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Prospective evaluation of cell-free DNA fragmentomes for lung cancer detection

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Disclosure Information

Founder and Officer of Delfi Diagnostics

Founder of Personal Genome Diagnostics (acquired by LabCorp in 2022)

Scientific Advisory Board of Epitope and Viron Therapeutics.

Dr. Velculescu is an inventor on patent applications submitted by Johns Hopkins University related to cancer genomic analyses and cell-free DNA for cancer detection that have been licensed to one or more entities, including Delfi Diagnostics, LabCorp, Qiagen, Sysmex, Agios, Genzyme, Esoterix, Ventana and ManaT Bio. Under the terms of these license agreements, the University and inventors are entitled to fees and royalty distributions. These arrangements have been reviewed and approved by the Johns Hopkins University in accordance with its conflict-of-interest policies.

Non-FDA Approved Use of Drugs or Products Referenced in this Presentation - NONE

One of the greatest achievements in public health in our lifetimes will be through early cancer detection

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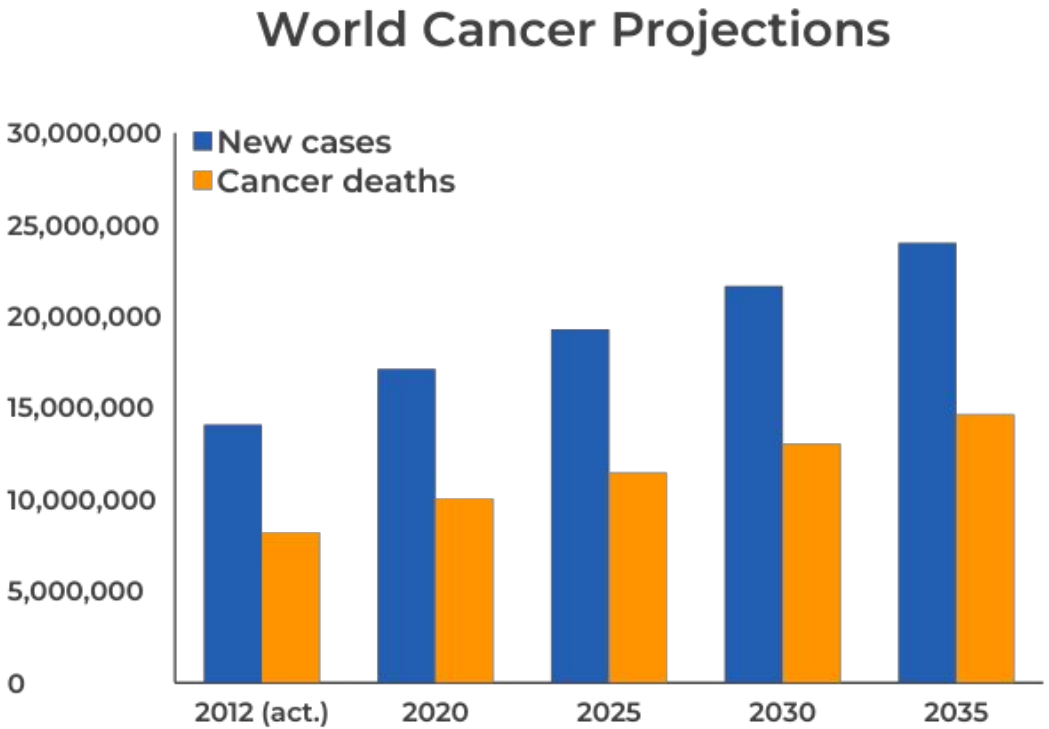

Cancer rates continue to rise world-wide

Largest potential strides against cancer are in early detection

For screening to work it must find early-stage disease as this has greatest impact on mortality

For screening to work it has to be a public health effort that is widely accessible at population scale price

1. Global Cancer Observatory. https://gco.iarc.fr/tomorrow/en/dataviz/isotype. 2. NCI SEER Cancer Stat Facts. https://seer.cancer.gov/statfacts/.

Mutations in cfDNA provide an avenue for noninvasive cancer detection

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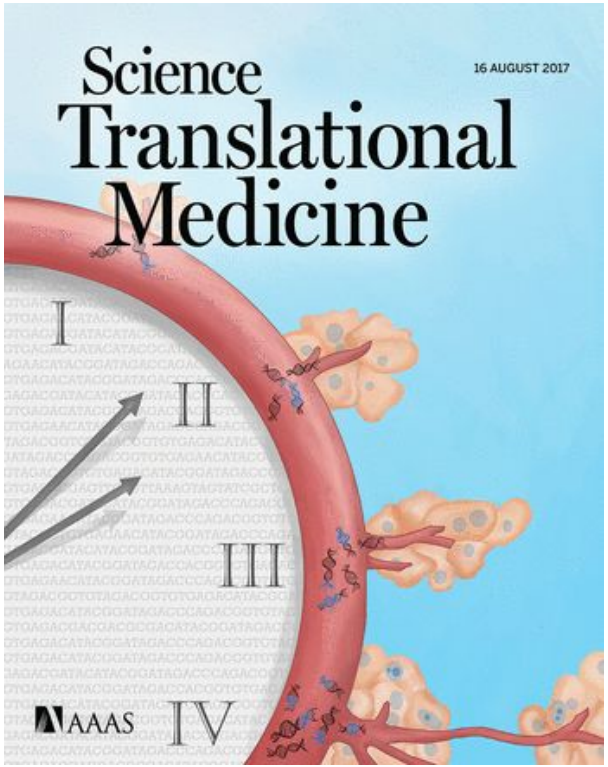


