Détection précoce des cancers digestifs

Pierre Laurent-Puig

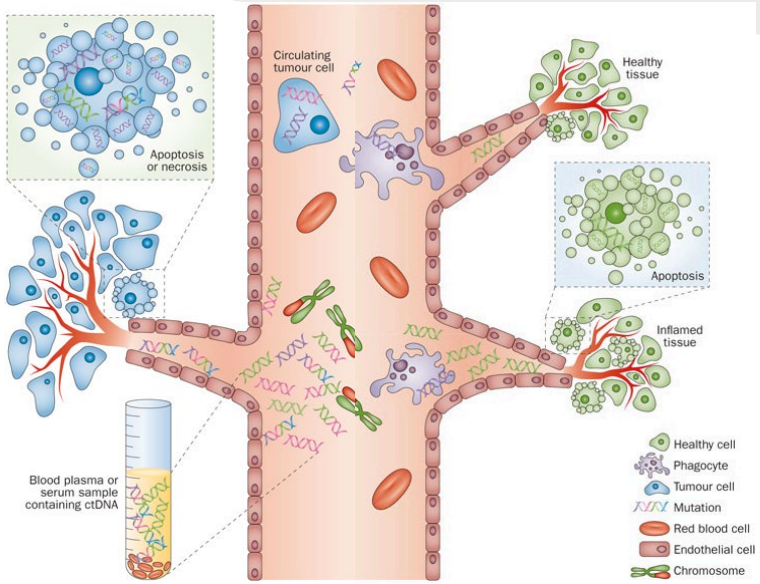
Service de Médecine Génomique des Tumeurs et Cancers

Hôpital Européen Georges Pompidou et Hôpital Cochin

Centre de Recherche des Cordeliers

MEdecine Personnalisée, Pharmacogénomique et Optimisation Thérapeutique

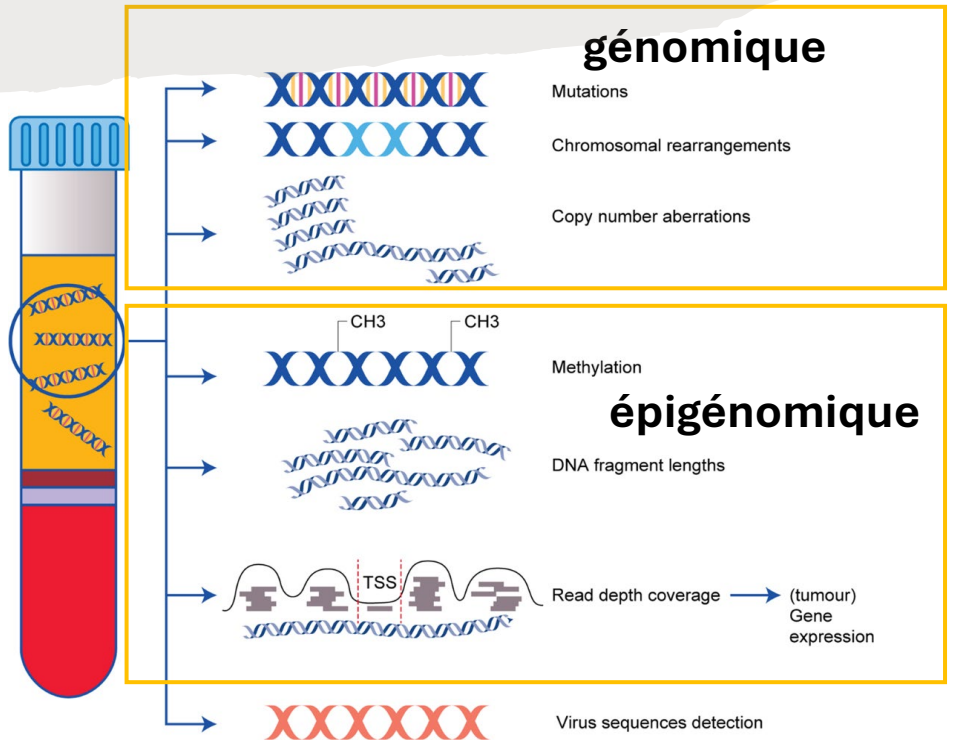
ADN circulant origine et caractérisation



(Crowley et al, 2013)

