Sistema Socio Sanitario
Fondazione IRCCS
Istituto Nazionale dei Tumori
via Venezian, 1 20133 Milano
Regione Lombardia

ON BIOMARKER DATA QUALITY

Paolo Verderio

Unit of Bioinformatic and Biostatistics

Fondazione IRCCS Istituto Nazionale dei Tumori di Milano

CHALLENGES IN DATA-DRIVEN GENOMIC COMPUTING

Organized by the Advanced ERC Project 693174 "GeCo" – data-driven Genomic Computing

Workshop in Como, Villa del Grumello – March 6-8 2019

Outline

I. Background

- ✓ Biomarker definition
- ✓ Biomarker state of the art

II. Biomarker development process

- ✓ Biomarker Discovery
- ✓ Biomarker Validation
- ✓ Clinical Utility

III. Concluding remarks

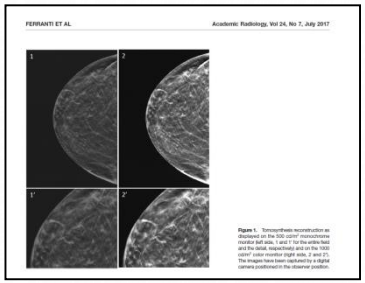
I. Background

What is a biomarker?

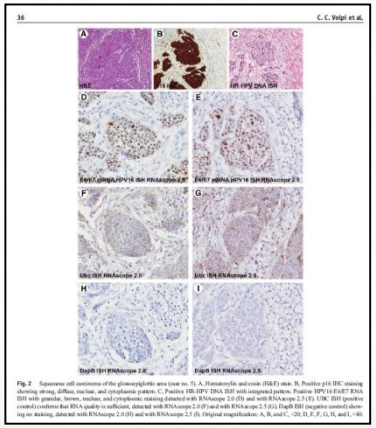
➢a defined characteristic that is measured as an indicator of normal biological processes, pathogenic processes, or responses to an exposure or intervention, including therapeutic interventions.

- ✓ molecular characteristic
- ✓ histological characteristic
- ✓ radiographical characteristic
- ✓physiological characteristic
- ✓ ....

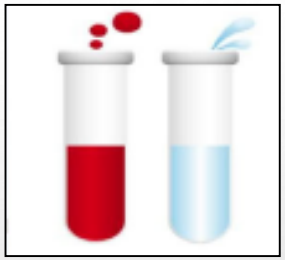
types of biomarkers



Ferranti C, et al. Acad Radiol. 2017, 24:795–801



Volpi CC, et al. Hum pathology, 2017, 74:32-42.



https://www.ncbi.nlm.nih.gov/books/NBK338448/#IX-B

Hayes DF. Biomarker validation and testing. Molecular Oncology. 2015(9): 960-966

What is a tumor biomarker?

might be a:

- molecular change (i.e. nucleic acid, protein, or metabolite) OR
- process change (i.e. alteration in tissue or image appearance)

Can be detected in:

- ✓ tissue
- ✓ blood
- ✓ secretions (i.e. urine, stool, sputum, or breast nipple aspirates, CSF, etc)

Hayes DF. Biomarker validation and testing. Molecular Oncology. 2015(9): 960-966
Kisser A. Issues and challenges in the systematic evaluation of biomarker tests. Precision in Medicine. 2018

What is a biomarker test?

- ✓ a test used to identify or measure the perturbation reflected by the tumor biomarker

- ✓ must be applicable in clinical routine and reproducible and accurately translate the biomarker into a measurement parameter

Hayes DF. Biomarker validation and testing. Molecular Oncology. 2015(9): 960-966
Kisser A. Issues and challenges in the systematic evaluation of biomarker tests. Precision in Medicine. 2018

Type of biomarker (1)

Diagnostic biomarker

Pharmacodynamic response biomarker

Prognostic biomarker

Predictive biomarker

Susceptibility risk biomarker

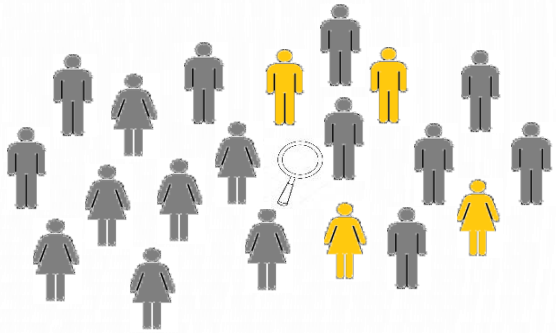
Monitoring biomarker

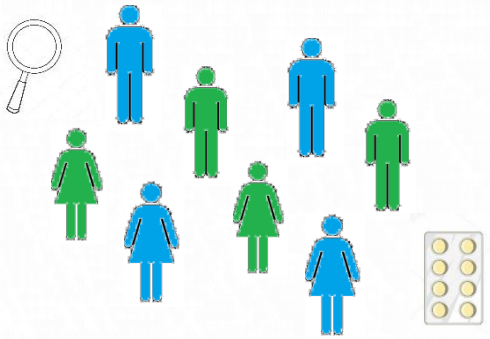
Safety biomarker

FDA-NIH Biomarker Working Group. BEST (Biomarkers, EndpointS, and other Tools) Resource [Internet]. Silver Spring (MD): Food and Drug Administration (US); 2016-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK326791/ Co-published by National Institutes of Health (US), Bethesda (MD).

Type of biomarker (2)

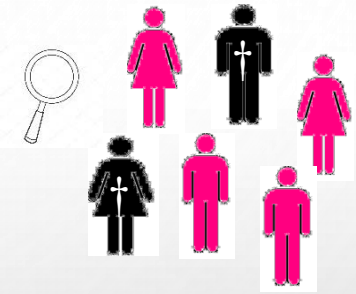
Diagnostic: "A biomarker used to detect or confirm presence of a disease or condition of interest or to identify individuals with a subtype of the disease."





Predictive: "A biomarker used to identify individuals who are more likely than similar individuals without the biomarker to experience a favorable or unfavorable effect from exposure to a medical product or an environmental agent."

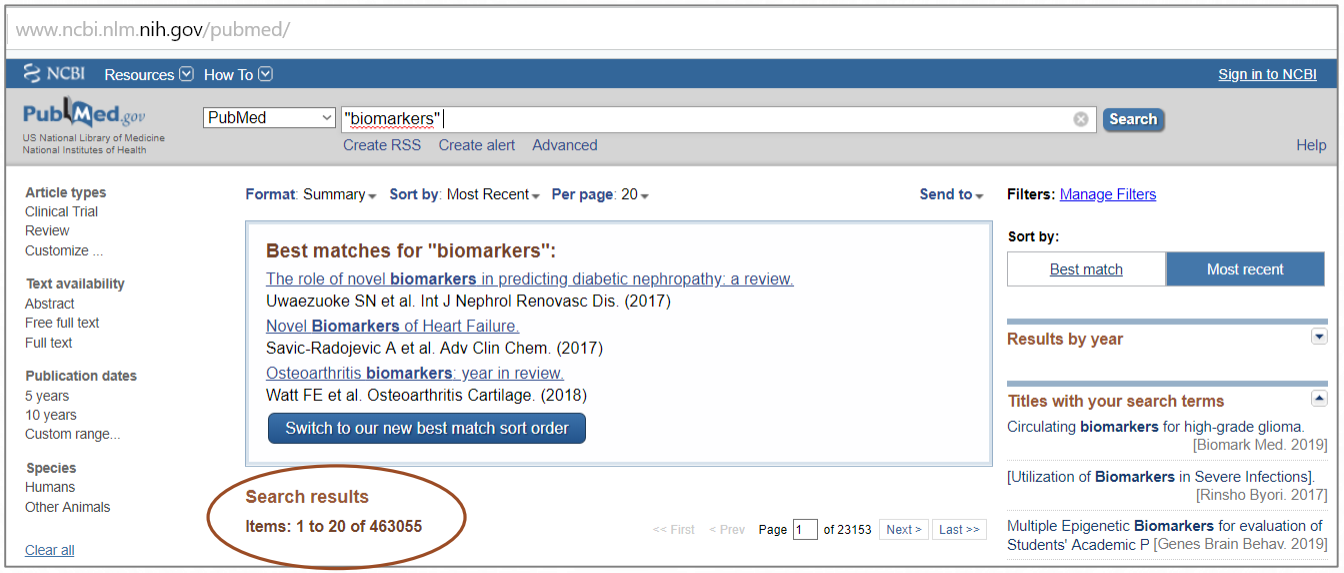
Prognostic: "A biomarker used to identify likelihood of a clinical event, disease recurrence or progression in patients who have the disease or medical condition of interest."