SCIENCE TRANSLATIONAL MEDICINE | RESEARCH ARTICLE

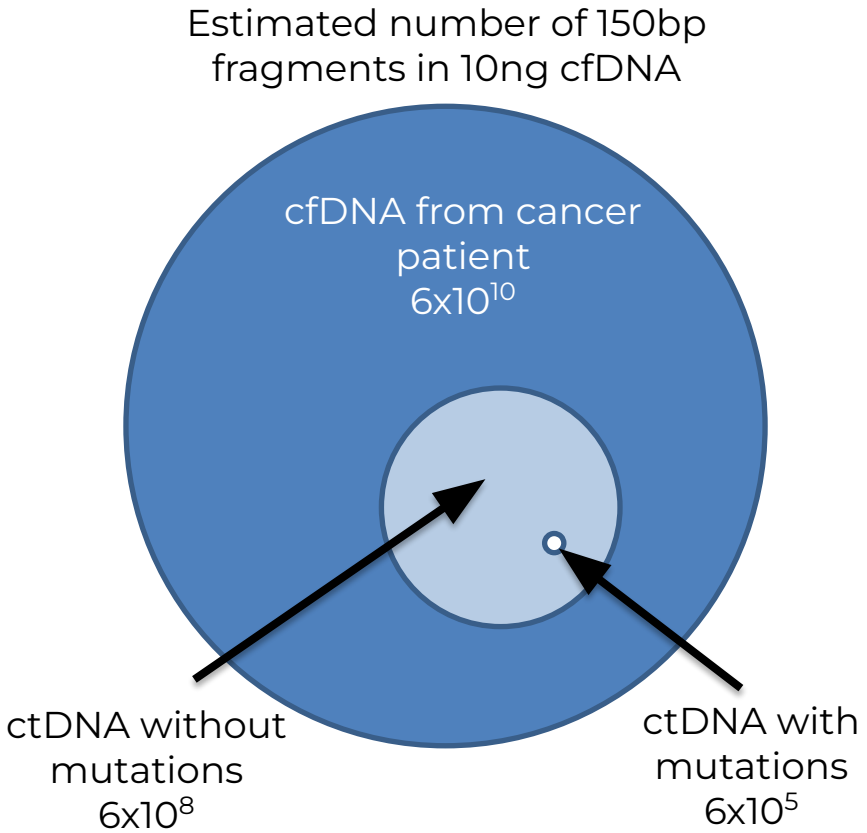
CANCER

Direct detection of early-stage cancers using circulating tumor DNA

Jillian Phallen,[1] Mark Sausen,[2] Vilmos Adleff,[1] Alessandro Leal,[1] Carolyn Hruban,[1] James White,[1] Valsamo Anagnostou,[1] Jacob Fiksel,[1] Stephen Cristiano,[1] Eniko Papp,[1]* Savannah Speir,[1] Thomas Reinert,[3] Mai-Britt Worm Orntoft,[3] Brian D. Woodward,[4] Derek Murphy,[2] Sonya Parpart-Li,[2] David Riley,[2] Monica Nesselbush,[2] Naomi Sengamalay,[2] Andrew Georgiadis,[2] Qing Kay Li,[1] Mogens Rørbæk Madsen,[5] Frank Viborg Mortensen,[6] Joost Huiskens,[7,8] Cornelis Punt,[8] Nicole van Grieken,[9] Remond Fijneman,[10] Gerrit Meijer,[10] Hatim Husain,[4] Robert B. Scharpf,[1] Luis A. Diaz Jr.,[1†] Siân Jones,[2] Sam Angiuoli,[2] Torben Ørntoft,[3] Hans Jørgen Nielsen,[11] Claus Lindbjerg Andersen,[3] Victor E. Velculescu[1‡]

First systematic analysis of sequence alterations in cfDNA for direct detection of early-stage cancers

But targeted cfDNA changes have limited signal and may not be suitable for population screening



Approaches using targeted detection of mutations or methylation only detect a tiny fraction of ctDNA

Miss many cancers, especially early stage

cfDNA mutations occur in normal blood cells, confounding analyses

Deep sequencing makes these methods too expensive for broad screening

Moving beyond mutations and methylation in cfDNA

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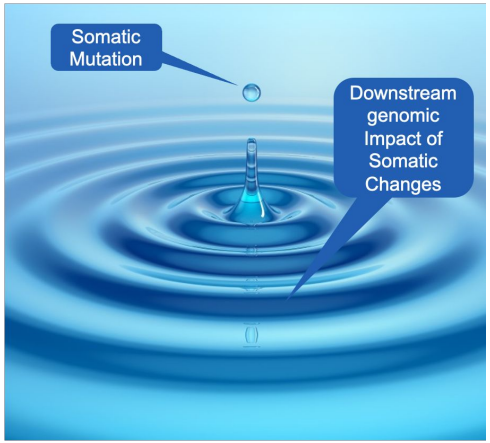

nature
International weekly journal of science

Genome-wide cell-free DNA fragmentation in patients with cancer

Stephen Cristiano[1,2,15], Alessandro Leal[1,15], Jillian Phallen[1,15], Jacob Fiksel[1,2,15], Vilmos Adleff[1], Daniel C. Bruhm[1], Sarah Østrup Jensen[3], Jamie E. Medina[1], Carolyn Hruban[1], James R. White[1], Doreen N. Palsgrove[1], Noushin Niknafs[1], Valsamo Anagnostou[1], Patrick Forde[1], Jarushka Naidoo[1], Kristen Marrone[1], Julie Brahmer[1], Brian D. Woodward[4], Hatim Husain[4], Karlijn L. van Rooijen[5], Mai-Britt Worm Ørntoft[3], Anders Husted Madsen[6], Cornelis J. H. van de Velde[7], Marcel Verheij[8], Annemieke Cats[9], Cornelis J. A. Punt[10], Geraldine R. Vink[5], Nicole C. T. van Grieken[11], Miriam Koopman[5], Remond J. A. Fijneman[12], Julia S. Johansen[13], Hans Jørgen Nielsen[14], Gerrit A. Meijer[12], Claus Lindbjerg Andersen[3], Robert B. Scharpf[1,2]* & Victor E. Velculescu[1]*

DELFI: DNA evaluation of fragments for early interception

Collaborative and interdisciplinary effort involving cancer genomics, biostatistics, computational biology, oncology, pathology from JHU School of Medicine and School of Public Health as well as national and international collaborators

Cristiano S, et al. *Nature*. 2019;570(7761):385-389.

DELFI Approach for Detection of ctDNA using Genome-Wide Fragmentation Profiles

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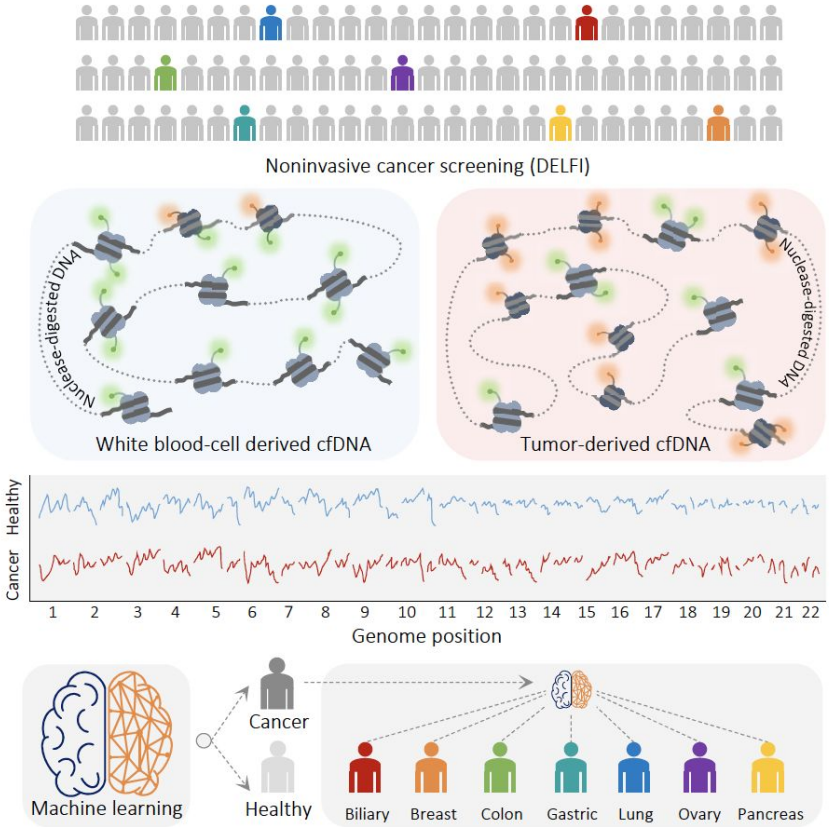

