

HOW CAN WE DEFINE A BIOMARKER - ACADEMIC PERSPECTIVE

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ISTITUTO NAZIONALE PER LO STUDIO E LA CURA DEI TUMORI
FONDAZIONE "G. Pascale" – NAPOLI

SC Biologia Cellulare e Bioterapie

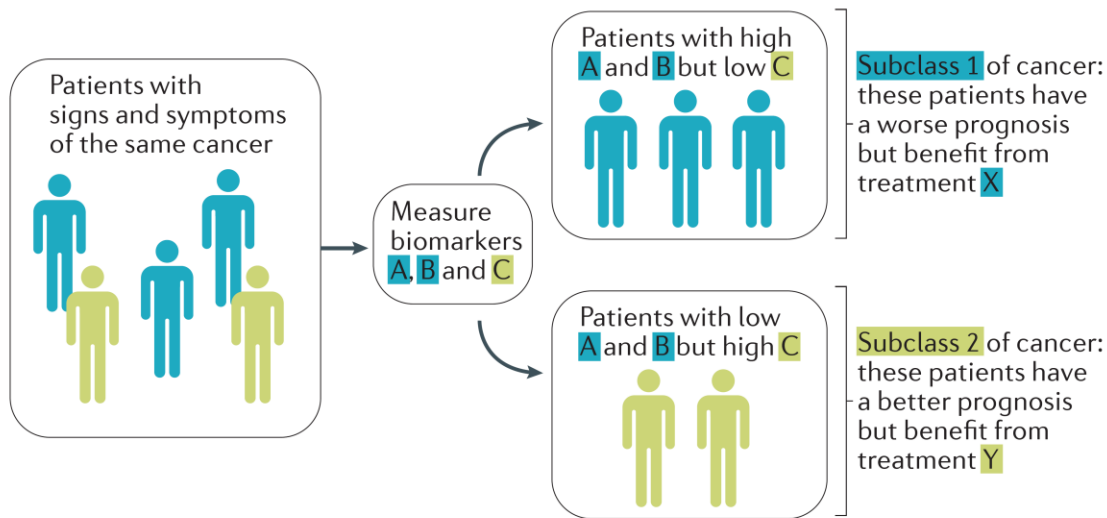
DISCLOSURES

- **Personal financial interests (speaker's fee and/or advisory boards):** MSD, Bayer, Biocartis, Illumina, Incyte, Roche, BMS, MERCK, Thermofisher, Astrazeneca, Eli Lilly, Novartis
- **Institutional financial interests (financial support to research projects):** MERCK, Thermofisher, QIAGEN, Roche, Astrazeneca, Biocartis, Illumina
- **Non-financial interests:** President, International Quality Network for Pathology (IQN Path); Past President, Italian Cancer Society (SIC)

What are cancer biomarkers?

- **A cancer biomarker is a characteristic of the tumor that can be objectively measured and evaluated (i.e. the levels of a protein, a sequence of DNA etc.)**
- **Biomarkers can be detected in the tumor tissue or in the blood**
- **Cancer biomarkers can provide important information for:**
 - ✓ **The diagnosis of the tumor and of its recurrence**
 - ✓ **The definition of the prognosis (i.e. may indicate the aggressiveness of the tumor)**
 - ✓ **The prediction of toxicity of therapeutics**
 - ✓ **The prediction of response to specific drugs**

Patients with the same signs and symptoms of cancer often have different outcomes



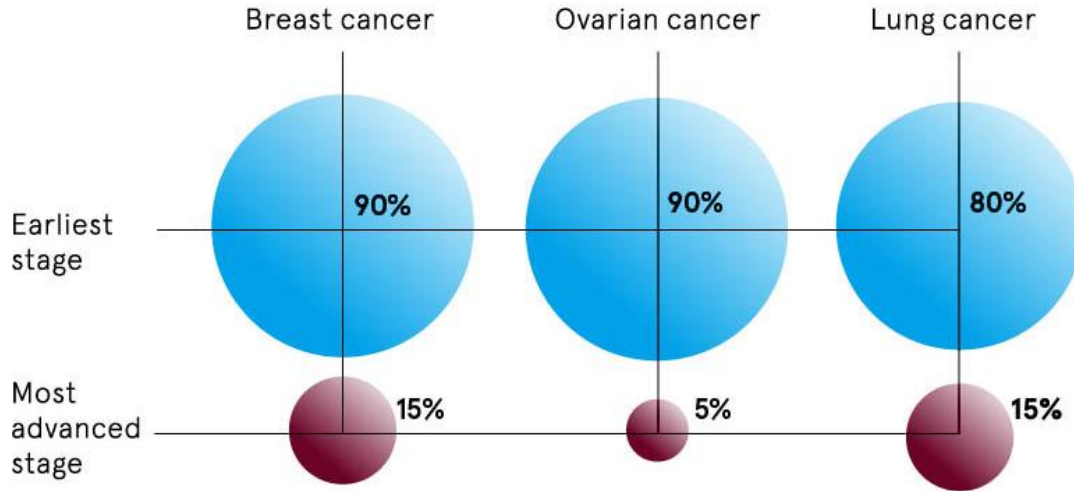
Biomarker testing is an important part of precision oncology, in which disease treatment are tailored to the characteristics of any individual patient