- **Cancer**

- **Lyse cellulaire spécifique d'organe**
  - Greffe
  - Maladie auto-immune

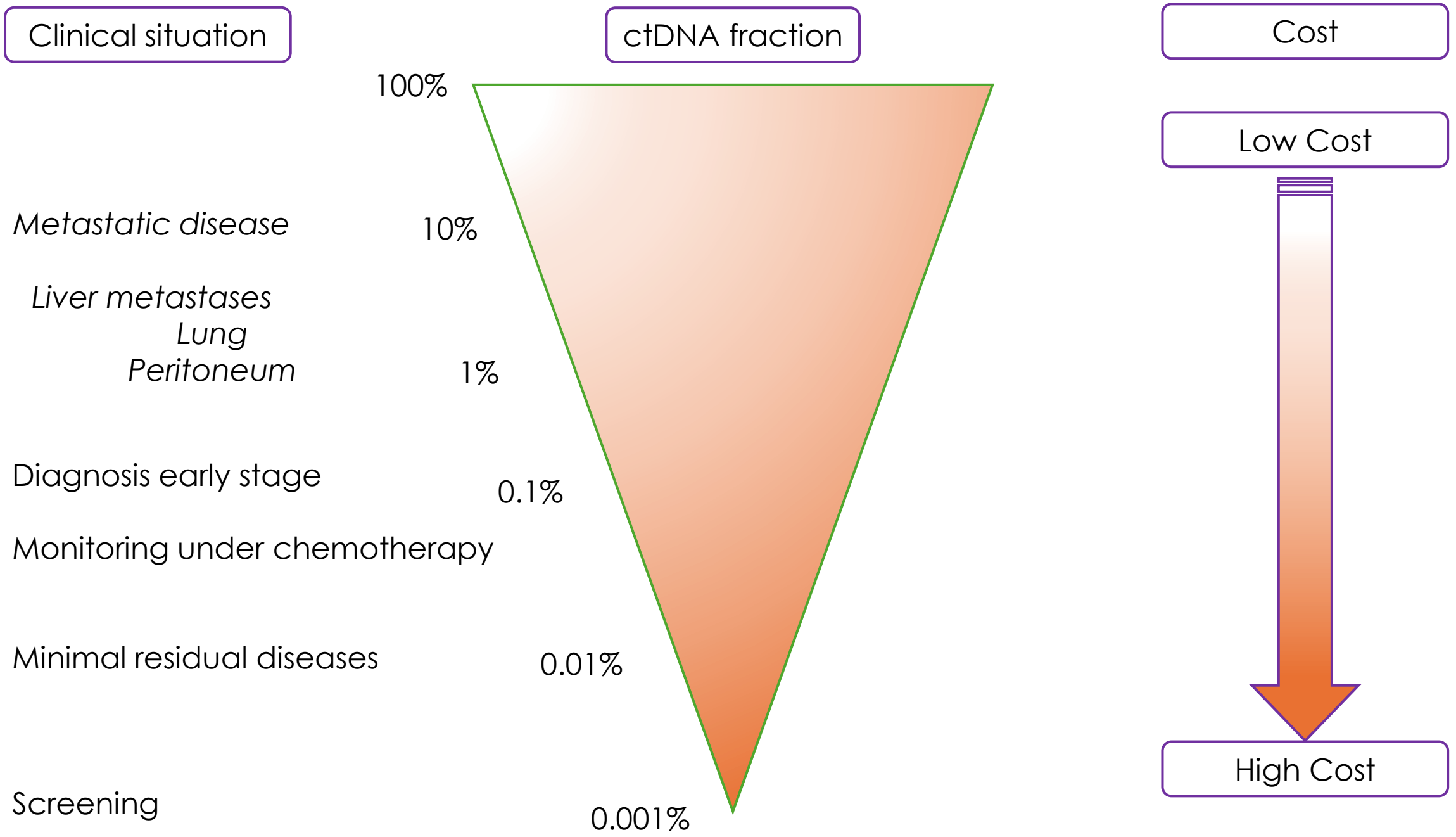


Biomarqueurs

ADN = 400-4000 équivalents génome / mL

Fraction pathologique faible, souvent <1%

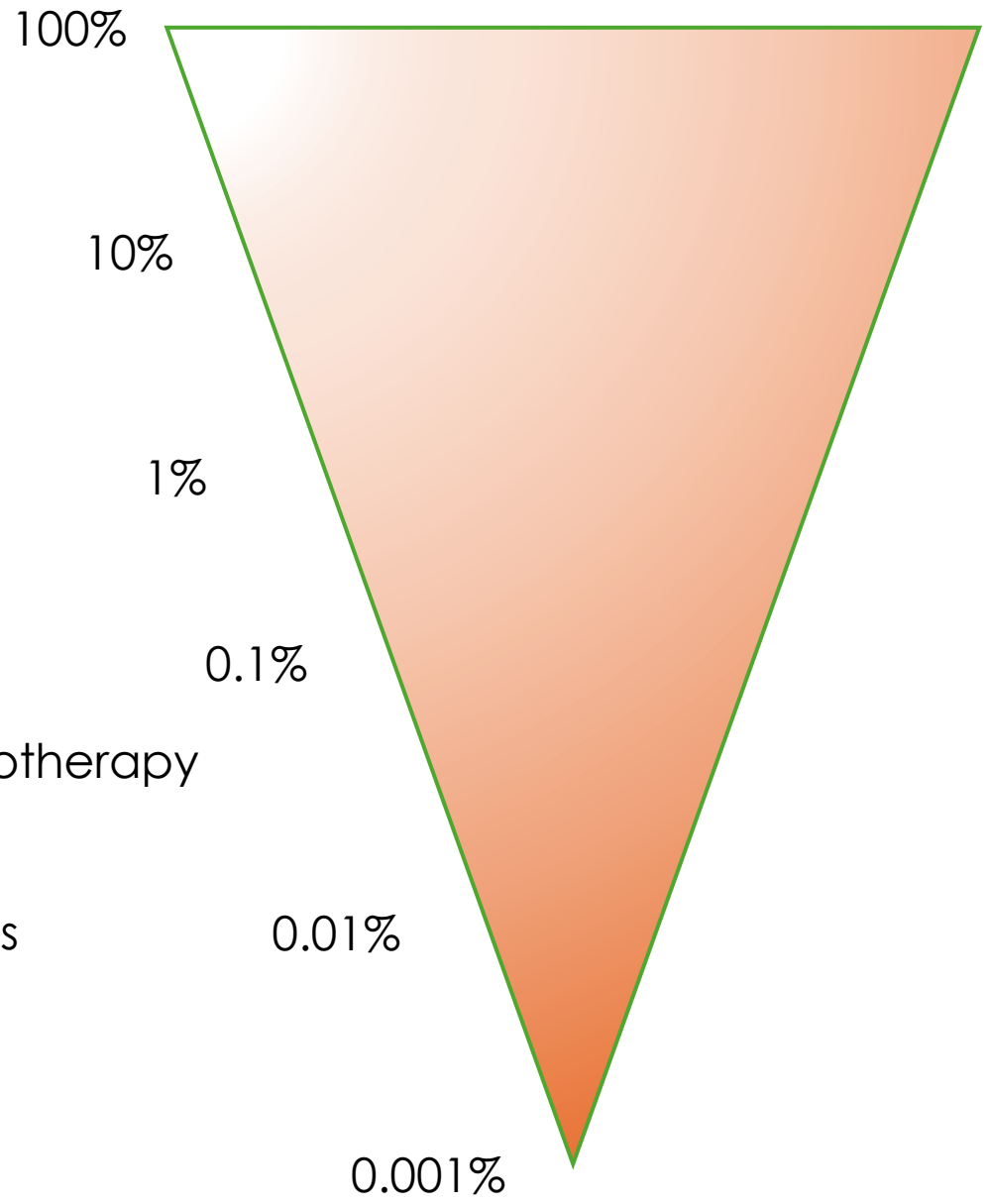
Technologies spécifiques /Séquençage à haut débit


Clinical situation
ctDNA fraction
Cost
100%
Low Cost
Metastatic disease
10%
Liver metastases
Lung
Peritoneum
1%
Diagnosis early stage
0.1%
Monitoring under chemotherapy
Minimal residual diseases
0.01%
Screening
0.001%
High Cost

Clinical situation

ctDNA fraction

Trap



Metastatic disease

Liver metastases
Lung
Peritoneum

Diagnosis early stage

Monitoring under chemotherapy

Minimal residual diseases

Screening

Clonal hematopoeisis

PCR artefact

Les différents tests avantages /incovénients

JAMA Oncology | Original Investigation

Analysis of Plasma Cell-Free DNA by Ultradeep Sequencing in Patients With Stages I to III Colorectal Cancer

Thomas Reinert, PhD; Tenna Vesterman Henriksen, MSc; Emil Christensen, PhD; Shruti Sharma, PhD; Raheleh Salari, PhD; Himanshu Sethi, MPH; Michael Knudsen, PhD; Iver Nordentoft, PhD; Hsin-Ta Wu, PhD; Antony S. Tin, PhD; Mads Heilskov Rasmussen, PhD; Søren Vang, PhD; Svetlana Shchegrova, PhD; Amanda Frydendahl Boll Johansen, MSc; Ramya Srinivasan, Msc; Zoe Assaf, PhD; Mustafa Balcioglu, PhD; Alexander Olson, BSc; Scott Dashner, BSc; Dina Hafez, PhD; Samantha Navarro, BSc; Shruti Goel, PhD; Matthew Rabinowitz, PhD; Paul Billings, MD, PhD; Styrmir Sigurjonsson, PhD; Lars Dyrskjøt, PhD; Ryan Swenerton, PhD; Alexey Aleshin, MBA, MD; Søren Laurberg, DMSc; Anders Husted Madsen, MD, PhD; Anne-Sofie Kannerup, MD, PhD; Katrine Stribolt, MD; Søren Palmelund Krag, MD, PhD; Lene H. Iversen, MD, PhD; Kåre Gotschalck Sunesen, MD, PhD; Cheng-Ho Jimmy Lin, MD, PhD, MHS; Bernhard G. Zimmermann, PhD; Claus Lindbjerg Andersen, PhD

WES of leucocytes

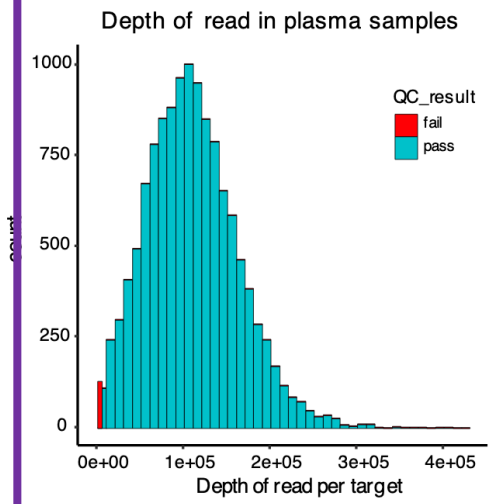
WES tumor DNA

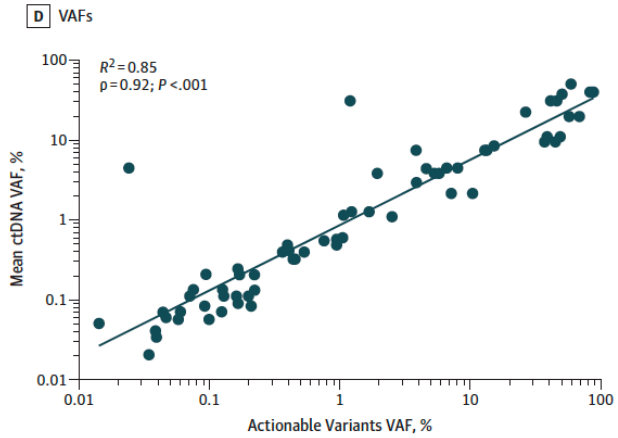
List of variants specific of tumor DNA

Selection of up to 16 variants based on observed VAF in tissue, sequence context of variants and short INDELs. These variants are expected to have a low plasma background mutation rate.