Cristiano S, et al. *Nature*. 2019;570(7761):385-389.

cfDNA Fragmentome: The genome-wide compendium of cfDNA fragments in the circulation, providing an integrated view of the chromatin, genome, and transcriptome states of normal and cancer cells of an individual

Multi-Cancer Early Detection: DELFI Is Highly Sensitive and Specific for Detection of Cancer and Tissue of Origin

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Cancer Type	Individuals analyzed	Sensitivity: 95% specificity	Sensitivity: 98% specificity		Tissue of Origin: Individuals analyzed	Tissue of Origin: Top 2 Predictions
Lung	12	100%	100%		30	77%
Ovarian	28	89%	89%		27	59%
Bile duct	26	88%	81%		23	65%
Gastric	27	81%	81%		24	79%
Colorectal	27	81%	80%		24	79%
Pancreatic	34	71%	65%		24	67%
Breast	54	70%	57%		42	91%
Healthy	215	—	—	Total	194	75%

Cristiano S, et al. *Nature*. 2019;570(7761):385-389.

DELFI Detects cfDNA Fragmentomes Altered in Lung Cancer and Is Not Fooled by Benign Lesions

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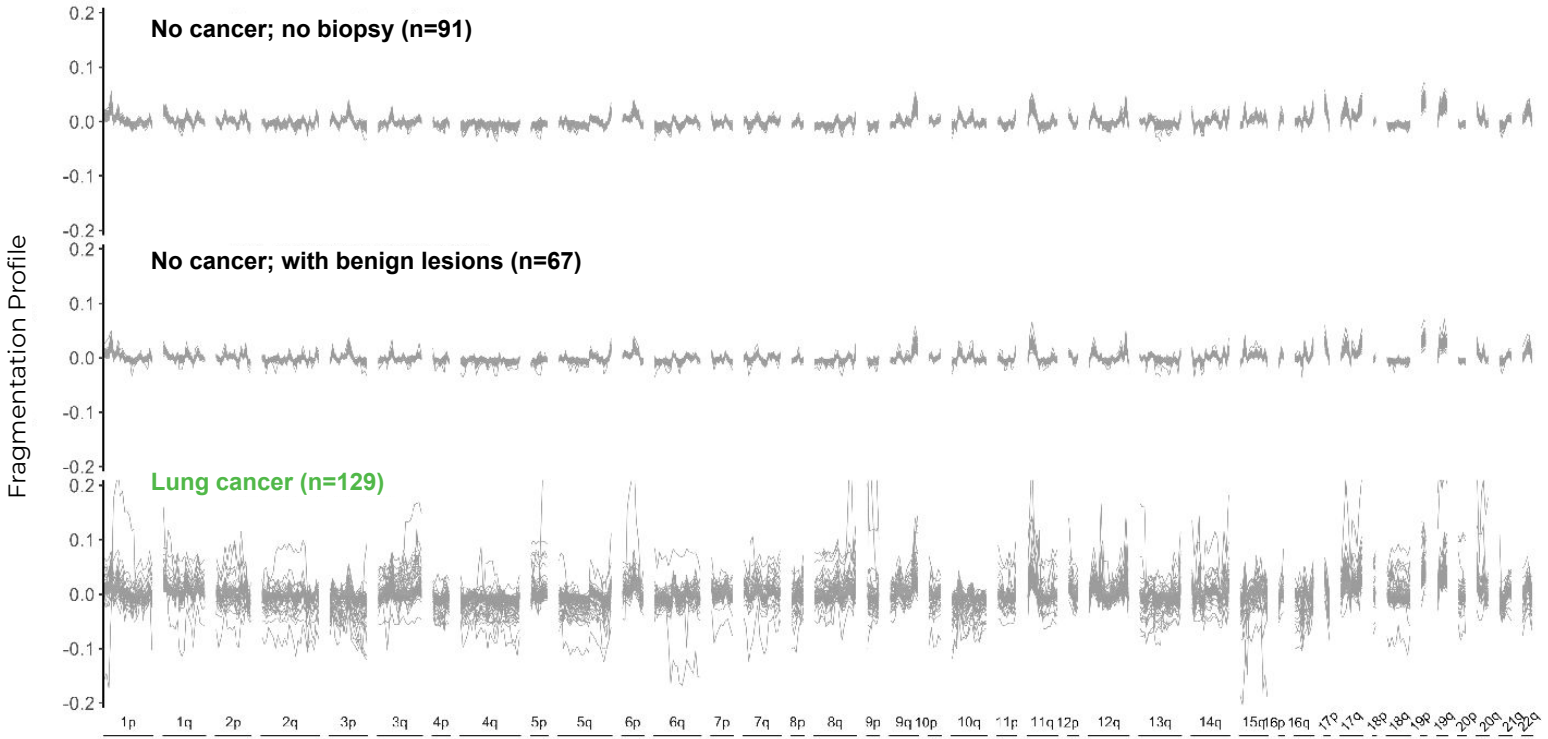
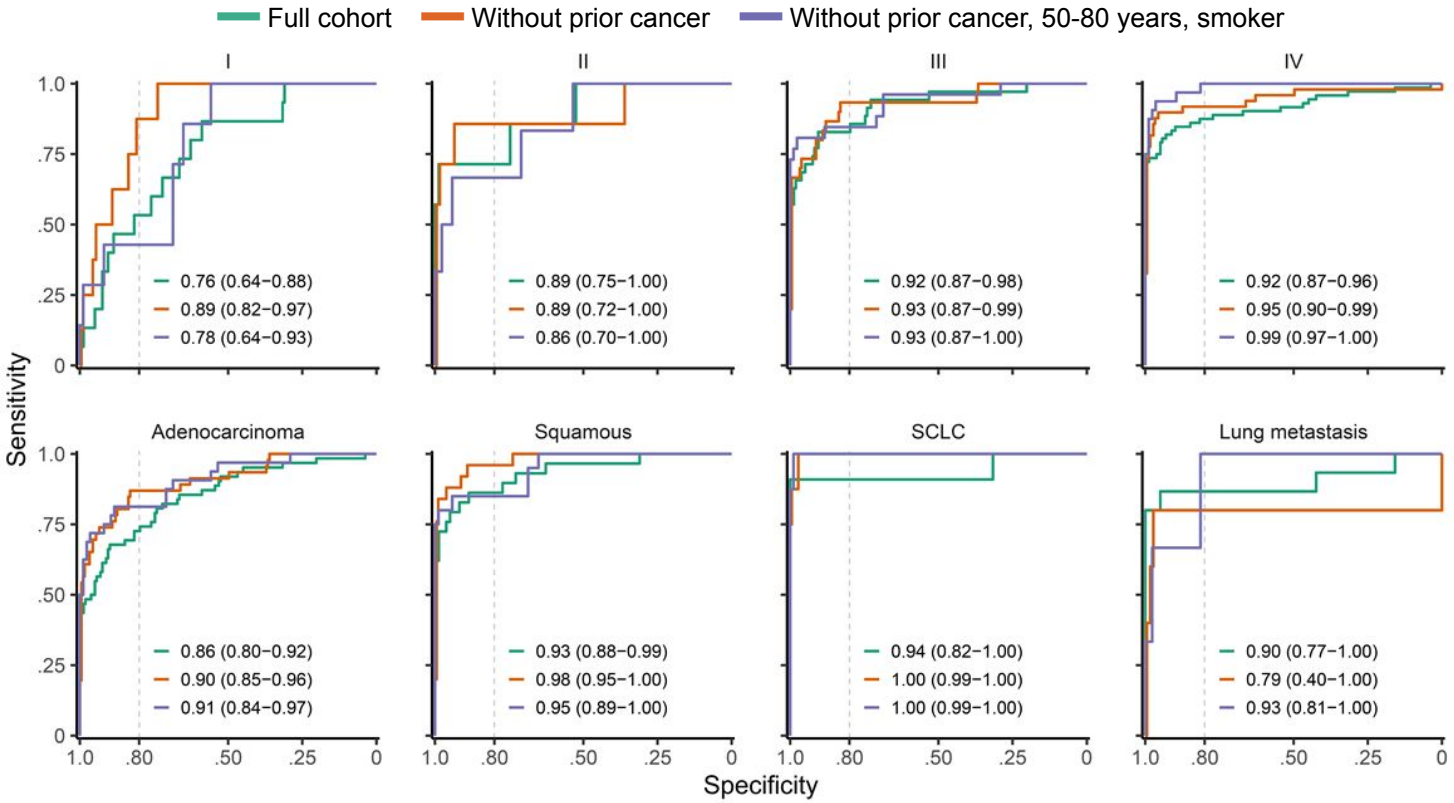