Clinical applications of cancer biomarkers

- **Early diagnosis**
- **Prognosis**
- **Response to therapy**

Impact of early detection of cancer

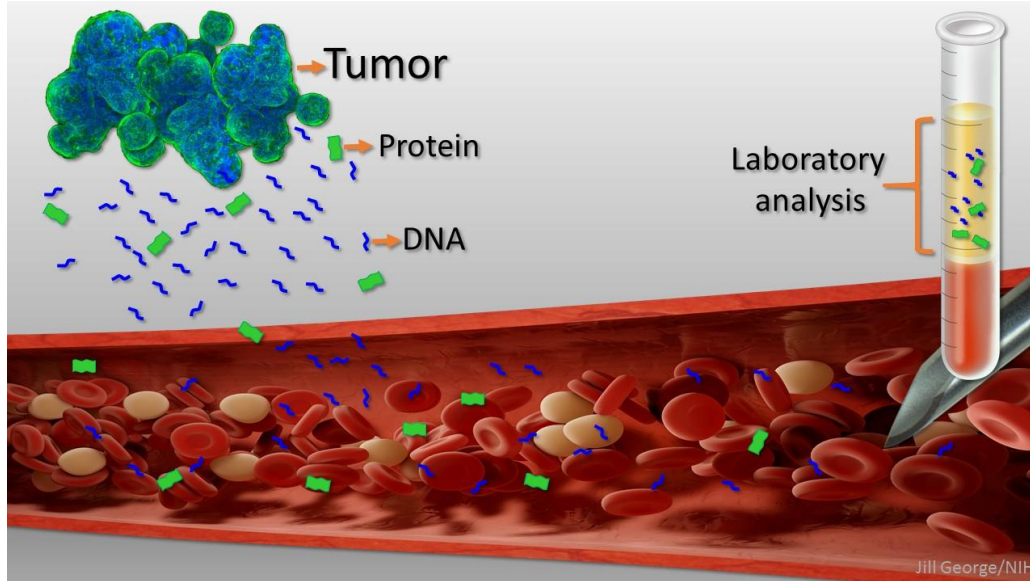
Survival rates if disease is diagnosed at earliest and most advanced stage



Cancer Research UK 2018

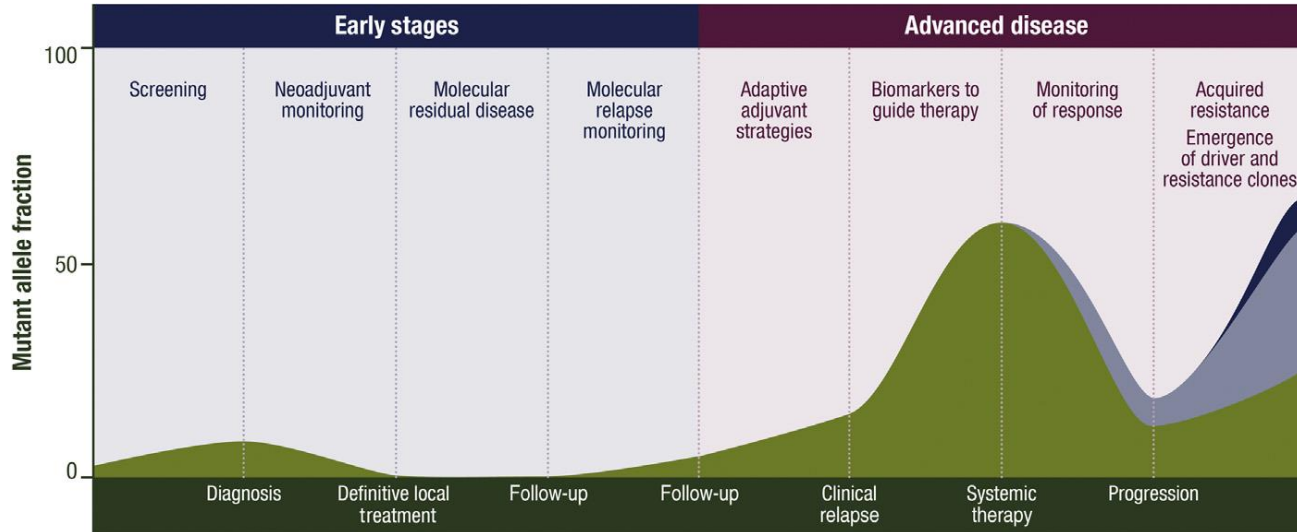
- Screening programs are available only for breast, cervical and colon cancer and for lung cancer (only for high-risk individuals in some counties)
- We need easy, non-invasive methods for early detection of different tumor types