FDA-NIH Biomarker Working Group. BEST (Biomarkers, EndpointS, and other Tools) Resource [Internet]. Silver Spring (MD): Food and Drug Administration (US); 2016-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK326791/ Co-published by National Institutes of Health (US), Bethesda (MD).

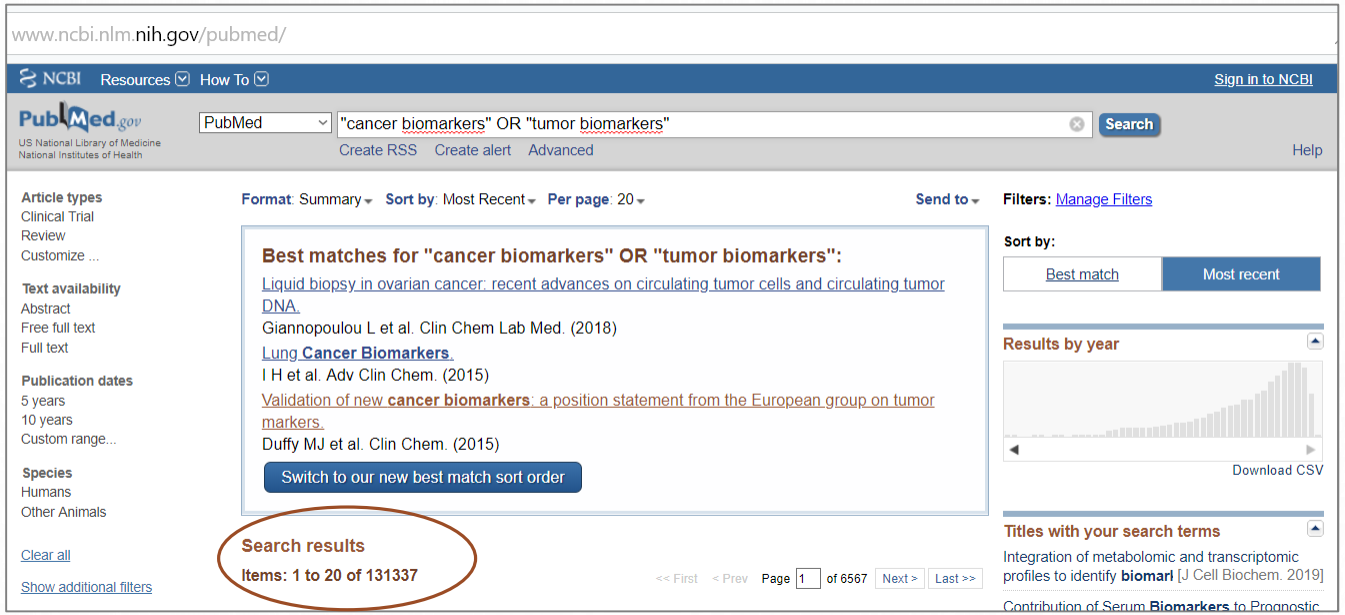
Biomarkers state of the art (1)

PubMed Central® (PMC) is a free full-text archive of biomedical and life sciences journal literature at the U.S. National Institutes of Health's National Library of Medicine (NIH/NLM) (https://www.ncbi.nlm.nih.gov/pmc/)



huge number of research studies focused on biomarkers in many different field and specimen

Biomarkers state of the art (2)

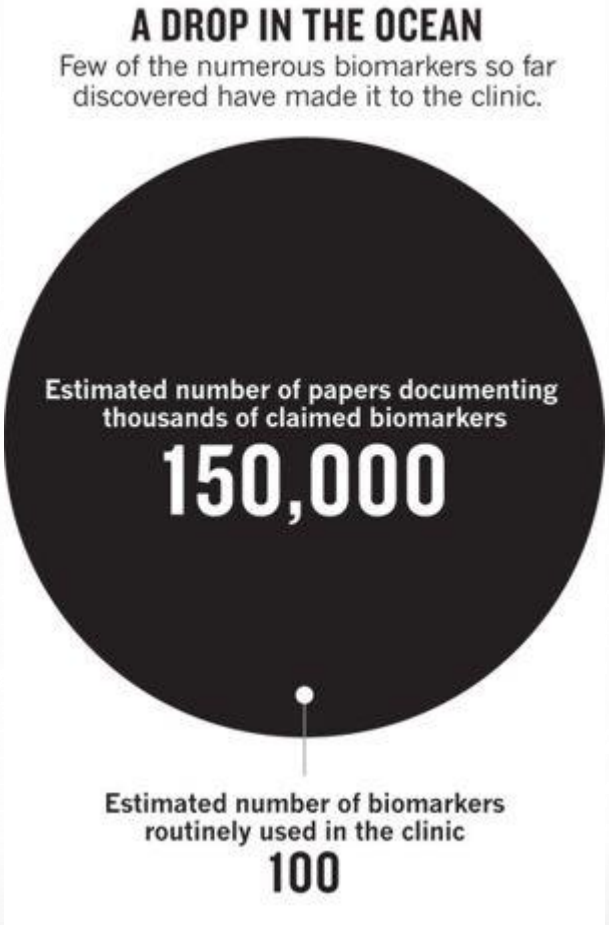


~30% are focused "*oncological*" biomarkers

However....

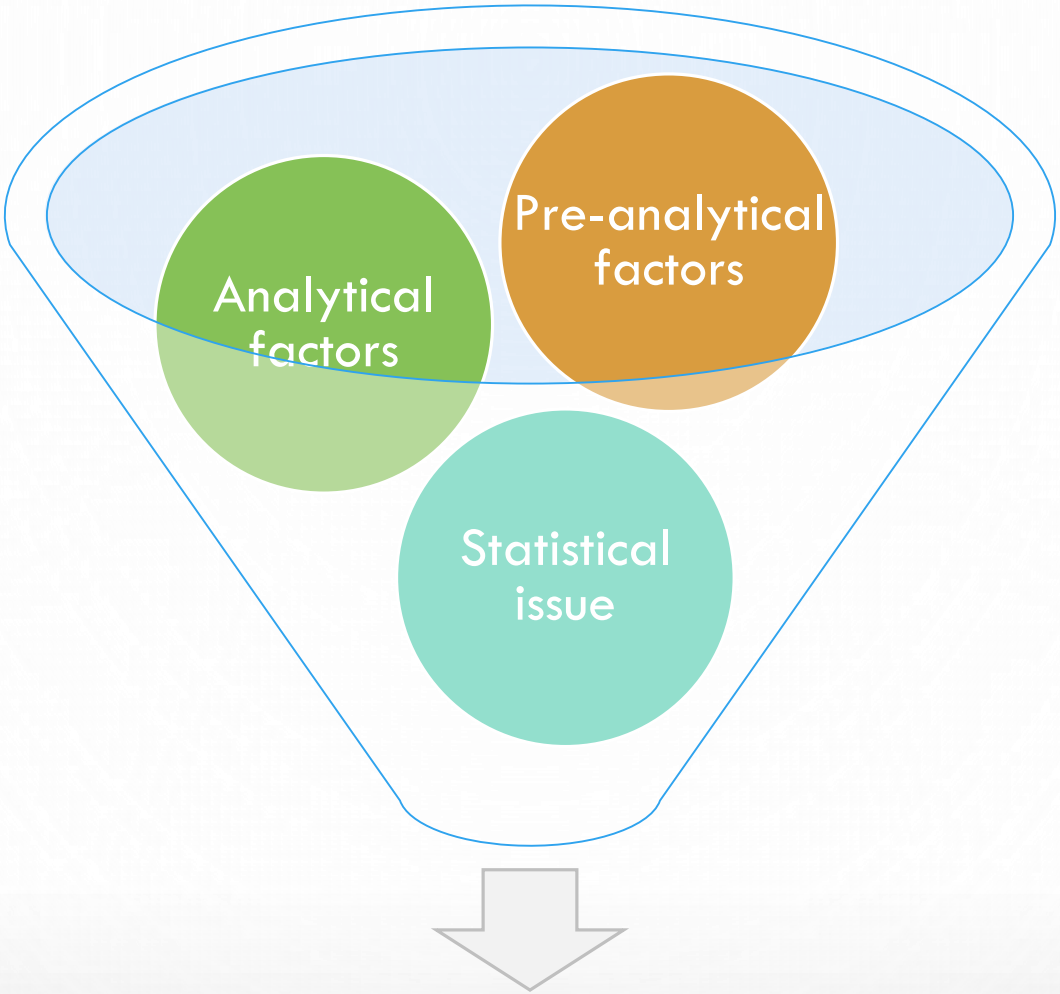
- a limited number of biomarkers is really translated and fully applied in the real clinical practice
- very few discovered biomarkers are used in the routine clinical setting

Why?