Mathios D, et al. *Nature Communications*, 2021;12(1):5060.

DELFI has high sensitivity and specificity for detection of lung cancer in prospective cohort



Mathios D, et al. *Nature Communications*, 2021;12(1):5060.

DELFI Detects cfDNA Fragmentomes Altered in Liver Cancer and Is Not Affected by Liver Disease

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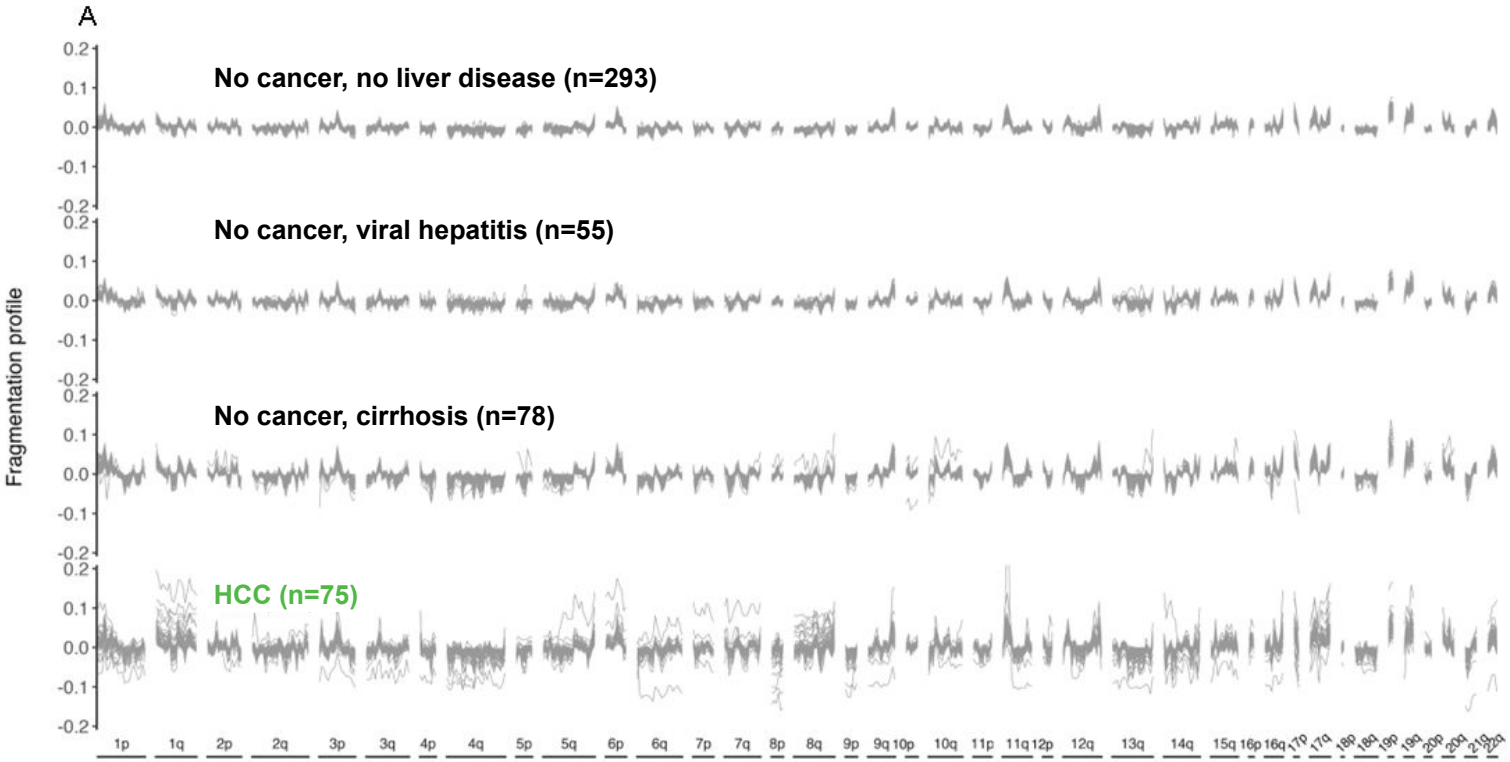

Foda, Annapragada, Kim, et al., *Cancer Discovery*, 2022:CD-22-0659.