Liquid biopsy for early cancer detection

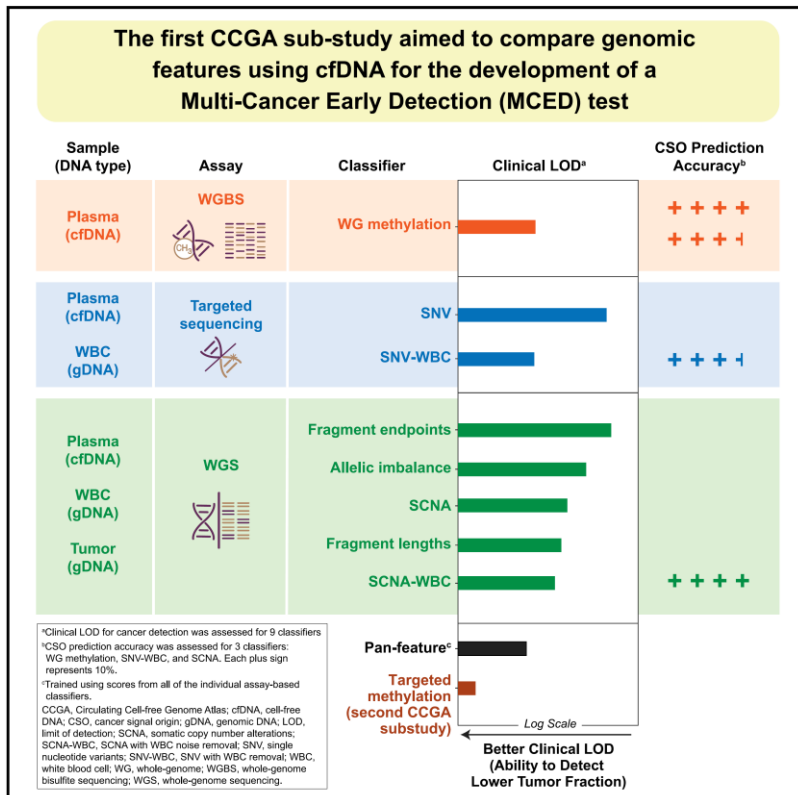


- Liquid biopsy is a generic term that refers to the possibility to identify biomarkers in bodily fluids and particularly in blood
- Different types of biomarkers can be detected in blood, including proteins, circulating tumor DNA (ctDNA) and circulating tumor cells (CTC)
- ctDNA testing is emerging as an adequate biomarker for early cancer detection

Clinical applications of ctDNA assays for patients with cancer and expected DNA levels



Evaluation of cell-free DNA approaches for multicancer early detection



Whole-genome (WG) methylation, single nucleotide variants with paired white blood cell background removal, and combined scores from classifiers evaluated in this study show the highest cancer signal detection sensitivities

DETECT-A: Assessment of clinical outcomes for participants following true positive and false positive test results



[§]Data is based on electronic medical records reviews and last documented follow-up with a healthcare provider. Patients to be followed through to a 5-year survey.

[†]N=98 Further evaluation in a large-scale pivotal study is needed to confirm the effectiveness of the imaging-based diagnostic resolution pathway.

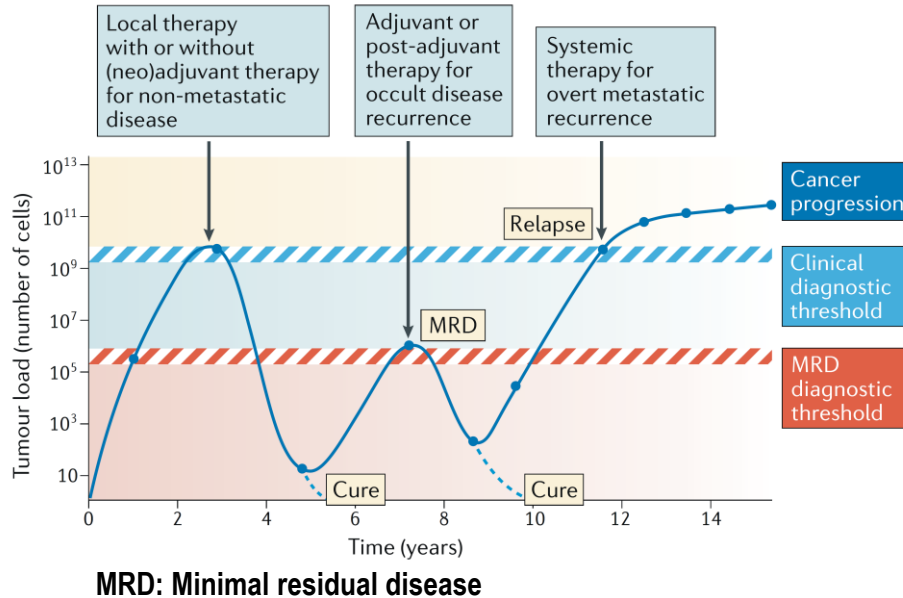
References: 1. Buchanan, et al. Presented at the ASCO Annual Meeting in Chicago, IL on June 3, 2023. 2. Lennon, et al. Presented at the ASCO Annual Meeting in Chicago, IL on June 3, 2023. 3. Data on file.

Disclaimer: This technology is under development. The features above describe current development goals. It has not been cleared or approved by the US FDA or any other national regulatory authority.