Poste G. Nature 2011;469:156-157

Challenges in biomarker development (1)



Challenges in biomarker development (2)

Pre-analytical factors

- ✓ starting material
- ✓ sample collection and processing
- ✓ sample storage conditions
- ✓ type of extraction method
- ✓ ...

Analytical factors

- ✓type of detection method
- ✓ type of throughput
- ✓ type of commercial platform
- ✓ ...

Statistical analysis

- ✓data normalization
- ✓level and type of analysis
- ✓ type of predictor
- ✓methods for multivariate model building
- ✓ ...

Challenges in biomarker development (3)

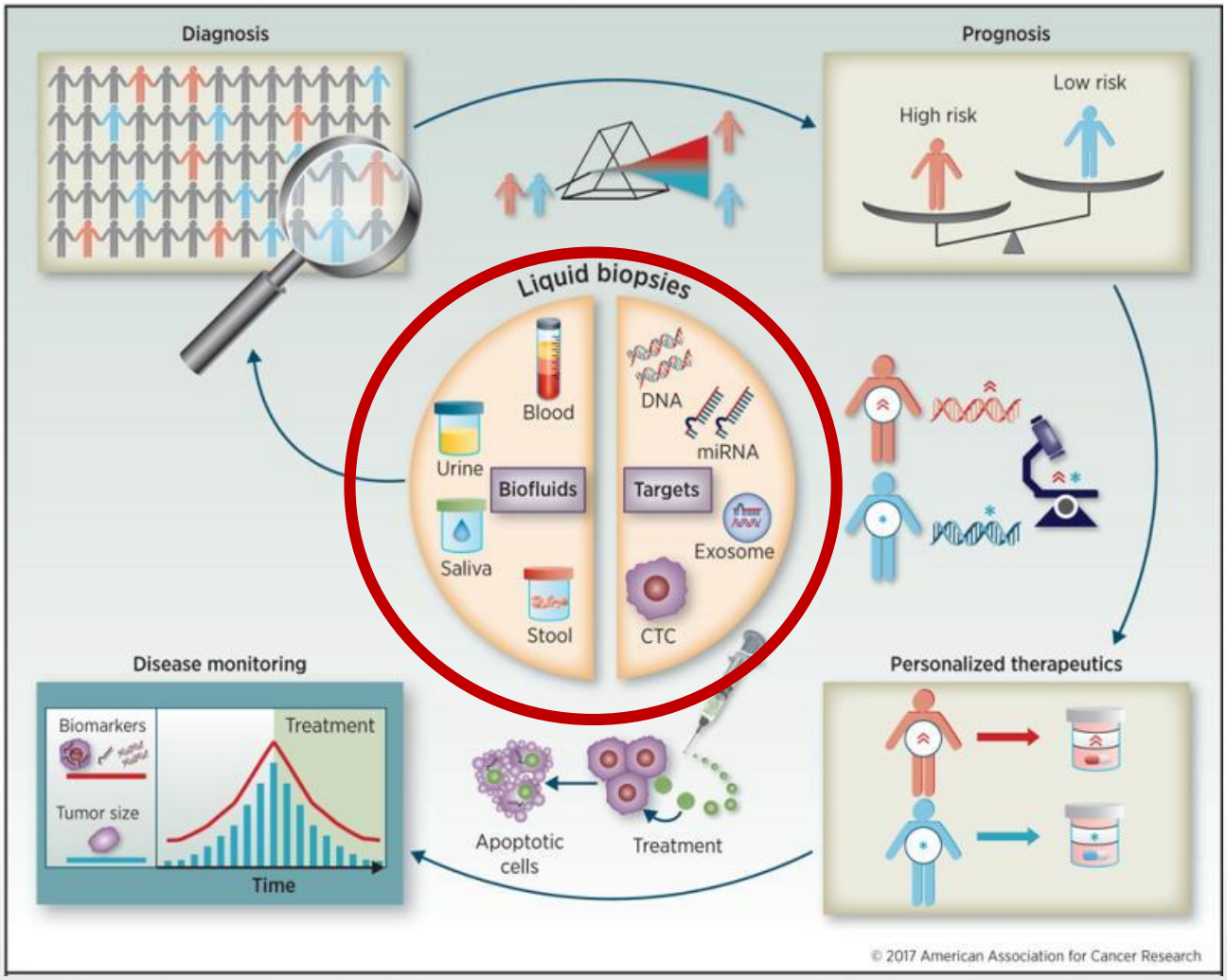
- Molecular features measurable from **body fluids (i.e. Liquid biopsies)**

  → a great opportunity to develop non invasive biomarkers

- Availability of **high-throughput technologies**

  → new diagnostic and therapeutic perspective especially in the oncological scenario

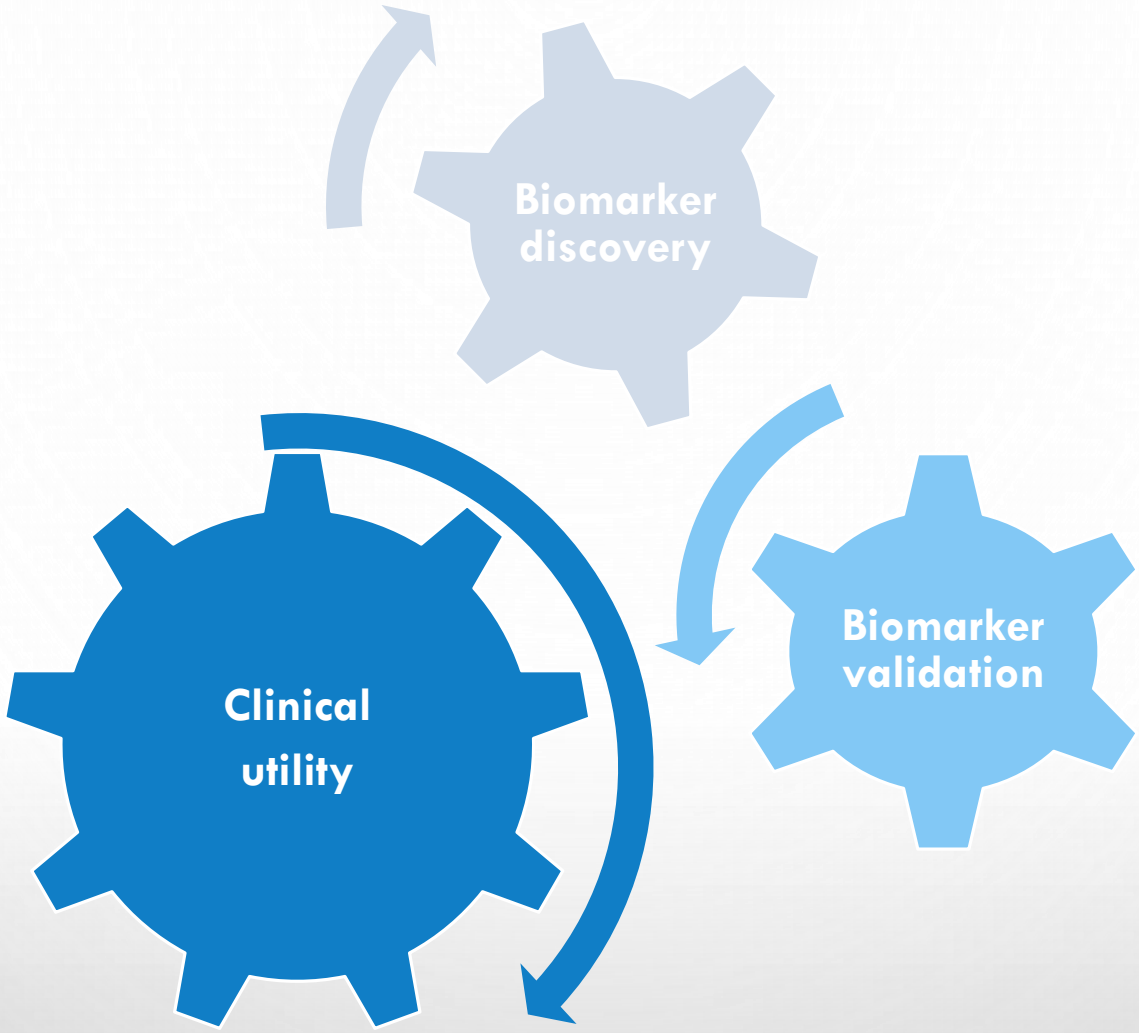
Challenges in biomarker development (3)