DELFI has high sensitivity and specificity for detection of liver cancer

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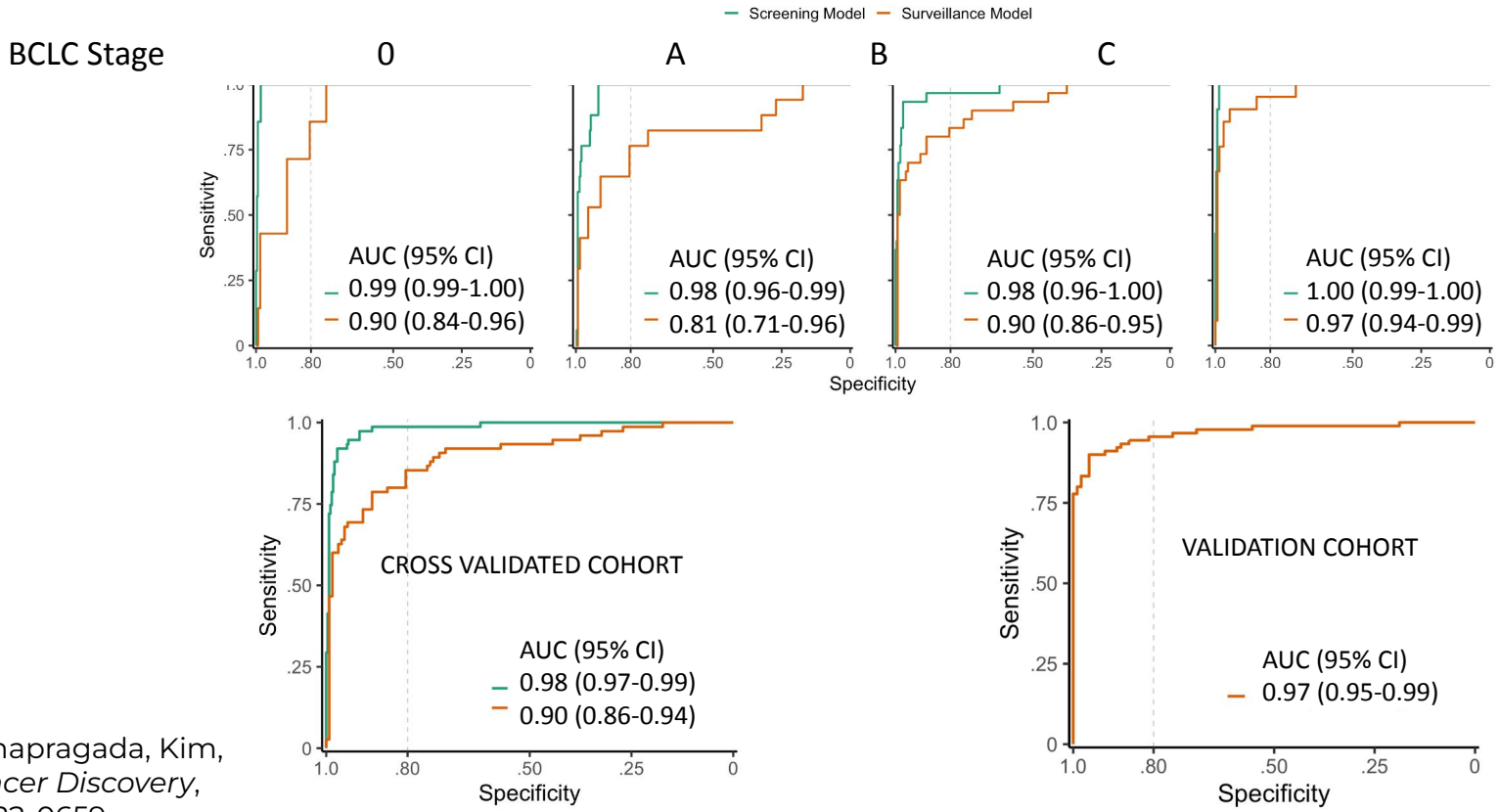

Foda, Annapragada, Kim, et al., *Cancer Discovery*, 2022:CD-22-0659.

DELFI for Monitoring Therapeutic Response to Immune Checkpoint Inhibition in Advanced Solid Tumors

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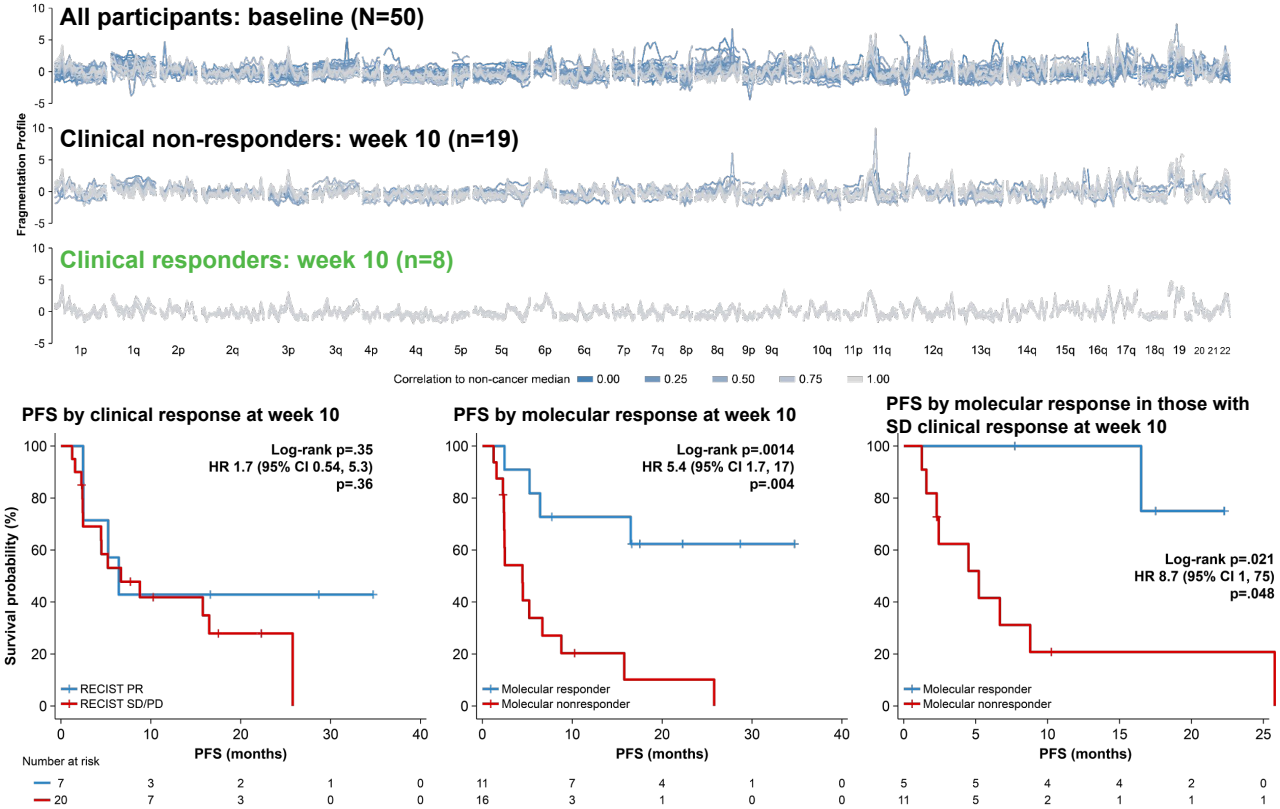