Design of primers for the 16 amplicons

Ultra deep sequencing





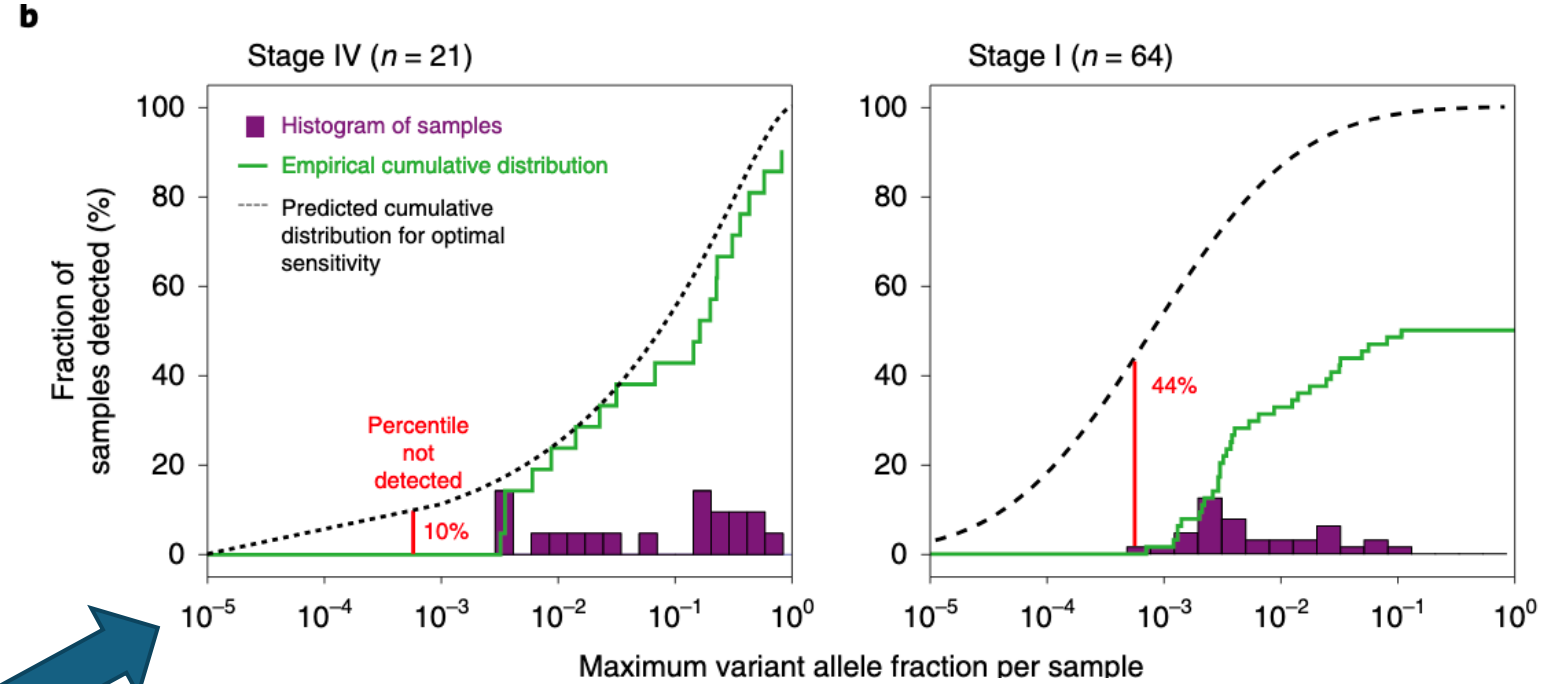
Mean 100 000 reads

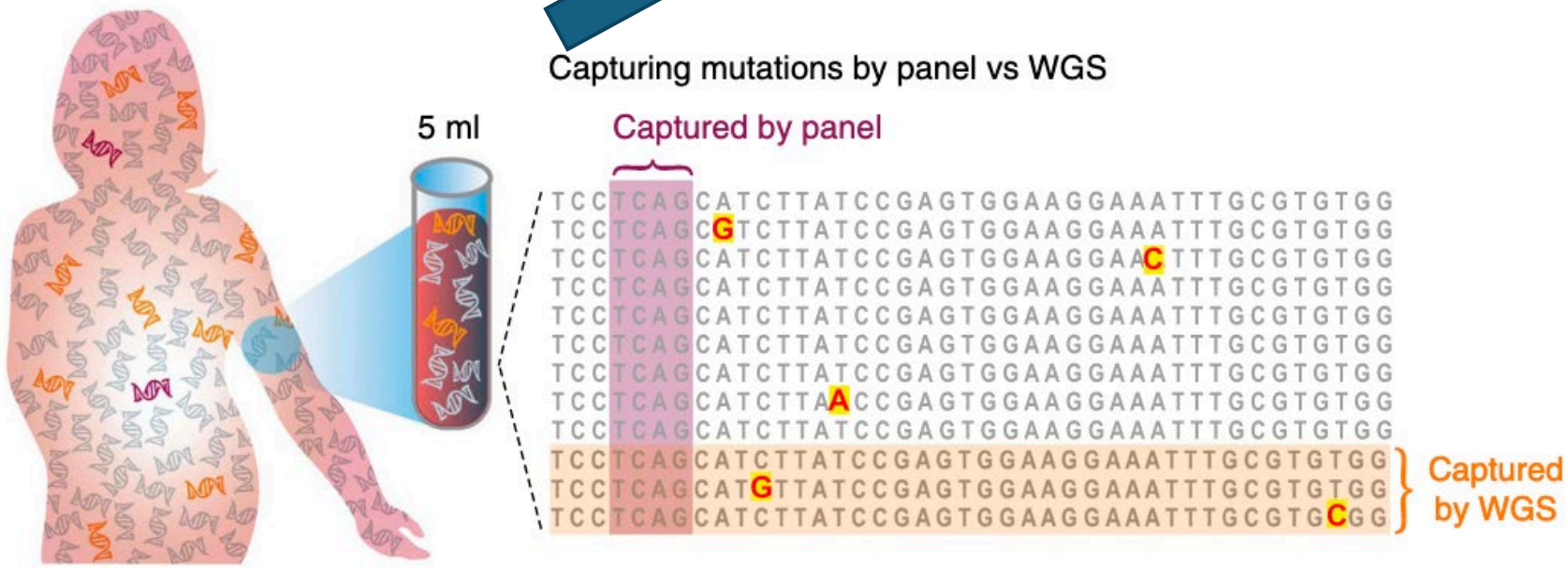
Avoid clonal hematopoiesis false positive
Avoid sequencing error by redundancy
Limited background noise by an educated choice
But 2 WES - Amplicon design for each patients

ARTICLES
https://doi.org/10.1038/s41591-020-0915-3
nature medicine

Genome-wide cell-free DNA mutational integration enables ultra-sensitive cancer monitoring

Asaf Zviran[1,2,7], Rafael C. Schulman[1,2,7], Minita Shah[1,7], Steven T. K. Hill[1,2], Sunil Deochand[1,2], Cole C. Khamnei[1,2], Dillon Maloney[1], Kristofer Patel[1,2], Will Liao[1], Adam J. Widman[1,2,3], Phillip Wong[3], Margaret K. Callahan[3], Gavin Ha[4], Sarah Reed[5], Denisse Rotem[5], Dennie Frederick[6], Tatyana Sharova[6], Benchun Miao[6], Tommy Kim[6], Greg Gydush[5], Justin Rhoades[5], Kevin Y. Huang[1,2], Nathaniel D. Omans[1,2], Patrick O. Bolan[2], Andrew H. Lipsky[2], Chelston Ang[1,2], Murtaza Malbari[2], Catherine F. Spinelli[2], Selena Kazancioglu[1], Alexi M. Runnels[1], Samantha Fennessey[1], Christian Stolte[1], Federico Gaiti[1,2], Giorgio G. Inghirami[2], Viktor Adalsteinsson[5], Brian Houck-Loomis[1], Jennifer Ishii[1], Jedd D. Wolchok[3], Genevieve Boland[6], Nicolas Robine[1], Nasser K. Altorki[2] and Dan A. Landau[1,2]✉