Clinical applications of cancer biomarkers

- Early diagnosis
- **Prognosis**
- Response to therapy

Therapeutic strategies depending on tumour burden in patients with cancer



- Some patients with early cancer are cured by surgery alone
- The presence of minimal residual disease (MRD) is associated with a high risk of disease recurrence
- The identification of MRD can therefore allow the adoption of a therapeutic strategy based on the level of risk of the individual patient

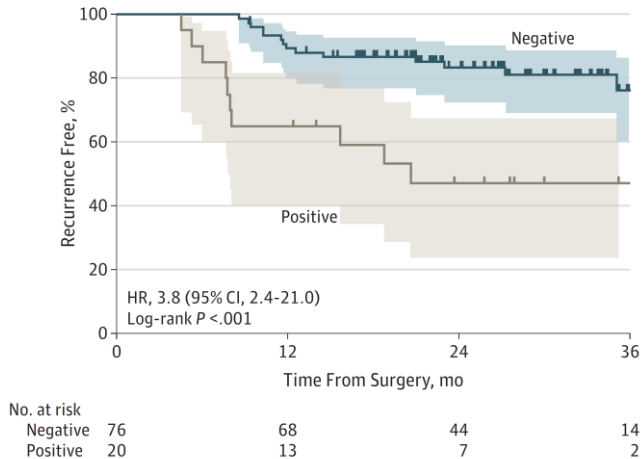
MRD detection by ctDNA testing – CRC

Tumor type	Pts (screened)	Stage	Time point	Method	HR RFS	Author, year
CRC	230 (231)	II	4-10 w post-surgery	Safe-SeqS	18	Tie, 2016
CRC	96	III	32-52 d post-surgery	Safe-SeqS	3.8	Tie, 2019
CRC	130	I-III	30 d post-surgery	Signatera	7.2	Reinert, 2019
CRC	132 (155)	I-III	6-8 w post-surgery	dPCR	11.6	Tarazona, 2019
CRC	84 (103)	I-IV	1 mo after therapy	LUNAR-1	11.28	Parikh, 2021
CRC	59 (485)	II-III	4–10 w post-surgery	Safe-SeqS	7.56	Tie, 2020
CRC	240	II-III	4–10 w post-surgery	Safe-SeqS	5.66	Chen, 2021
CRC	250	II	4–10 w post-surgery	LUNAR-1	18	Masfarré, 2021
CRC	160	III	4-8 w post-surgery	Signatera	7	Henriksen, 2022

MRD detection: open issues

Prognostic value of postoperative ctDNA in CRC patients

A Postoperative ctDNA

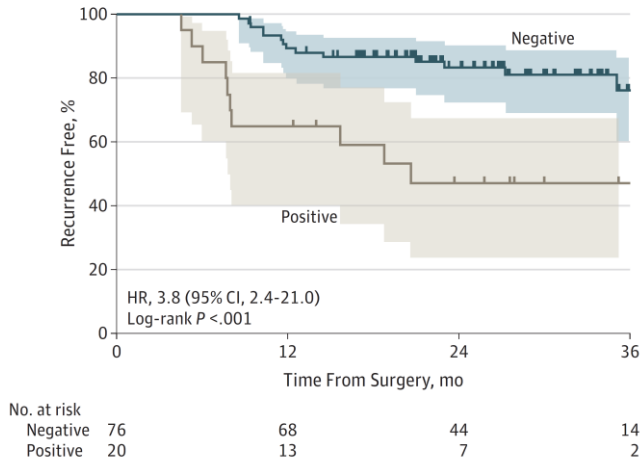


MRD: Minimal residual disease

MRD detection: open issues

Prognostic value of postoperative ctDNA in CRC patients

A Postoperative ctDNA

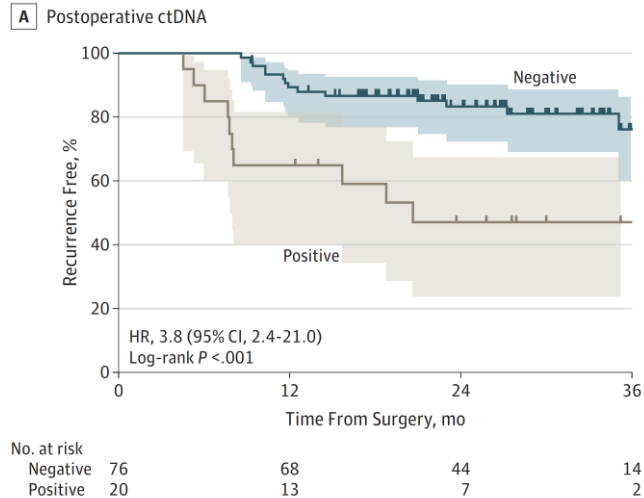


- Almost all MRD positive patients have a disease recurrence
- Interventional studies are needed to develop effective therapeutic strategies to eradicate MRD