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Medina J, et al. ESMO 2022 - In Collaboration with Evanthia Roussos-Torres, Roisin Connolly and Liz Jaffee (NCT02453620)

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Cell-free DNA Fragmentation Profiling for Monitoring Therapeutic Response in Metastatic Colorectal Cancer

Bahar Alipanahi,[1] Iris van 't Erve,[2] Keith Lumbard,[1] Laurel K. Millberg,[1] Zachary L. Skidmore,[1] Lorenzo Rinaldi,[1] Jacob Carey,[1] Jennifer Tom,[1] Cornelis J. A. Punt,[3] Nicholas C. Dracopoli,[1] Gerrit A. Meijer,[2] Robert B. Scharpf,[4] Victor E. Velculescu,[4] Remond J.A. Fijneman,[2] Alessandro Leal [1]

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2.Department of Pathology, Netherlands Cancer Institute, Amsterdam, Netherlands.
3.Julius Center for Health Sciences and Primary Care, University Medical Center Utrecht, Utrecht, the Netherlands.
4.The Sidney Kimmel Comprehensive Cancer Center, Johns Hopkins University School of Medicine, Baltimore, MD, United States

Increasing Stages of Rigor From Scientific Discovery to Validated Diagnostic Product

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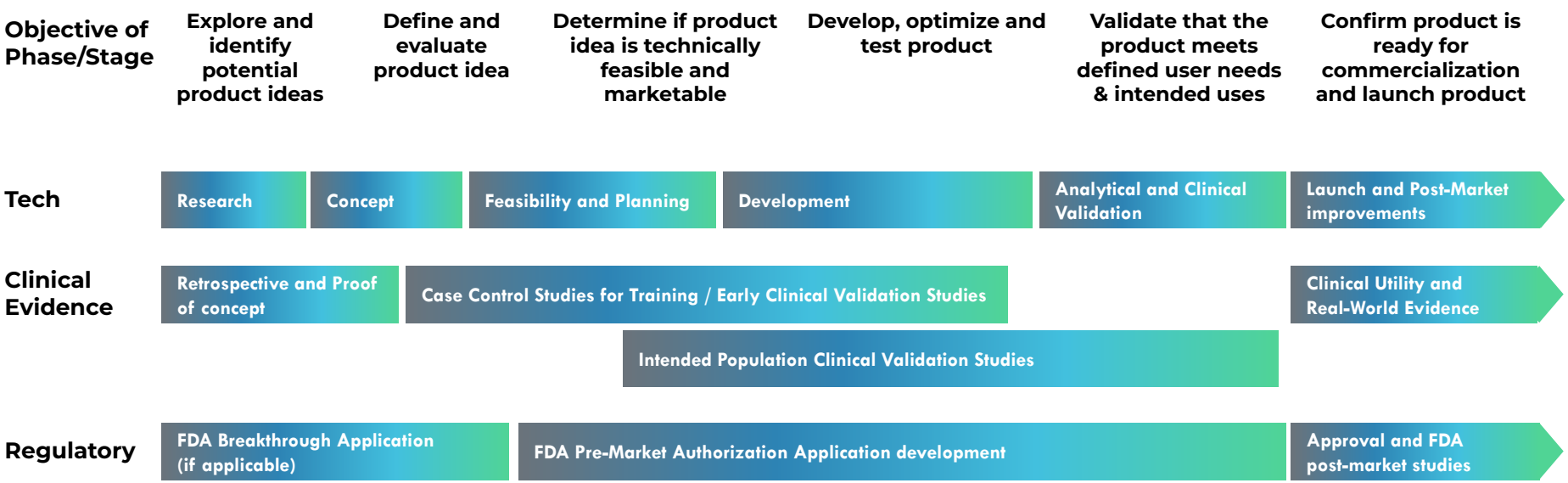

DELFI

Delfi Diagnostics: spinout from JHU to develop clinically useful and widely accessible screening products based on DELFI technology

Courtesy of Jennifer Beuchel, 2023

Prospective National Clinical Trials to Validate DELFI Technology for Detection of Lung and Other Cancers

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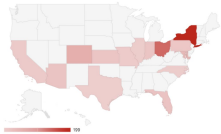
L101	Prospective, observational case-control NCT04825834	***Train*** classifiers for lung cancer detection… … and multi-cancer detection	20+ sites 	N=2,500 participants with lung cancer, non-lung cancers*, and no cancer	≥50 years of age; current or former smoker; ≥20 pack-years	FPI March ‘21 2,000+enrolled
CASCADE - LUNG	Prospective observational cohort NCT05306288	***Validate*** clinical performance of the DELFI lung cancer screening	40+ sites 	N≈15,000 participants undergoing LDCT screening	≥50 years of age; current or former smoker; ≥20 pack-years	FPI April ‘22

* bladder, colorectal, esophageal, gastric, head and neck, kidney, liver, and pancreatic