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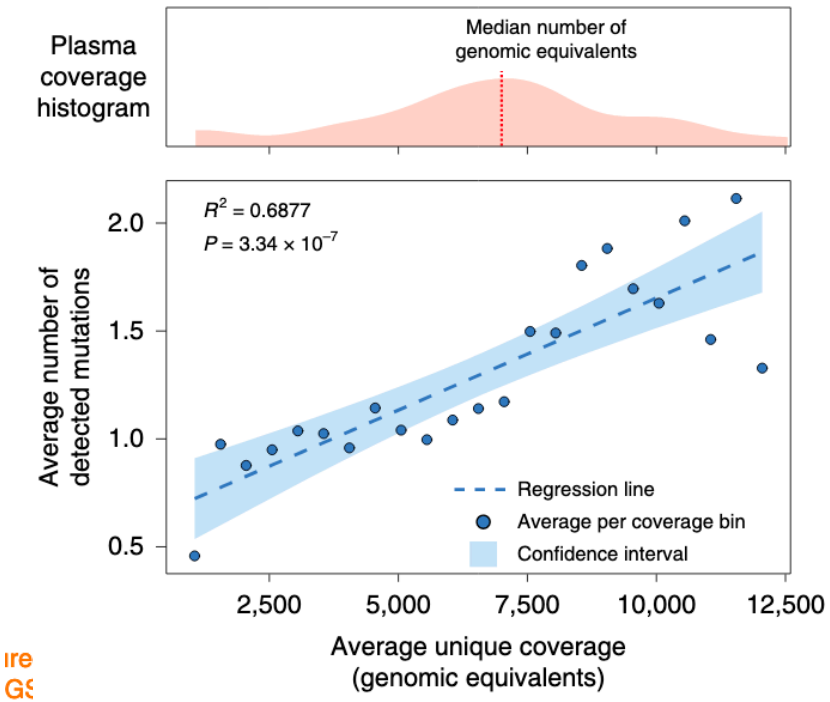
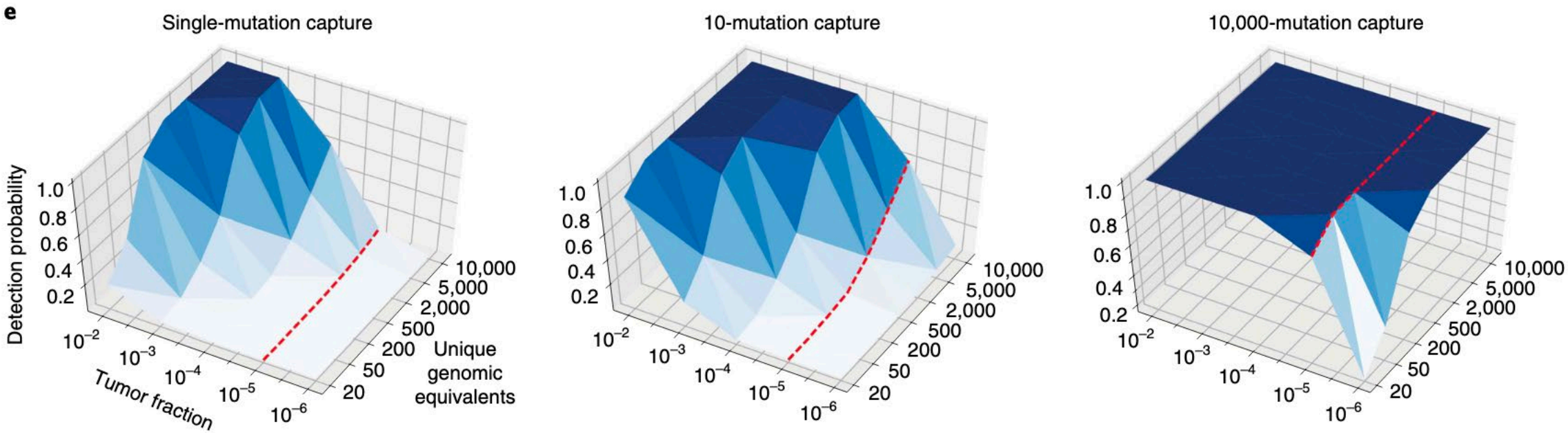
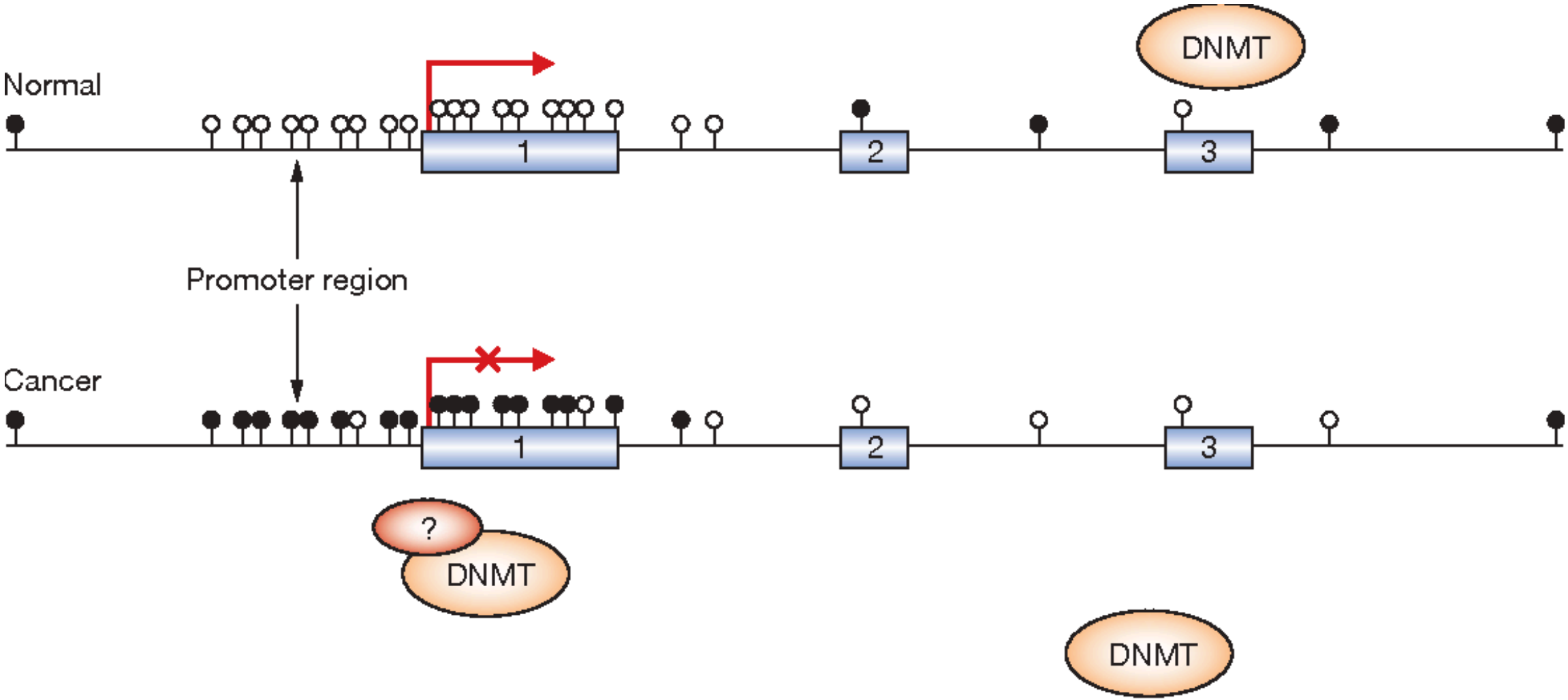


194 patients, across different tumor types and disease stages sequenced with exhaustive deep sequencing (median, 40,729×).

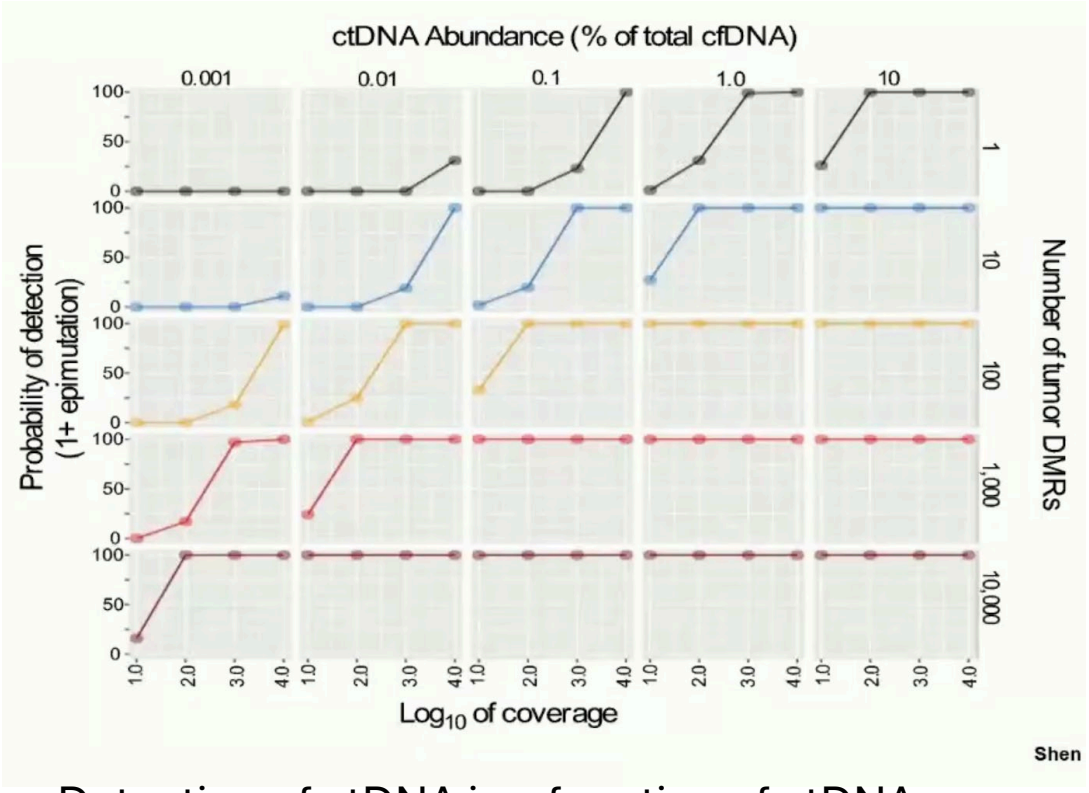
A median of 6,000 (range, 400–14,000) unique reads (GEs) was found in a median of 4.2 ml of plasma (range, 1.6–9.8)