MRD: Minimal residual disease

MRD detection: open issues

Prognostic value of postoperative ctDNA in CRC patients

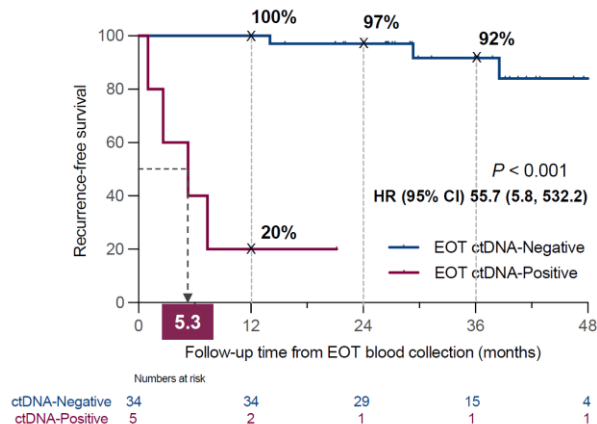


MRD: Minimal residual disease

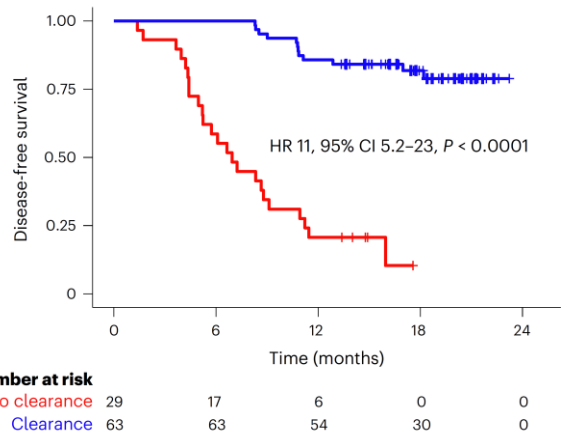
- A fraction of MRD negative patients still has a disease recurrence
- It is not clear whether this is due to limited sensitivity of the assays or to biological features of the tumor
- In MRD negative patients, recurrence is more frequently locoregional or in lymph nodes or intracranial

- Almost all MRD positive patients have a disease recurrence
- Interventional studies are needed to develop effective therapeutic strategies to eradicate MRD

ctDNA clearance after adjuvant chemotherapy



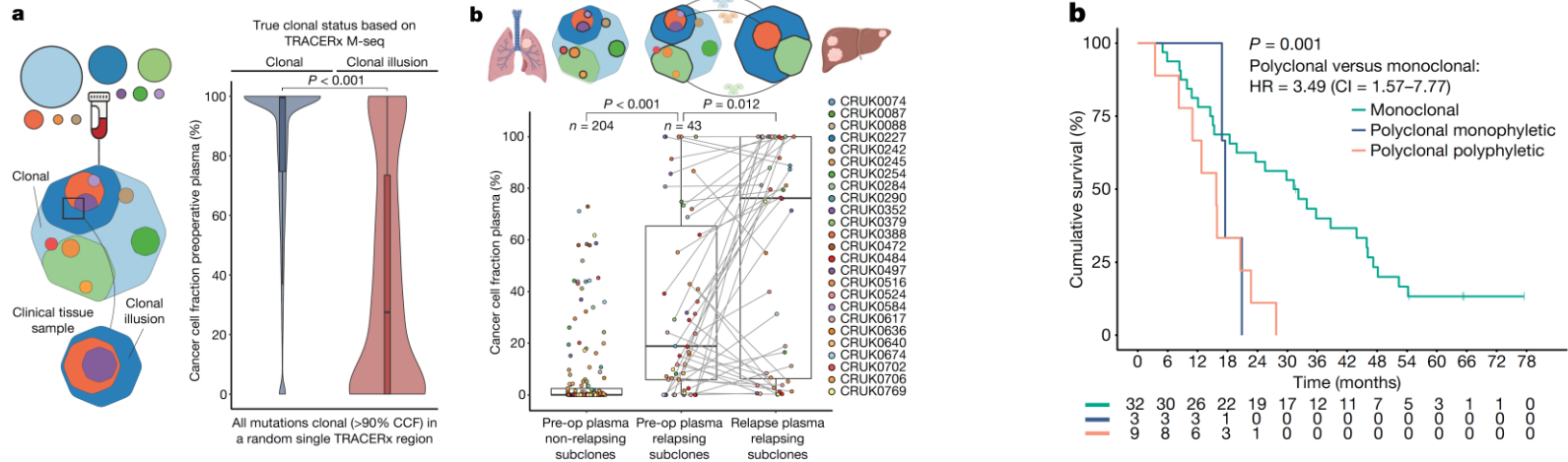
Tie ESMO Congress 2022



Kotani Nat Med 2023

- ctDNA clearance after adjuvant chemotherapy is emerging as an early biomarker of response to treatment
- Opportunity to improve the chance of cure by testing new treatments in patients with no clearance of the ctDNA
- Early biomarker of activity of new drugs in clinical trials of adjuvant therapy?