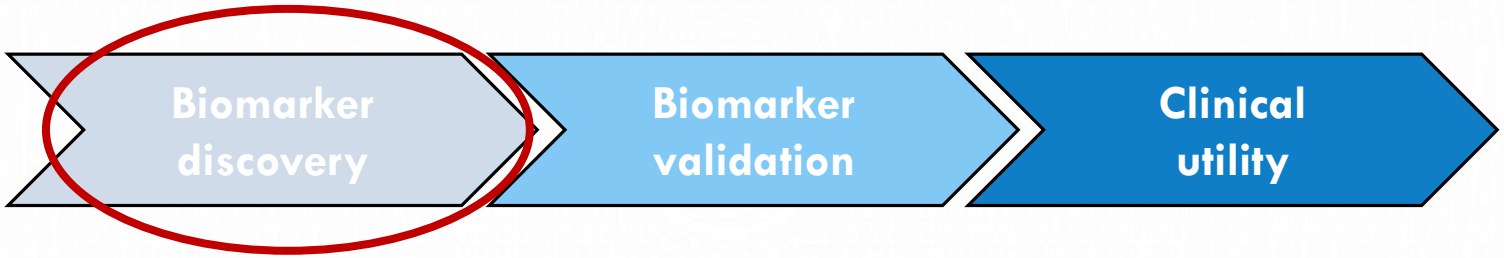
Shigeyasu K, Toden S, Zumwalt TJ, Okugawa Y, Goel A.Clin Cancer Res..2017.23(10):2391-2399.

II. Biomarker development process

Comprehensive evaluation of biomarker development process

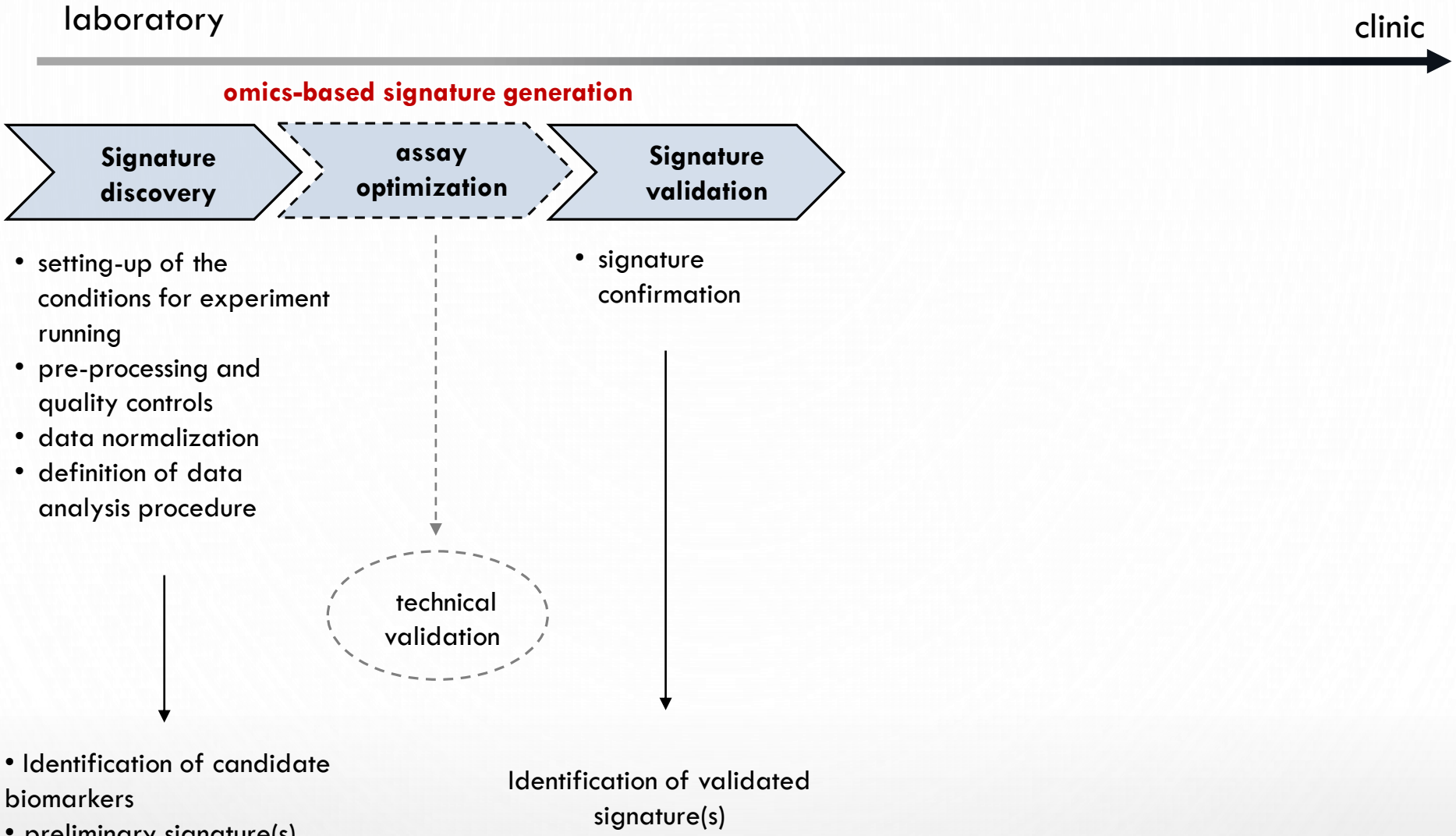


Biomarker discovery (1)



1. **identification of the context of use [study planning]**
   - the fundamental use of the biomarker
   - the target population (with inclusion and exclusion criteria)
   - specific clinical context
   - …
2. **setting-up of the conditions for experiment running [data generation]**
   - sample specimen and collection (e.g. collection tubes, timing of collection)
   - sample processing (e.g. timing of processing, temperature of storage)
   - sample analysis (e.g. type of extraction, platform)
   - …
3. **biomarker identification/selection [data analysis]**
   - pre-processing and quality controls
   - definition of data analysis procedure
   - identification of candidate biomarker
   - development of omics-based signatures (e.g. statistical model)
   - …

Biomarker discovery (2)



Adapted from Verderio P et al. Br J Cancer. 2016;115:1-4.