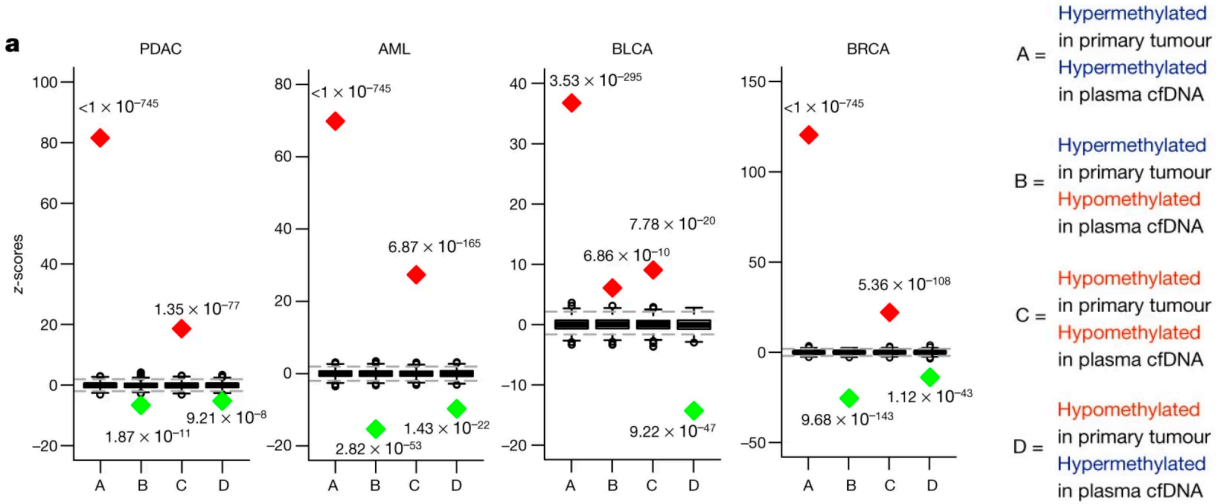

Normal
DNMT
1
2
3
Promoter region
Cancer
1
2
3
?
DNMT
DNMT

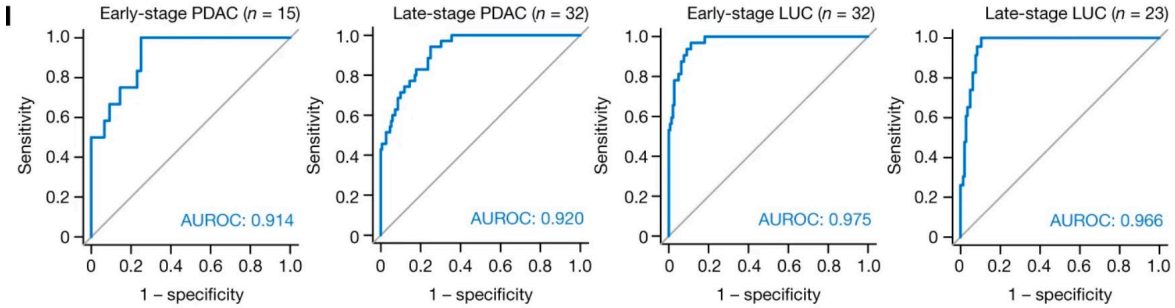
Sensitive tumour detection and classification using plasma cell-free DNA methylomes

Shu Yi Shen[1,12], Rajat Singhania[1,12], Gordon Fehringer[2,12], Ankur Chakravarthy[1,12], Michael H. A. Roehrl[1,3,4], Dianne Chadwick[1], Philip C. Zuzarte[5], Ayelet Borgida[2], Ting Ting Wang[1,4], Tiantian Li[1], Olena Kis[1], Zhen Zhao[1], Anna Spreafico[1], Tiago da Silva Medina[1], Yadon Wang[1], David Roulois[1,6], Ilias Ettayebi[1,4], Zhuo Chen[1], Signy Chow[1], Tracy Murphy[1], Andrea Arruda[1], Grainne M. O'Kane[1], Jessica Liu[4], Mark Mansour[4], John D. McPherson[7], Catherine O'Brien[1], Natasha Leighl[1], Philippe L. Bedard[1], Neil Fleshner[1], Geoffrey Liu[1,4,8], Mark D. Minden[1], Steven Gallinger[9,10], Anna Goldenberg[11], Trevor J. Pugh[1,4], Michael M. Hoffman[1,4,11], Scott V. Bratman[1,4], Rayjean J. Hung[2,8]* & Daniel D. De Carvalho[1,4]*



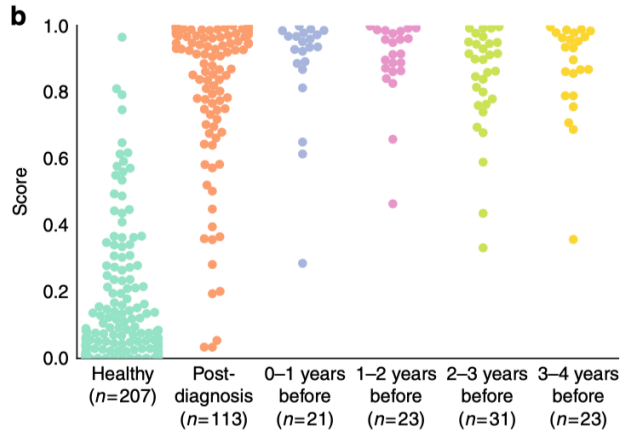
Detection of ctDNA is a function of ctDNA abondance, sequencing depth and number of features investigated





Non-invasive early detection of cancer four years before conventional diagnosis using a blood test

Xingdong Chen[1,2,3,12], Jeffrey Gole[4,12], Athurva Gore[4,12], Qiye He[5,12], Ming Lu[2,6,12], Jun Min[4], Ziyu Yuan[2], Xiaorong Yang[2,6], Yanfeng Jiang[1,2], Tiejun Zhang[7], Chen Suo[7], Xiaojie Li[5], Lei Cheng[5], Zhenhua Zhang[5], Hongyu Niu[5], Zhe Li[5], Zhen Xie[5], Han Shi[4], Xiang Zhang[8], Min Fan[9], Xiaofeng Wang[1,2], Yajun Yang[1,2], Justin Dang[4], Catie McConnell[4], Juan Zhang[2], Jiucun Wang[1,2,3], Shunzhang Yu[2,7], Weimin Ye[2,10]✉, Yuan Gao[4]✉, Kun Zhang[11]✉, Rui Liu[4,5]✉ & Li Jin[1,2,3]✉

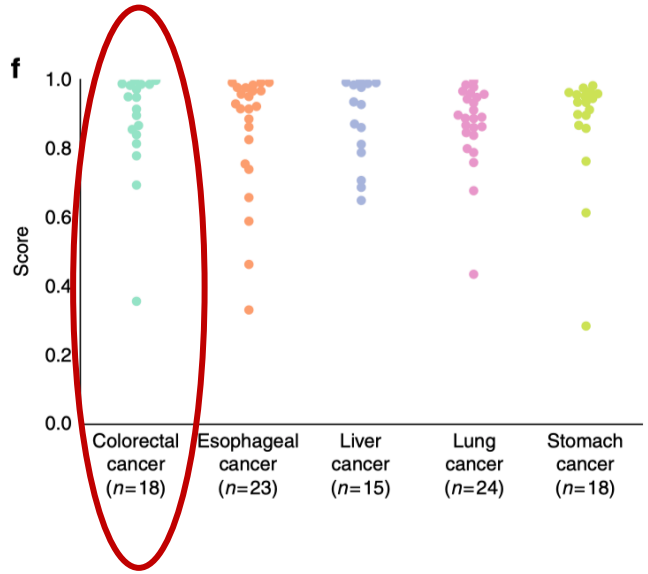


The PanSeer assay utilizes methylation biomarkers to their fullest extent by sensitively targeting 10,613 CpG sites across 477 genomic regions

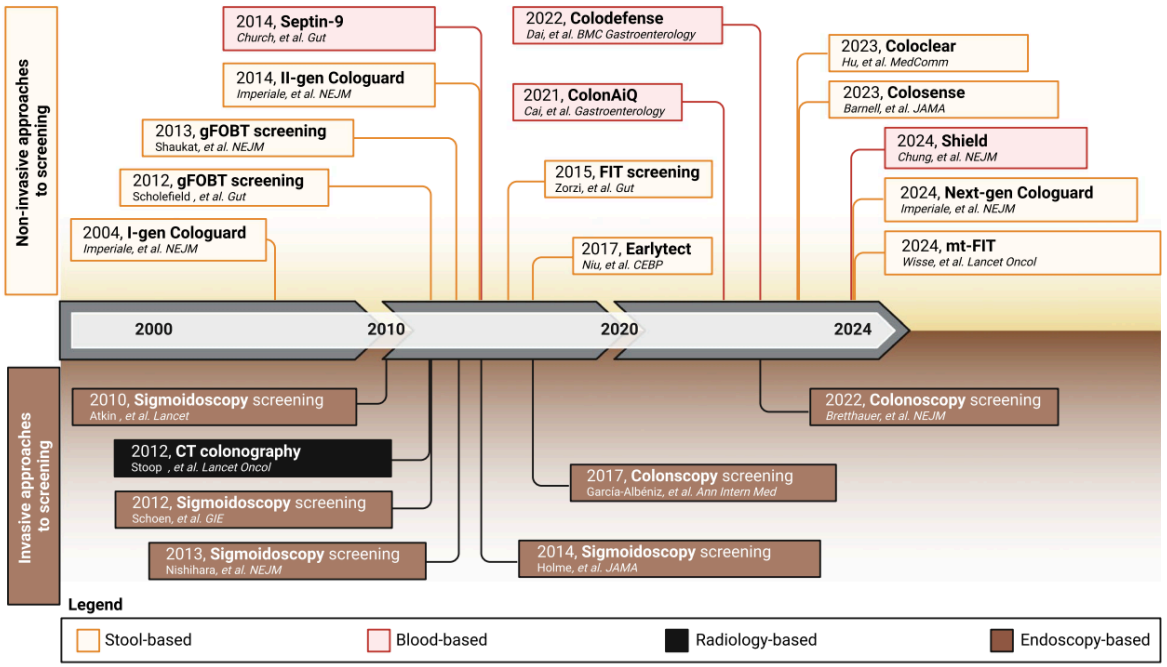
Table 1 Accuracy of PanSeer.