Clonality in pre- and post-operative plasma from early NSCLC patients



Clinical applications of cancer biomarkers

- Early diagnosis
- Prognosis
- **Response to therapy**

nature medicine

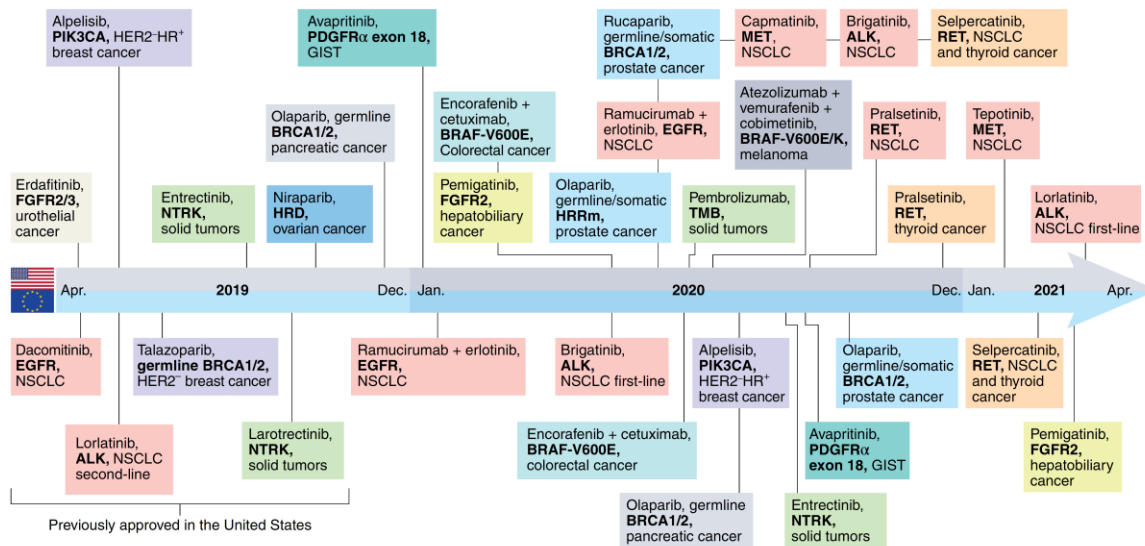
Focus on The Future of Cancer Research



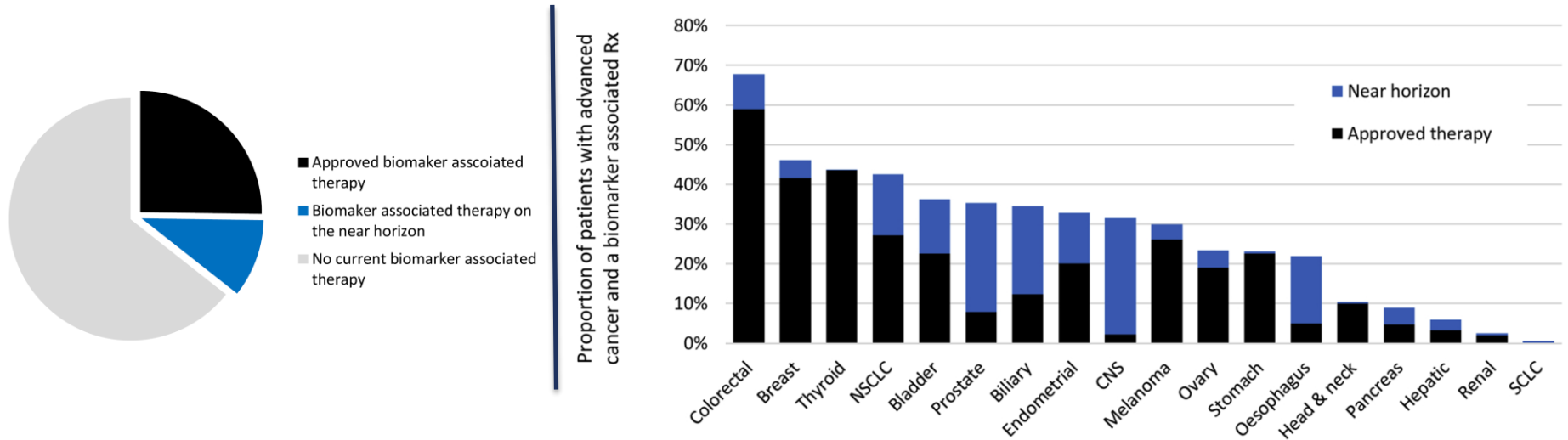
Delivering precision oncology to patients with cancer

[Joaquin Mateo](#), [Lotte Steuten](#), [Philippe Aftimos](#), [Fabrice André](#), [Mark Davies](#), [Elena Garralda](#), [Jan Geissler](#), [Don Husereau](#), [Iciar Martinez-Lopez](#), [Nicola Normanno](#), [Jorge S. Reis-Filho](#), [Stephen Stefani](#), [David M. Thomas](#), [C. Benedikt Westphalen](#) & [Emile Voest](#) ✉

Nature Medicine 28, 658–665 (2022) |



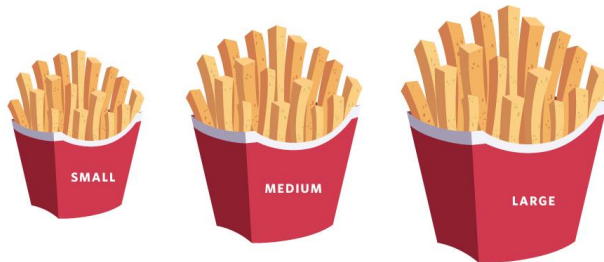
Advanced cancer patients eligible for a biomarker associated therapy



Which NGS panel fits better in my workflow?

SMALL PANELS (<50 genes)

- Specific panels by tumor histotype
- Limitations of pooling samples from patients with various diseases
- Complex workflow to manage
- Need to validate several panels (IVDR?)



LARGE PANELS (>300 genes)

- Comprehensive genomic profiling that covers all actionable mutations
- Provides information on complex biomarkers
- Costs increased
- Considerable complexity of data interpretation

MEDIUM PANELS (50-100 genes)

- Single panel covering all clinical needs
- Single workflow with the possibility of pooling multiple samples and reducing costs
- Opportunity to validate a single panel to respond to multiple needs

Characteristics of patients enrolled in RATIONAL

- **Pathway A:** Patients with advanced solid tumors who already received a NGS test in clinical practice or clinical trials. In Pathway A, 70.6% tumor or liquid biopsy tests were performed with NGS panels covering ≤52 genes
- **Pathway B:** Patients with no NGS data available were tested with a comprehensive genomic profiling assay
- In pathway A, at the time of testing 98/218 patients (45.0%) did not receive any treatment for metastatic disease, whereas 103/218 (47.3 %) had at least one line of therapy, with a maximum of five
- In pathway B, patients with no previous therapy were 112/520 (21.5%), while 404/520 (77.7%) received up to a maximum of 11 treatments before testing