Biomarker discovery (3)

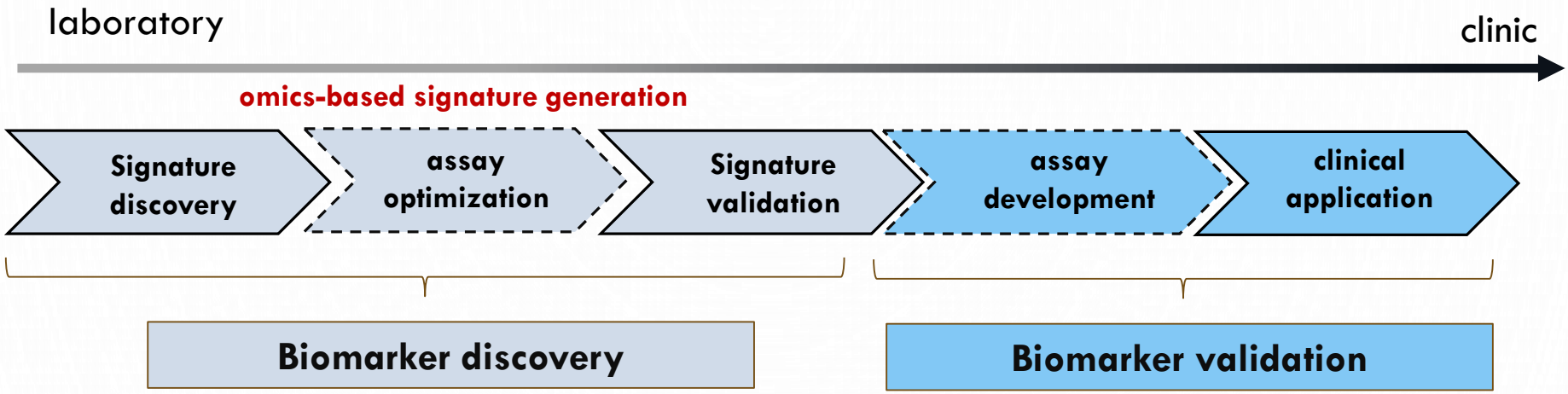
laboratory → clinic

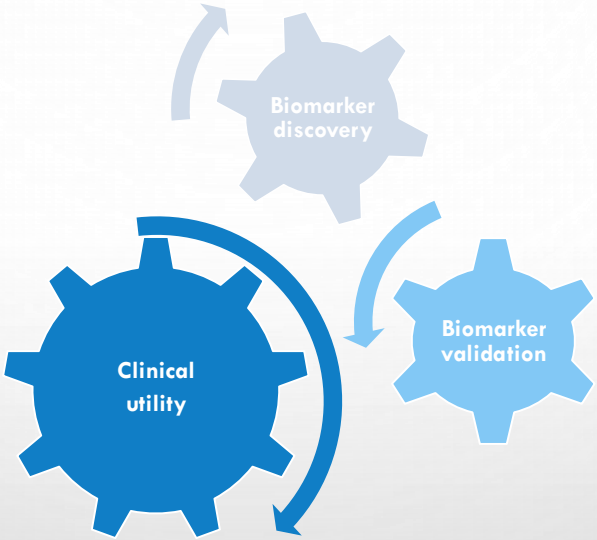
omics-based signature generation

Signature discovery → **assay optimization** → **Signature validation**

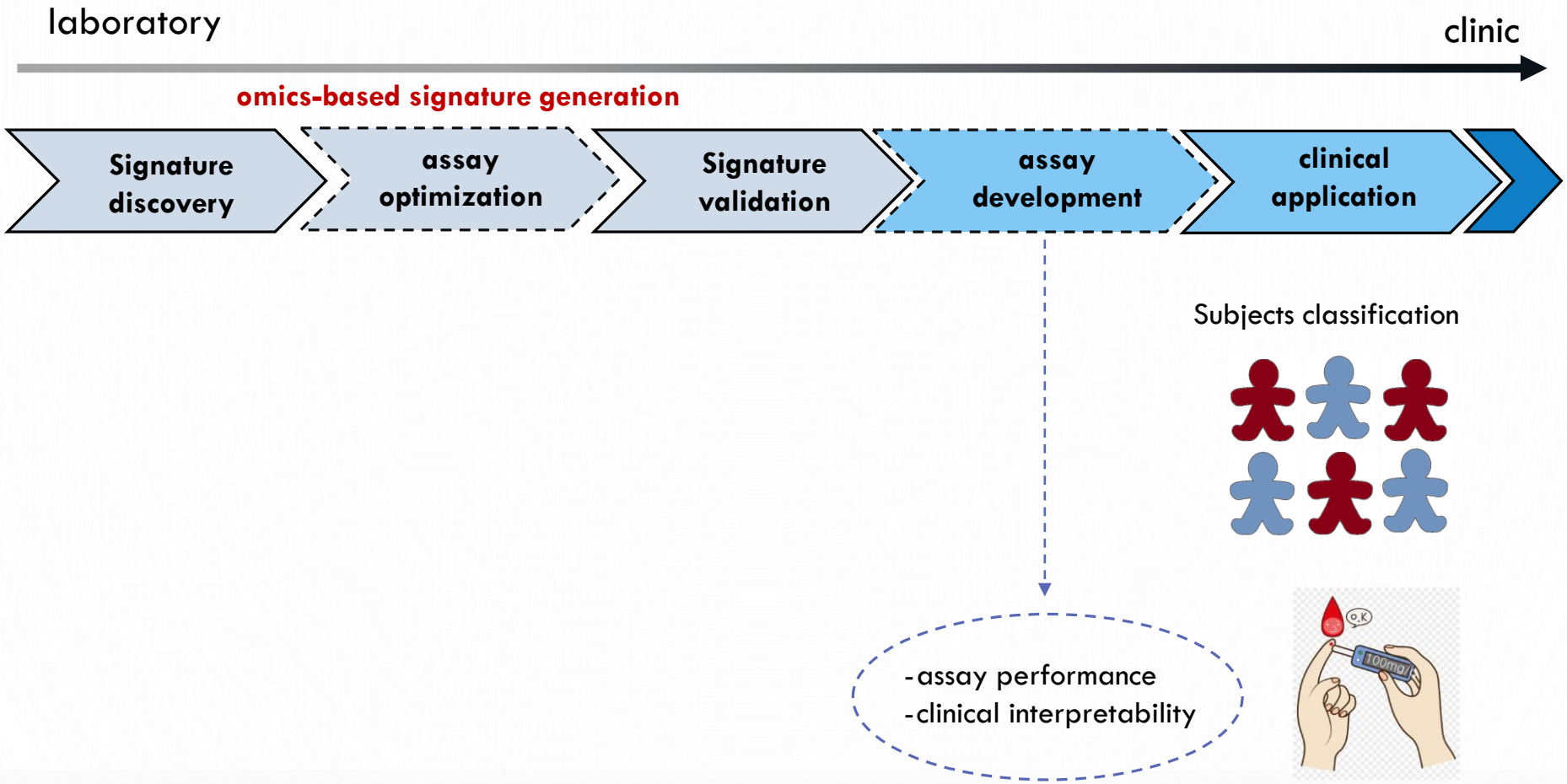
Validated omics-signature(s) = biomarkers

From biomarker discovery … to biomarker validation



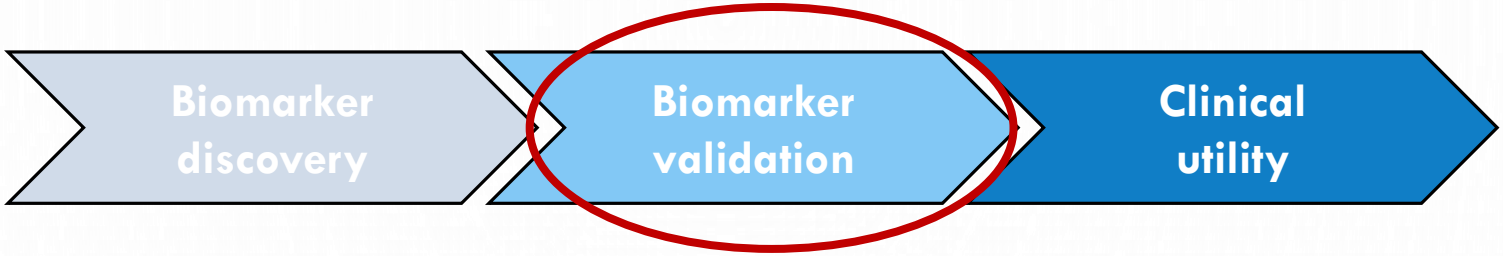


Biomarker validation (1)



Adapted from Verderio P et al. Br J Cancer. 2016;115:1-4.