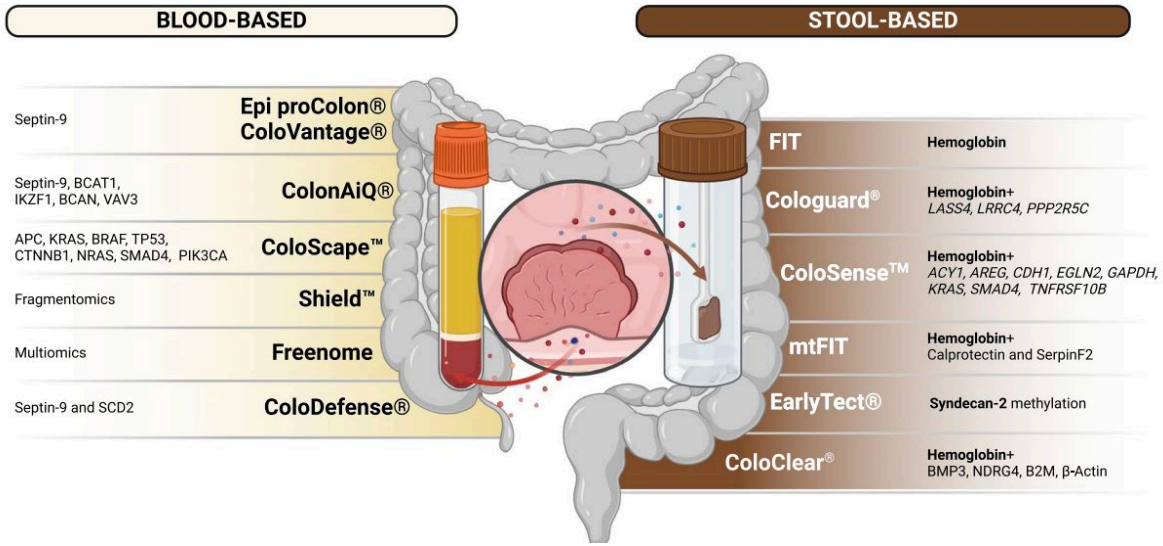
		Training set			Test set		
Category	**Total**	**# of Samples**	**Specificity (%, 95% CI)**	**Sensitivity (%, 95% CI)**	**# of Samples**	**Specificity (%, 95% CI)**	**Sensitivity (%, 95% CI)**
Healthy	414	207	94.7 (90.7-97.3)		207	96.1 (92.5-98.3)	
Post-diagnosis	223	110		88.2 (80.6-93.6)	113		87.6 (80.1-93.1)
Pre-diagnosis	191	93		91.4 (83.8-96.2)	98		94.9 (88.5-98.3)
0-1 year before diagnosis		22		100 (84.6-100)	21		95.2 (76.2-99.9)
1-2 year before diagnosis		21		90.5 (69.6-98.8)	23		95.7 (78.1-99.9)
2-3 year before diagnosis		19		94.7 (74.0-99.9)	31		93.6 (78.6-99.2)
3-4 year before diagnosis		31		83.9 (66.3-94.6)	23		95.7 (78.1-99.9)

Sensitivity and specificity for the training set and test set are presented and divided into subcategories by the number of years prior to cancer diagnosis by conventional testing.



Chen et al. Nature Comm 2020





Mannucci and Goel Molecular Cancer (2024) 23:259

Les tests de dépistage basés sur la detection de l'ADNtc

Étude (Test)	Marqueurs / n / type d'étude	Sensibilité CRC	Spécificité
Church et al. — Epi proColon™ (NCT00855348)	SEPT9 (méthylation) n = 7 941 • Prospectif	48,2 % (IC95 % 32,4–63,6) Stades : I 35 % • II 63 % • III 46 % • IV 77,4 %	91,5 % (IC95 % 89,7–93,1)
Chung et al. — Shield™ (NCT04136002)	Méthylation + mutations + fragmentomique n = 7 861 • Prospectif	83,1 % (IC95 % 72,2–90,3) Stades : I 65 % • II 100 % • III 100 % • IV 100 %	89,6 % (IC95 % 88,8–90,3)
PREEMPT CRC™ [11]	Multi-omique (Méthylation + fragmentation+ altérations nombre de copies n = 27 010	79,2 % Stades : I 57,1 % • II 100 % • III 82,4 % • IV 100 %	91,5 %
Cai et al. — ColonAiQ™	SEPT9, BCAT1, IKZF1, BCAN, VAV3 (méthylation) n = 348 • Rétrospectif	86,1 % (IC95 % 79,9–90,7) Stades : I 78,3 % • II 82,0 % • III 86,0 % • IV 100 %	91,9 % (IC95 % 85,7–95,7)
Zhao et al. — ColoDefense™	SEPT9 + SDC2 (méthylation) n = 384 • Rétrospectif	88,9 % (IC95 % 81,4–93,7) Stades : I 80 % • II 90 % • III 89,5 % • IV 100 %	92,8 % (IC95 % 87,4–96,0)

AA : adénome avancé • CRC : cancer colorectal. Laurent-Puig P., Bull Acad Natl Med 209 (2025) 1002–1012.

Les tests de dépistage comparaison plasma/selles

	FIT	Cologuard Plus	ColoSense	Shield (Guardant) ou Freenome
Type d'analyse	Test immunochimique fécal (détection d'hémoglobine)	ADN fécal multi-cibles + FIT	8 marqueurs d'ARN d'origine fécale + FIT	ADN tumoral circulant (sang) ; approche épigénomique
Sensibilité CRC	67–91 %	95 %	94 %	80,5–83 %
Sensibilité pour adénome avancé	20–30 %	43 %	46 %	13–16,4 %
Sensibilité pour adénome dysplasie de haut grade	47 %	74 %	N/A	23 %
Spécificité	95 %	91–94 %	88 %	90 %

Imperiale TF, et al NEJM 2024, Ann Intern Med 2019 Chung, D et al, NEJM 2024

Feces-based Tests for Advanced Adenomas

Mannucci and Goel Molecular Cancer (2024) 23:259

Test's Name	Company	Sample Type	Targets Tested	Population & Study Design	Sensitivity for AA (95% CI)
FIT	–	Feces	Hemoglobin	Meta-analysis of FITs vs colonoscopy (31 studies, 120,255 participants)	40% (33–47%)
FIT	–	Feces	Hemoglobin	Meta-analysis, 10 studies (63,473 participants)	27% (23–31%)
Cologuard	Exact Sciences	Feces	Hb + 3 KRAS + 10 APC + 8 TP53	Asymptomatic, average-risk ≥50 yrs (N=2,507)	12.1% (12.0–19.0%)
Cologuard	Exact Sciences	Feces	Hb + 7 KRAS + NDGR4, BMP3 + β-Actin	Asymptomatic, average-risk 50–84 yrs (N=9,989)	42.4% (38.9–46.0%)
Cologuard	Exact Sciences	Feces	Hb + LASS4, LRRC4, PPP2R5C, ZDHHC1	Asymptomatic, average-risk ≥40 yrs (N=20,176)	43.4% (41.3–45.6%)
ColoSense	Geneoscopy	Feces	Hb + Smoking + 8 mRNA (ACY1, AREG, CDH1, EGLN2, GAPDH, KRAS, SMAD4, TNFRSF10B)	Asymptomatic, average-risk ≥45 yrs (N=8,920)	45.9% (42–50%)
EarlyTect / Colosafe	Genomic Tree / Creative Biosciences	Feces	SDC2 methylation	Meta-analysis of blood Septin-9 (25 studies)	15% (11–19%)
Coloclear	Prenetics	Stool	Hb + KRAS + BMP3, NDRG4, β-Actin, B2M	High-risk CRC (N=4,758)	63.5% (58.6–68.3%)

Imperiale et al. Ann Intern Med. 2019;170:319–29 Niedermaier et al. Eur J Epidemiol. 2017;32:481–93. Imperiale et al. N Engl J Med. 2004;351:2704–14. Imperiale et al. N Engl J Med. 2014;370:1287–97. Imperiale et al. N Engl J Med. 2024;390:984–93. Barnell EK et al. JAMA. 2023;330:1760–8 Niu et al. Cancer Epidemiol Biomarkers Prev. 2017;26:1411–9. Han et al. Clin Epigenetics. 2019;11:51. Hu et al. multicenter clinical study. MedComm 2020. 2023;4:e345.