Characteristics	All, n = 738	Pathway A, n = 218	Pathway B, n = 520
<i>Age, years, median (range)</i>	63 (18–87 years)	66 (18–85 years)	62 (18–87 years)
<i>Sex</i>			
Male	391 (53.0%)	109 (50.0%)	282 (54.2%)
Female	347 (47.0%)	109 (50.0%)	238 (45.8%)
<i>Lines of previous therapy</i>			
0	210 (28.5%)	98 (45.0%)	112 (21.5%)
1	195 (26.4%)	44 (20.2%)	151 (29.1%)
≥2	312 (42.3%)	59 (27.1%)	253 (48.6%)
NA	21 (2.9%)	17 (7.8%)	4 (0.8%)

Patients with GAs treated with matched targeted therapies

- We identified 324 GAs belonging to ESCAT levels I, II and III in 254/554 (45.8%) patients
- Overall, 67/573 (11.7%) patients, for which FU data were available, received a targeted therapy, based on the result of molecular analysis
- In Pathway A, 25 patients with actionable GAs did not receive therapy due to lack of clinical trials or other pathways of access to drugs (n. 21; 84%) or poor clinical conditions or death at the time of the report (n. 4; 16%).
- In Pathway B 16/115 patients (13.9%) died before the report was available, 32 (27.8%) died within 5 months suggesting poor clinical conditions and 67 (58.3%) did not receive targeted therapy because no drug was approved and reimbursed in Italy or clinical trials or other pathways of access to drugs were available, or patients already received suggested therapy in previous lines

ESMO recommendations for germline-focused analysis of tumour-only sequencing

Germline-focused tumour analysis can be restricted to variants of VAF >30% (SNVs) or >20% (small insertions/deletions)

Box 1. Recommendations for genes to be included for germline-focused analysis and triggering of germline sample laboratory confirmation

	Any tumour type		Associated tumour type only
Tumour arising any age	<i>BRCA1</i>	<i>RAD51C</i>	<i>FLCN</i>
	<i>BRCA2</i>	<i>RAD51D</i>	<i>FH</i>
	<i>BRIP1</i>	<i>RET</i>	<i>BAP1</i>
	<i>MLH1</i>	<i>SDHA</i>	<i>POLE</i>
	<i>MSH2</i>	<i>SDHAF2</i>	
	<i>MSH6</i>	<i>SDHB</i>	
	<i>PALB2</i>	<i>SDHC</i>	
	<i>PMS2</i>	<i>SDHD</i>	
	<i>VHL</i> ^a	<i>TSC2</i>	
		<i>MUTYH</i> ^b	
Tumour arising age <30 only	<i>RB1</i>		<i>TP53</i> ^c
	<i>APC</i>		<i>NF1</i>

- In patients analyzed with CGP tests in the RATIONAL study, potentially germline pathogenic/likely pathogenic (P/LP) variants were detected in 69/580 (11.9%) patients.
- Only 4/23 patients (17.4%), for which we had information about genetic counseling, were referred to genetics consultation after molecular testing.

SPECIAL ARTICLE

ESMO recommendations on the use of circulating tumour DNA assays for patients with cancer: a report from the ESMO Precision Medicine Working Group

J. Pascual¹, G. Attard², F.-C. Bidard^{3,4}, G. Curigliano^{5,6}, L. De Mattos-Arruda^{7,8}, M. Diehn⁹, A. Italiano^{10,11,12}, J. Lindberg¹³, J. D. Merker¹⁴, C. Montagut¹⁵, N. Normanno¹⁶, K. Pantel¹⁷, G. Pentheroudakis¹⁸, S. Popat^{19,20}, J. S. Reis-Filho²¹, J. Tie^{22,23}, J. Seoane^{24,25}, N. Tarazona^{26,27}, T. Yoshino²⁸ & N. C. Turner^{19,20*}

Tissue-based testing remains the preferred test for many cancer patients, due to limitations of ctDNA assays detecting fusion events and copy number changes, although ctDNA assays may be routinely used when faster results will be clinically important, or when tissue biopsies are not possible or inappropriate.

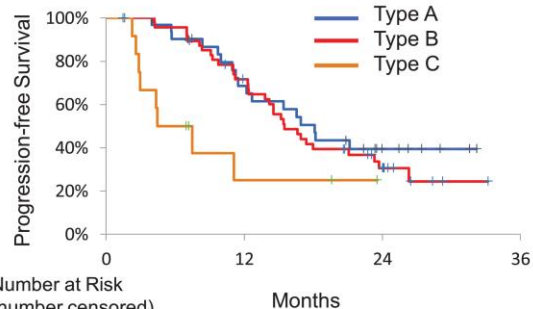
PFS of EGFR mut NSCLC patients treated with Bevacizumab/Erlotinib or Erlotinib

PFS among patients classified by plasma EGFR detection at pretreatment (P0) and at 6 weeks from the start of treatment (P1):
Type A, P0-/P1-
Type B, P0+/P1-
Type C, P0+/P1+