Biomarker validation (2)



- ✓ **Analytical validation:** demonstrates the **robustness** (accuracy, precision, reproducibility) of the test

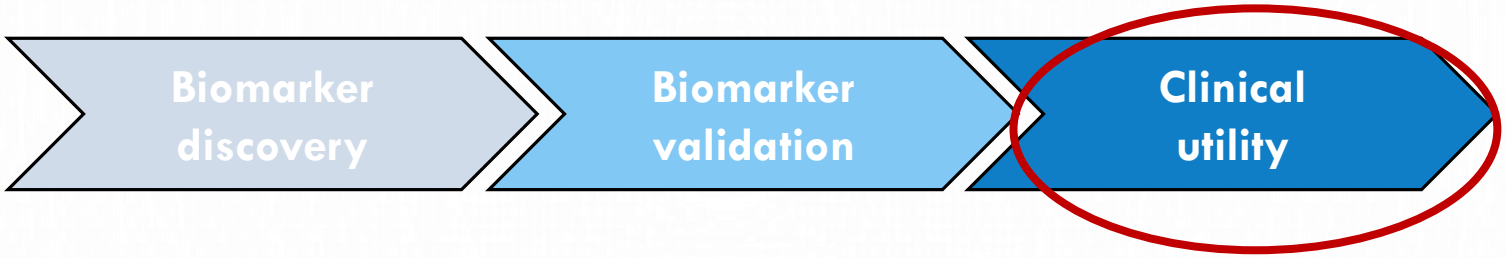
  how well does the test measure what it claims to measure?

- ✓ **Clinical Validation:** demonstrates the **effectiveness** of the test

  how relevant is the test measurement to the clinical condition?

  is the biomarker related to the clinical endpoint?

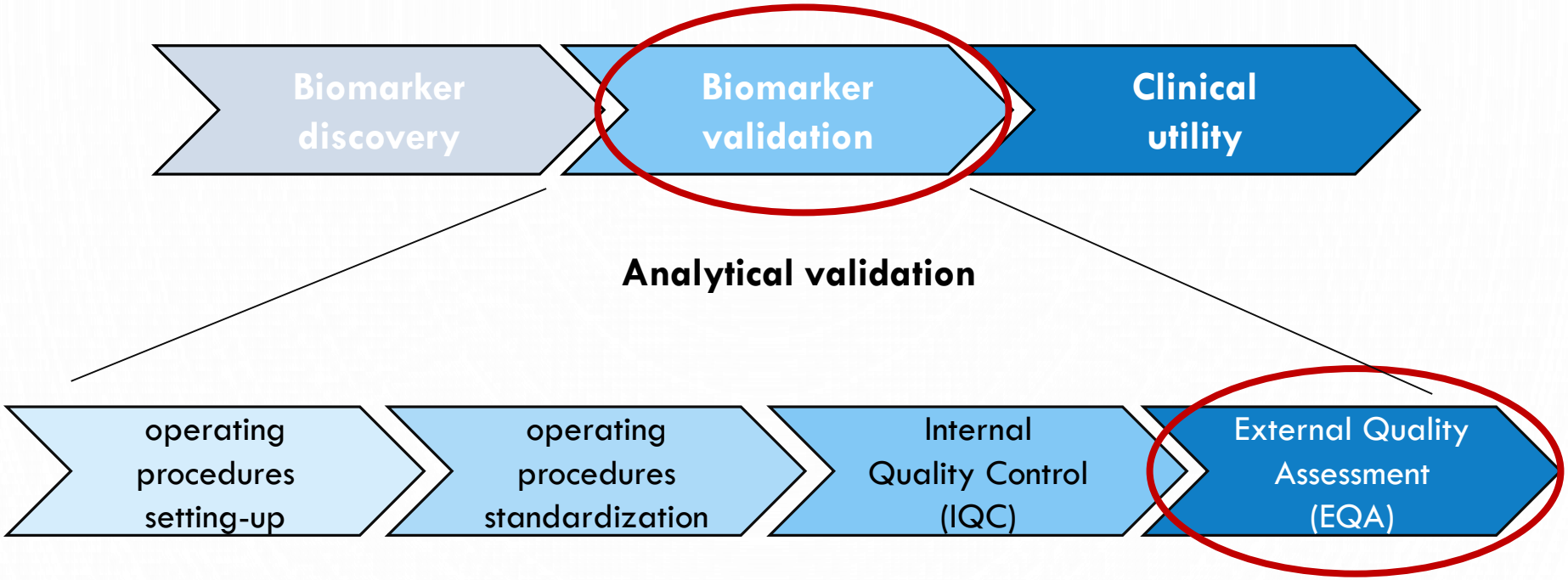
Clinical utility



The conclusion that a given use of a medical product **will lead to a net improvement in health outcome** or provide useful information about diagnosis, treatment, management, or prevention of a disease.

Does use of the biomarker result in patient benefit?

Focus on the analytical validation



Verderio P et al. New Biotechnology 2019 (submitted)

External Quality Assessment (EQA)

Purpose of EQAs:

(a) evaluation of laboratory performance for specific tests and its continuous monitoring

(b) identification of inter-laboratory differences

(c) evaluation of method/diagnostic system performances

(d) degree of comparability between methods/diagnostic systems

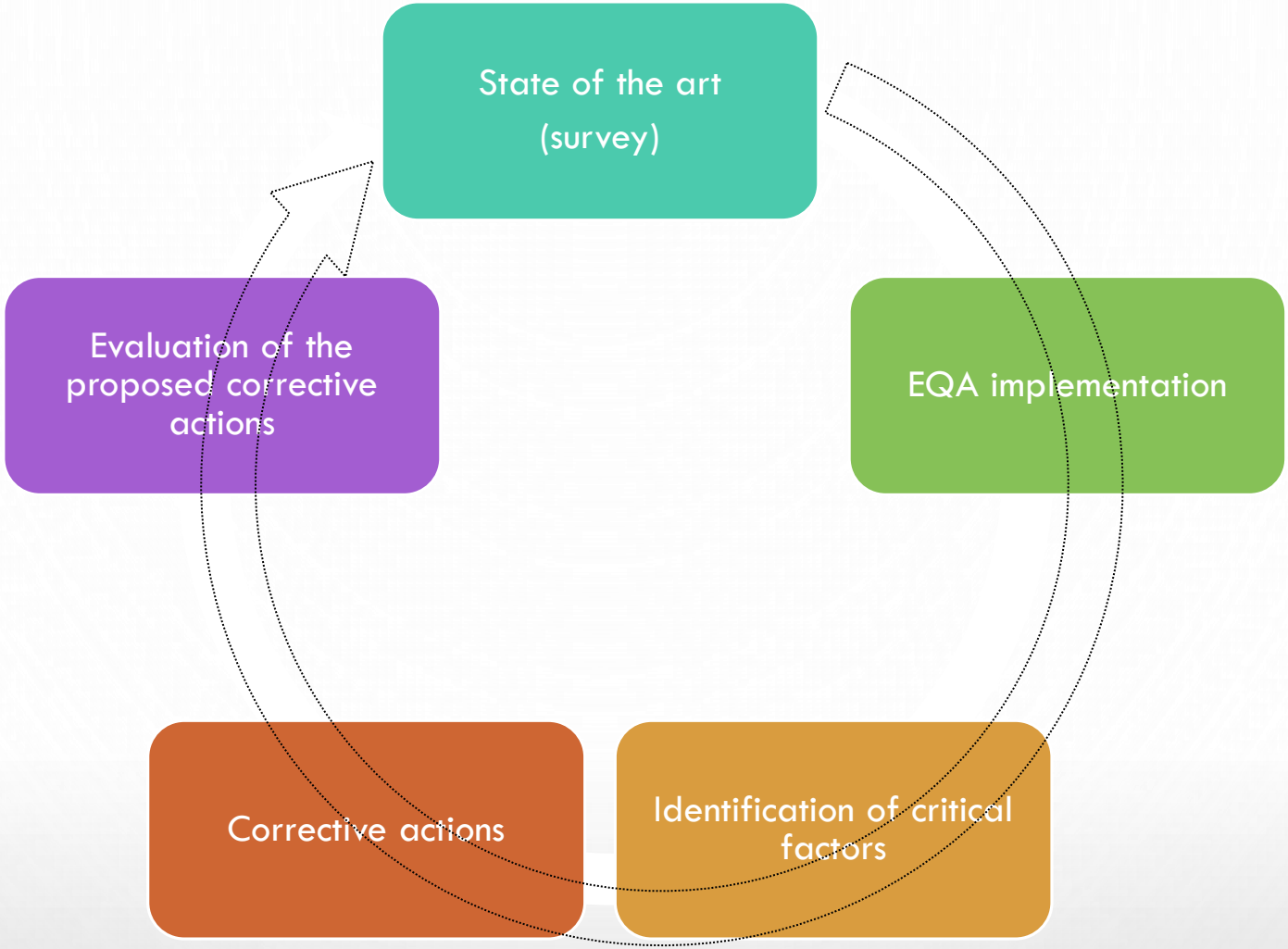
(e) monitoring of the success of harmonization/standardization efforts for improving results comparability