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Blood-based Tests for Advanced Adenomas

Mannucci and Goel Molecular Cancer (2024) 23:259

Test's Name	Company	Sample Type	Targets Tested	Population & Study Design	Sensitivity for AA (95% CI)
Epi proColon / Colovantage	Epigenomics / Quest	Blood	Septin-9 methylation	Prospective study, adults ≥50 yrs (N=7,941)	11.2%
Shield	Guardant Health	Blood	cfDNA fragmentomics	Prospective trial (N=7,861)	13.2% (11.3–15.3%)
Freenome	Freenome	Blood	Multiomics	Screen-eligible adults 45–85 yrs (N=48,995)	12.5% (11.3–13.8%)
ColonAiQ	Singlera / Clinomics	Blood	5 methylations (Septin-9, BCAT1, IKZF1, BCAN, VAV3)	Retrospective case–control (N=348)	42% (33.1–51.5%)
Coloscape	Diacarta	Blood	61 mutations (APC, KRAS, BRAF, TP53, CTNNB1, NRAS, SMAD4, PIK3CA) + 7 methylations	Retrospective case–control (N=NA)	60% (17–93%)
ColoDefense2.0	VersaBio Technologies	Blood	Methylation (Septin-9, SDC2)	Retrospective case–control (N=529)	55.0% (38.6–70.4%)

Church et al. Gut. 2014;63:317–25. Nian et al. Clin Transl Gastroenterol. 2017;8: e216. Cai et al. Colorectal Cancer. Gastroenterology. 2021;161:2053-6. Tanaka et al. Cancer Research. 2023;83:6505. Chung et al. N Engl J Med. 2024;390:973–83. Dai et al. BMC Gastroenterol. 2022;22:428. Freenome

Blood-based Tests for Advanced Adenomas

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Shield	Guardant Health	Blood	cfDNA fragmentomics	Prospective trial (N=7,861)	13.2% (11.3–15.3%)
Freenome	Freenome	Blood	Multiomics	Screen-eligible adults 45–85 yrs (N=48,995)	12.5% (11.3–13.8%)
ColonAiQ	Singlera / Clinomics	Blood	5 methylations (Septin-9, BCAT1, IKZF1, BCAN, VAV3)	Retrospective case–control (N=348)	42% (33.1–51.5%)
Coloscape	Diacarta	Blood	61 mutations (APC, KRAS, BRAF, TP53, CTNNB1, NRAS, SMAD4, PIK3CA) + 7 methylations	Retrospective case–control (N=NA)	60% (17–93%)
ColoDefense2.0	VersaBio Technologies	Blood	Methylation (Septin-9, SDC2)	Retrospective case–control (N=529)	55.0% (38.6–70.4%)

Church et al. Gut. 2014;63:317–25. Nian et al. Clin Transl Gastroenterol. 2017;8: e216. Cai et al. Colorectal Cancer. Gastroenterology. 2021;161:2053-6. Tanaka et al. Cancer Research. 2023;83:6505. Chung et al. N Engl J Med. 2024;390:973–83. Dai et al. BMC Gastroenterol. 2022;22:428. Freenome

PATHFINDER Study: Key Findings and Limitations

- 🔍 **Key Results**
- 6,621 analysable participants ≥50 years
- Cancer signal detected in 92 (1.4%)
  - 35 true positives (38%)
  - 57 false positives (62%)
- Sensitivity: 29% (35/121 cancers in 12 months)
- Specificity: 99.1%
- Positive Predictive Value (PPV): 38%
- Negative Predictive Value (NPV): 98.6%
- 48% of new cancers: stage I–II
- 0.26% underwent unnecessary procedures

⚠️ **Limitations**

- Sensitivity remains low (29%)
- Cannot replace standard screening programs
- High rate of false positives (esp. hematologic signals)
- Long diagnostic time for false positives (162 days
- Mortality benefit not demonstrated
- Cost-effectiveness still uncertain

Schrag et al. Lancet 2023; 402: 1251–60

SYMPLIFY Study: MCED Test in Symptomatic Patients

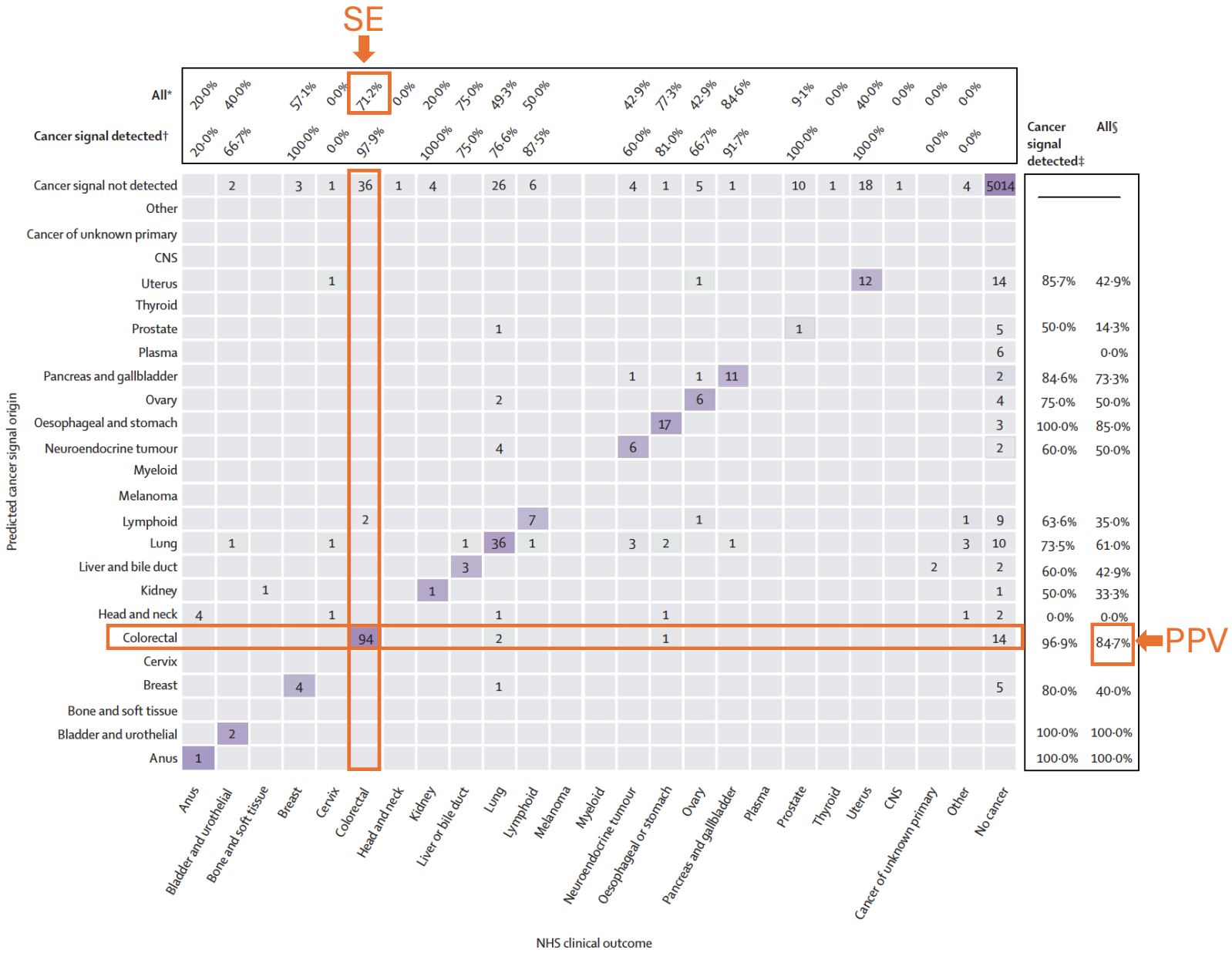
🔍 Key Results

- 5461 evaluable participants (England & Wales, NHS)
- Cancer prevalence: 6.7% (368 diagnosed)
- MCED test performance:
  - Sensitivity: 66.3% (61.2–71.1)
  - Specificity: 98.4% (98.1–98.8)
  - Positive Predictive Value (PPV): **75.5% (70.5–80.1)**
  - Negative Predictive Value (NPV): 97.6% (97.1–98.0)
- Sensitivity increased with stage: 24% (I) → 95% (IV)
- Correct cancer signal origin prediction: ~85%