L101 Prospective Trial: Participant Demographics and Clinical Characteristics

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Characteristic	Lung Cancer, N = 294[1]	Non-cancer, N = 661[1]
AGE	70 (65, 76)	66 (60, 71)
SEX		
Female	154 (52%)	313 (47%)
Male	140 (48%)	348 (53%)
BMI	26 (23, 31)	28 (25, 33)
SMOKING HISTORY		
Current	76 (26%)	267 (40%)
Former	193 (66%)	391 (59%)
Never	25 (8.5%)	3 (0.5%)
RACE		
Asian	12 (4.1%)	8 (1.2%)
Black or African American	23 (7.8%)	54 (8.2%)
Not Reported/Missing	14 (4.8%)	33 (5.0%)
Other	2 (0.7%)	9 (1.4%)
White	243 (83%)	557 (84%)

[1] Median (IQR); n (%)

Lung Cancer Characteristics

Characteristic	N = 294[1]
LUNG CANCER STAGE	
I	147 (50%)
II	40 (14%)
III	65 (22%)
IV	39 (13%)
Unknown Lung Cancer Stage	3 (1.0%)
LUNG CANCER HISTOLOGY	
Adenocarcinoma	177 (60%)
Other	26 (8.8%)
Small cell carcinoma (SCLC)	21 (7.1%)
Squamous cell carcinoma (SCC)	70 (24%)

[1] n (%)

L101 Prospective Trial: Fragmentome profiles are not affected by smoking status

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L101 Prospective Trial: Fragmentome profiles are not affected by sex

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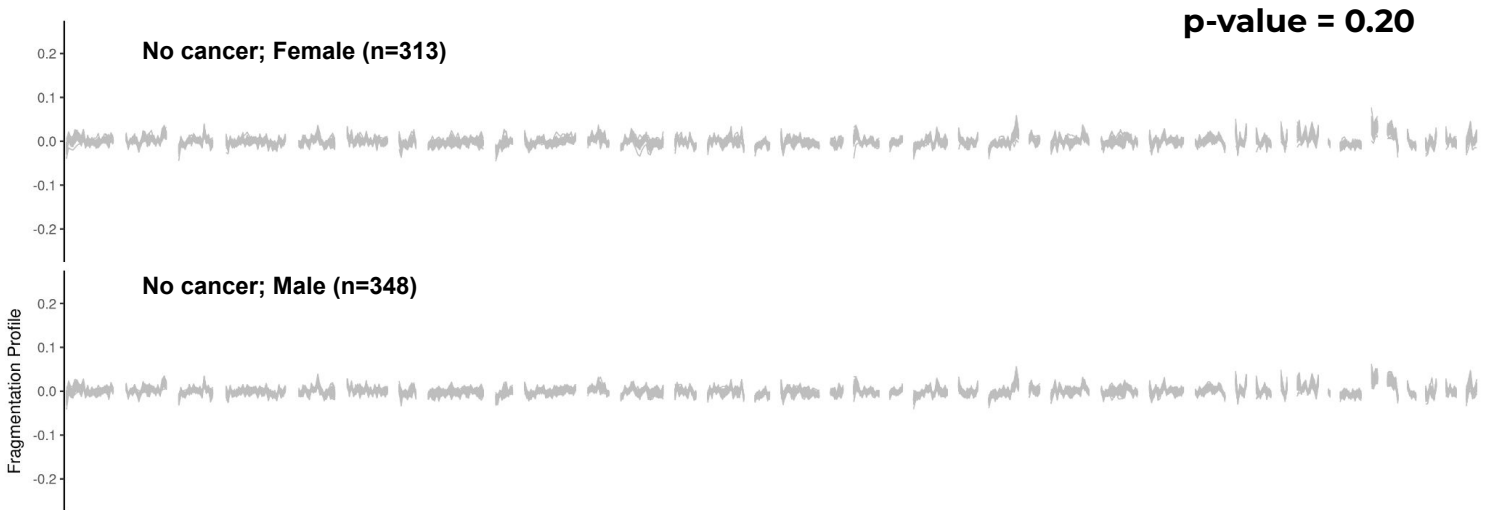

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L101 Prospective Trial: Fragmentome profiles are not affected by BMI

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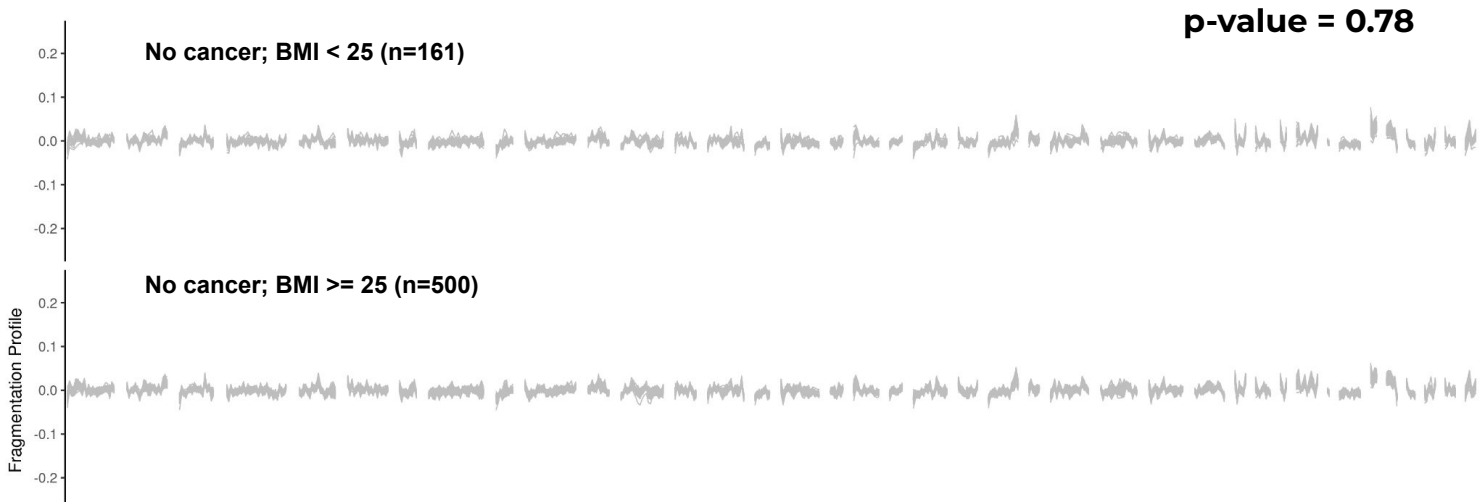

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L101 Prospective Trial: Fragmentome profiles are not affected by benign lesions