✓can be organized in various biomedical research field

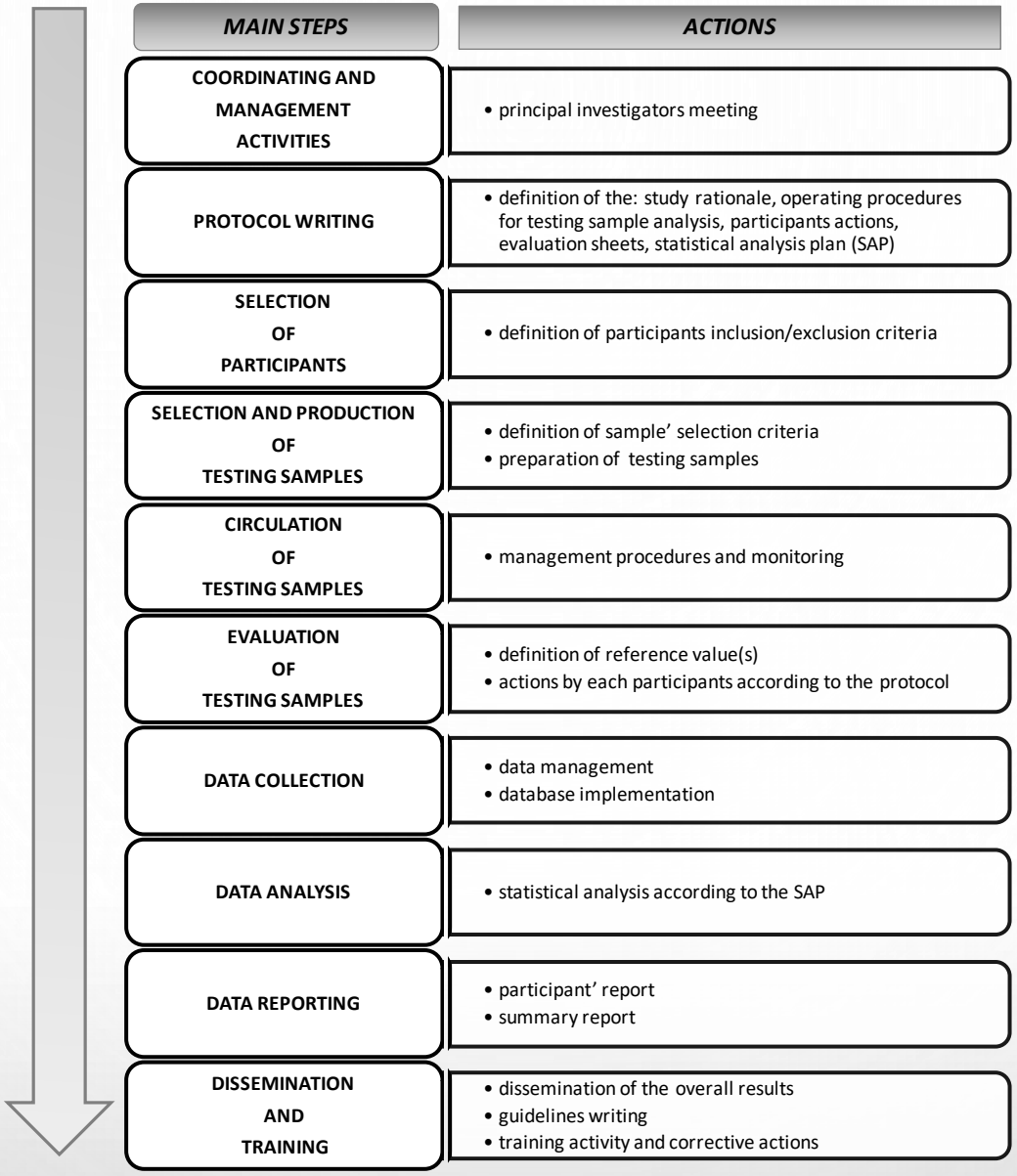
✓can be focused on the **pre-analytical, analytical or post-analytical** phase of biomarker determination

Verderio P et al. New Biotechnology 2019 (submitted)
Sciacovelli L, et al. Clin Chem Lab Med 2018;56:1644-54

Dynamicity of the EQA schemes

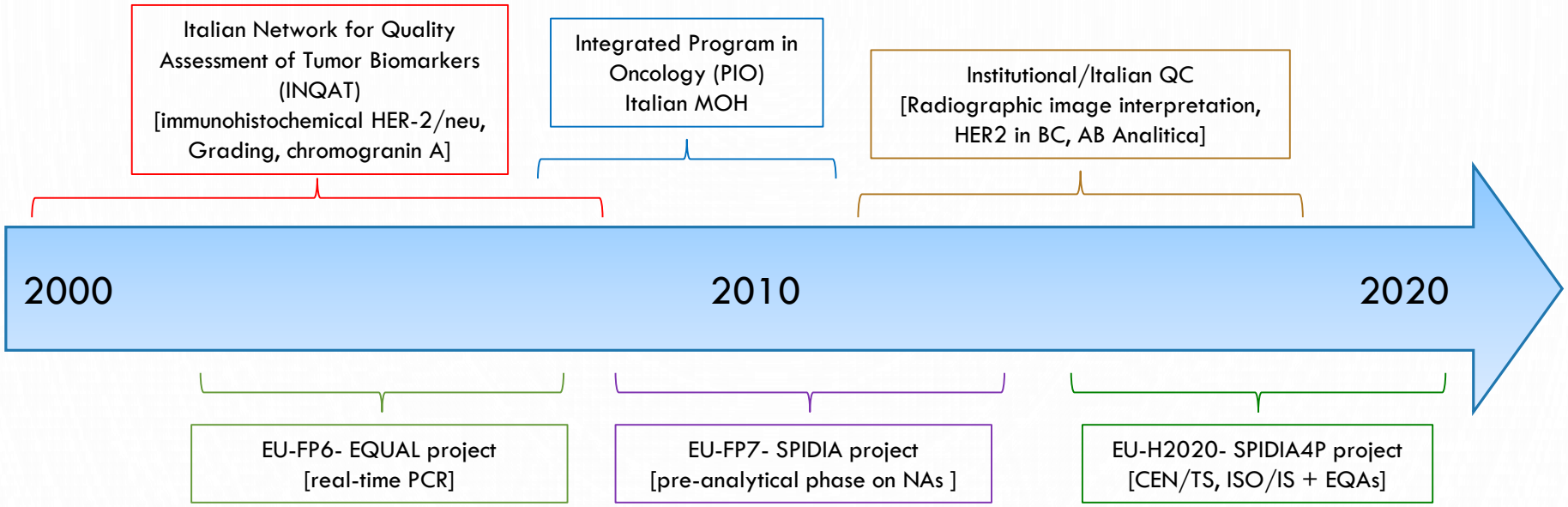


EQA: scheme design



Verderio P et al. New Biotechnology 2019 (submitted)