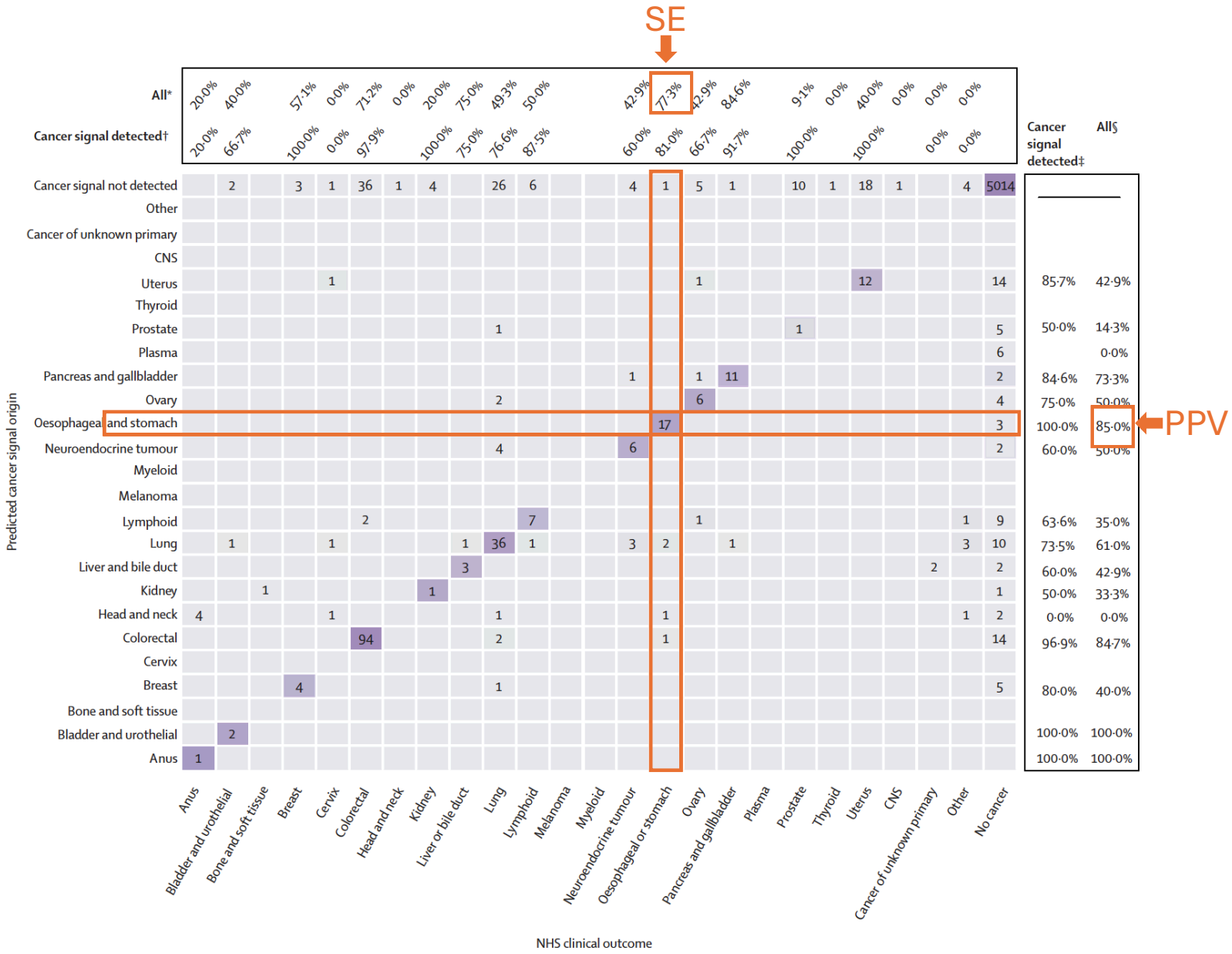
⚠️ Limitations & Interpretation

- Moderate sensitivity limits cancer rule-out potential
- Algorithm optimized for asymptomatic screening population
- Low early-stage sensitivity (37% for stage I–II)
- Lower performance in non-specific symptom referrals
- Observational study — no proven impact on mortality or diagnostic delay
- Requires interventional validation before clinical use
- May serve best as triage support for symptomatic patients

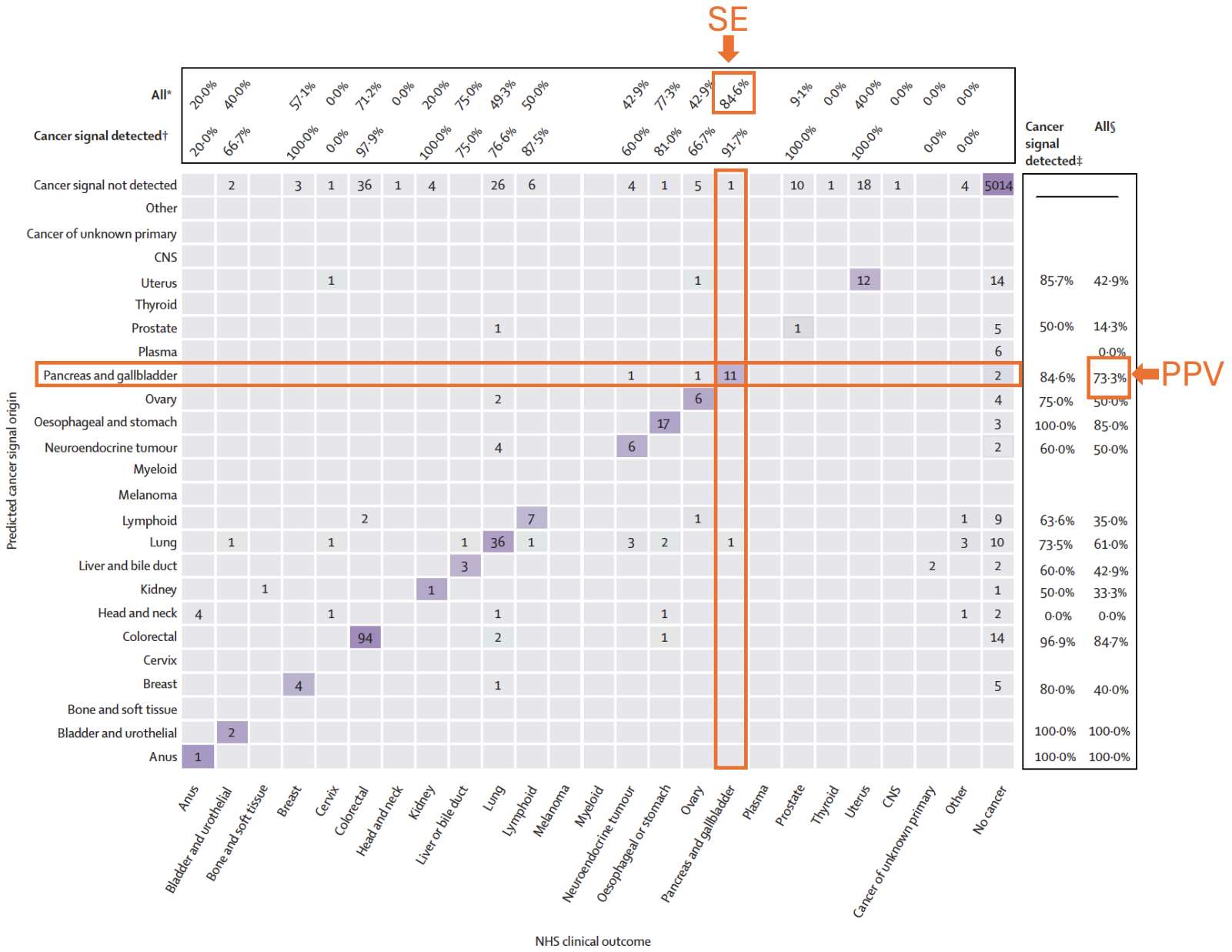
SYMPLIFY Study: MCED Test in Symptomatic Patients



SYMPLIFY Study: MCED Test in Symptomatic Patients



SYMPLIFY Study: MCED Test in Symptomatic Patients



Conclusions

- Le dépistage des cancers digestifs par la mise en évidence de l'ADN tumoral nécessite des techniques ultra sensibles
- Pour le cancer du côlon les performances des tests sanguins ne surpassent pas les tests à partir des selles
- Pour les autres cancers digestifs les données sont très parcellaires
- Chez les patients symptomatiques (loin du dépistage) donnés intéressantes
- Capacité de séquençage ? Pour un dépistage en population ?