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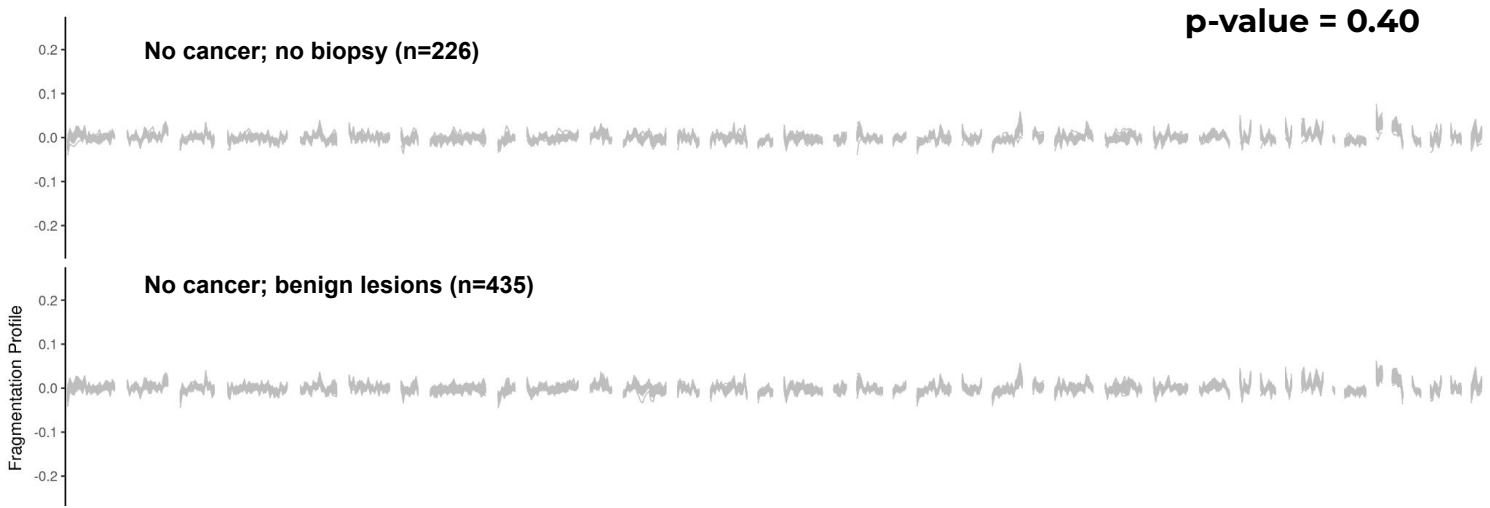

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DELFI Cross-validated Classifier Distinguishes Lung Cancer With High Performance in L101

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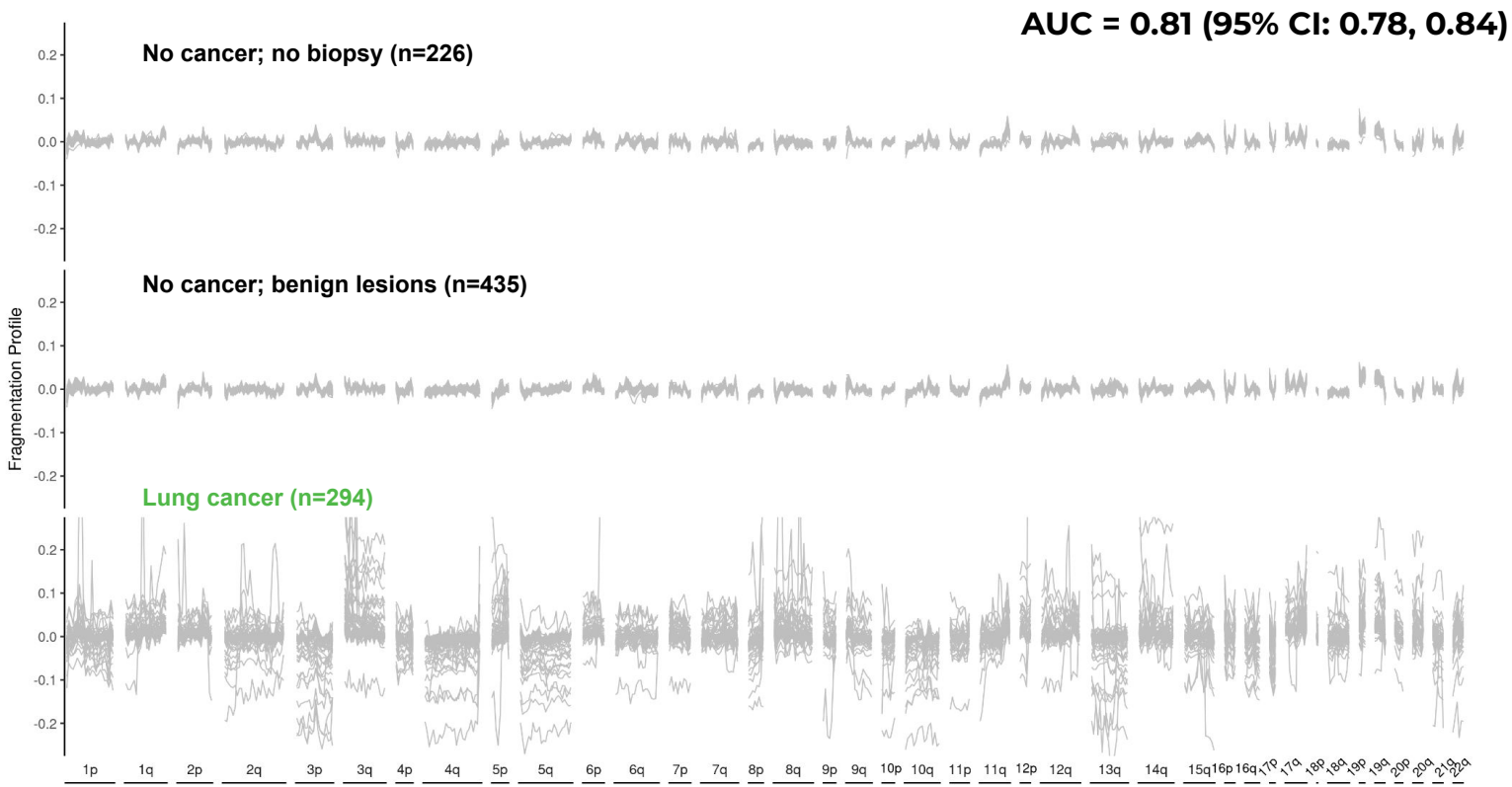

Conclusions and Opportunities

- Genome-wide cfDNA fragmentome features detect cancer with high sensitivity, including in early-stage disease.
- Fragmentation profiles capture genomic and chromatin characteristics, including alterations known to be important in cancer.
- The high performance of cfDNA fragmentome analyses for cancer detection, along with its cost-efficient characteristics, could overcome the limitations of current cancer screens.

Acknowledgments

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The generous participants in this study
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