correctly predicted top 1 signal origin divided by all cancer participants with a positive cancer signal, excluding participants with unknown primary.
CI, confidence interval.

Conclusions

In this pre-specified, large-scale, clinical validation sub-study 3 of CCGA:



- This MCED test detected cancer signals across >50 AJCC cancer types, which is critical to maximize the number of cancer cases detected in a population
- This MCED test performed with high specificity and high accuracy of signal origin prediction, which could help direct the follow-up diagnostic work-up
- These data support the foundation for population-scale clinical implementation of this test