A

BE

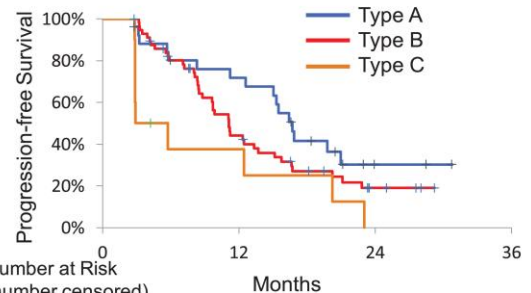
	Type A	Type B	Type C
mPFS	18.1m	15.5m	6.0m
(95%CI)	(11.5- NR)	(12.4-23.3)	(2.6-NR)
HR	reference	1.222	2.788
(95%CI)		(0.676-2.208)	(1.182-6.576)



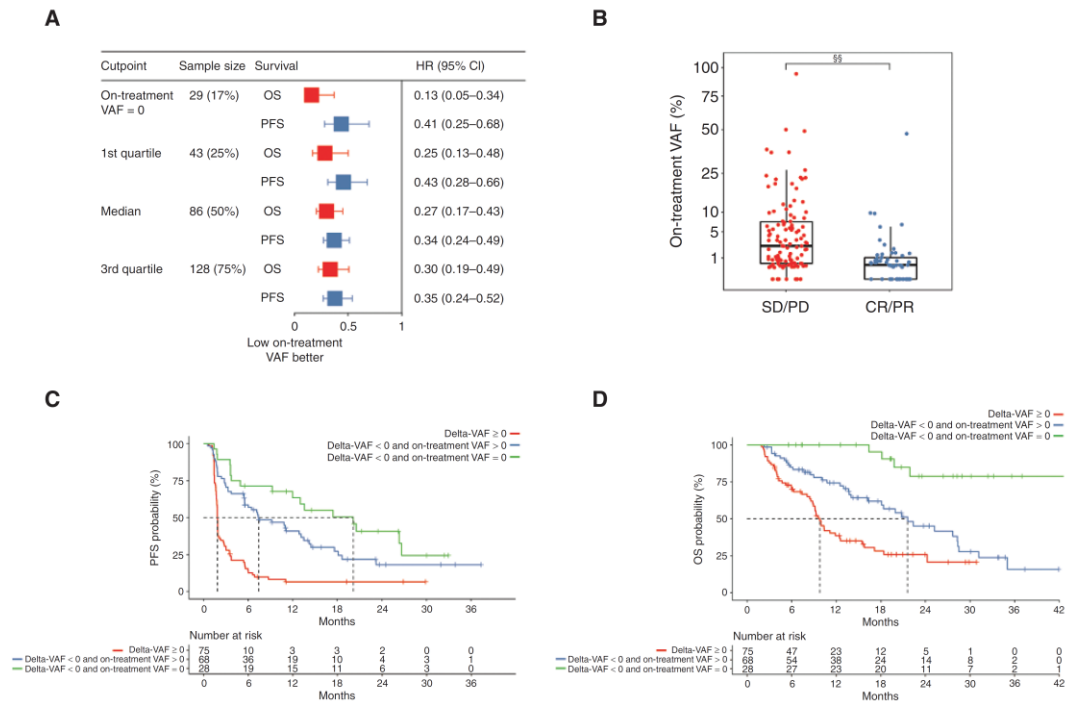
B

E

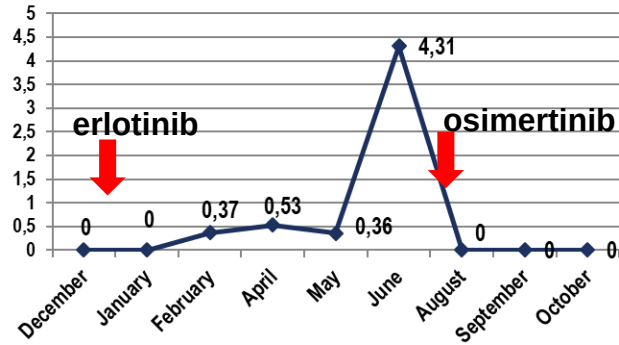
	Type A	Type B	Type C
mPFS	16.7m	11.1m	4.3m
(95%CI)	(11.2- NR)	(8.5-13.7)	(2.8-20.2)
HR	reference	1.533	2.618
(95%CI)		(0.855-2.750)	(1.141-6.006)



Dynamics of ctDNA levels and prognosis in patients treated with durvalumab $\pm$ tremelimumab



Monitoring response to EGFR TKI with cfDNA testing



July 2018



September 2018



Access to precision drugs and high quality biomarker tests in Europe

Medicines access



Single biomarker test access



Multi-biomarker test access



Test quality



Conclusions

- **Biomarkers play an important role in the diagnosis of tumors, in defining the prognosis of patients and in predicting the response to precision therapies**
- **Access to biomarkers testing must be guaranteed to all cancer patients**
- **The implementation of comprehensive genomic profiling and liquid biopsy in the diagnostic workup will:**
 - ✓ **Increase patients' chances of accessing new therapies through clinical trials**
 - ✓ **Identify families with hereditary cancer syndromes**
 - ✓ **Provide relevant information to improve the personalization and precision of therapies**



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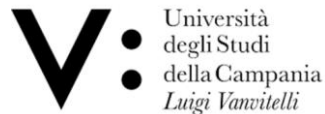
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