Our experience in EQAs

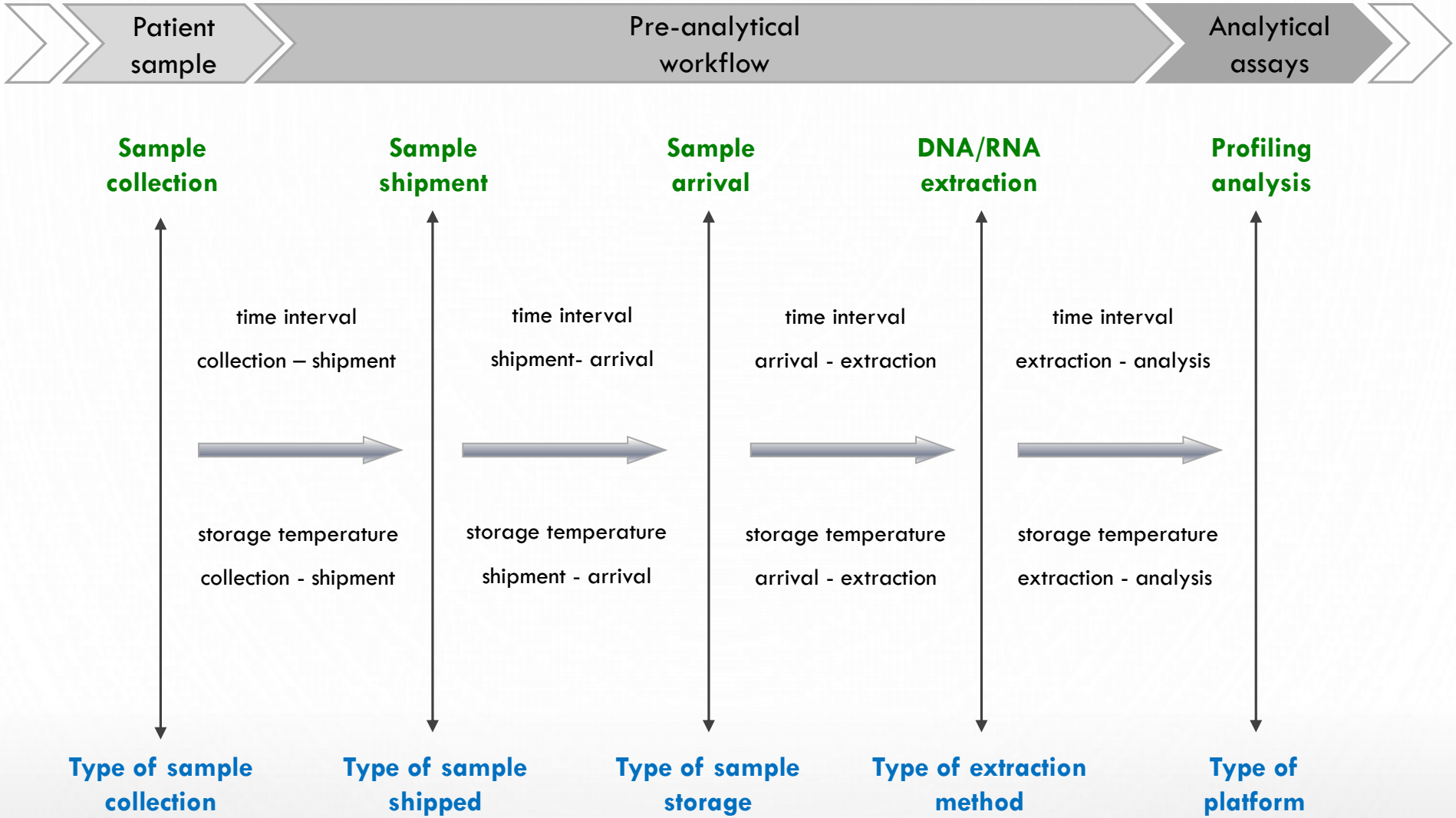


"Preanalytical errors still account for nearly 60%-70% of all problems occurring in laboratory diagnostics, most of them attributable to mishandling procedures during collection, handling, preparing or storing the specimens".

[Lippi G. et al. Clin Chem Lab Med. 2011; 49:1113-26]

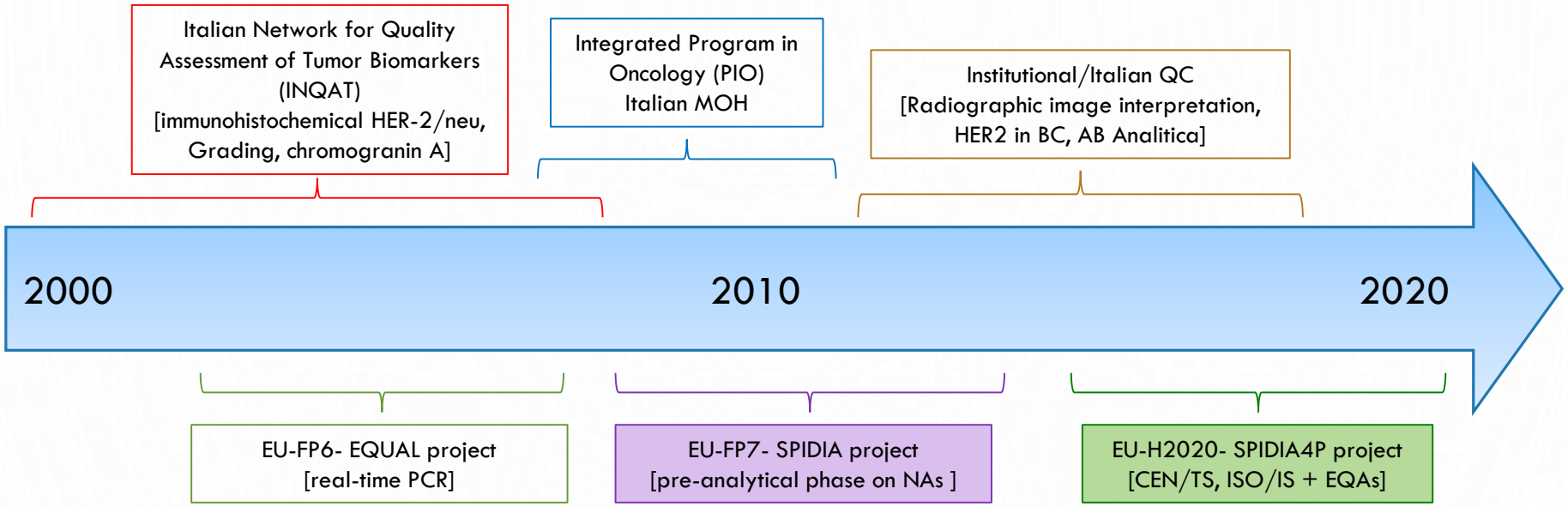
NAs: Nucleic Acids;
CEN/TS: Comité Européen de Normalisation/ Technical Specification
ISO/IS:International Organization for Standardization/Internation Standrds

Pre-analytical workflow



Adapted from Verderio P. J Clin Oncol. 2012;30:1912-5

Our experience in EQAs



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The EU SPIDIA and SPIDIA4P projects

www.spidia.eu

- **SPIDIA** (*Standardisation and improvement of generic pre-analytical tools and procedures for in-vitro diagnostics*) was a 4.5-year project funded by the EU-FP7 programme (GA n. 222916, 2008-2013)

- **SPIDIA4P** (*Standardisation and improvement of generic pre-analytical tools and procedures for in-vitro diagnostics for personalize medicine*) is a 4-year project funded by the H2020 programme (GA n. 73312, 2017-2020)
  - 19 partners coordinating by QIAGEN
  - built upon SPIDIA's results

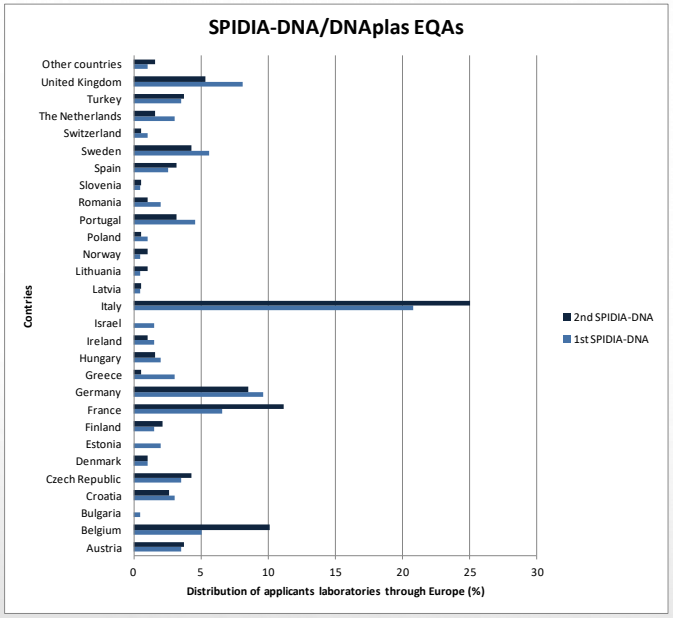
SPIDIA4P Kick-off Meeting, 30th Jan – 1st Feb 2017, Hilden

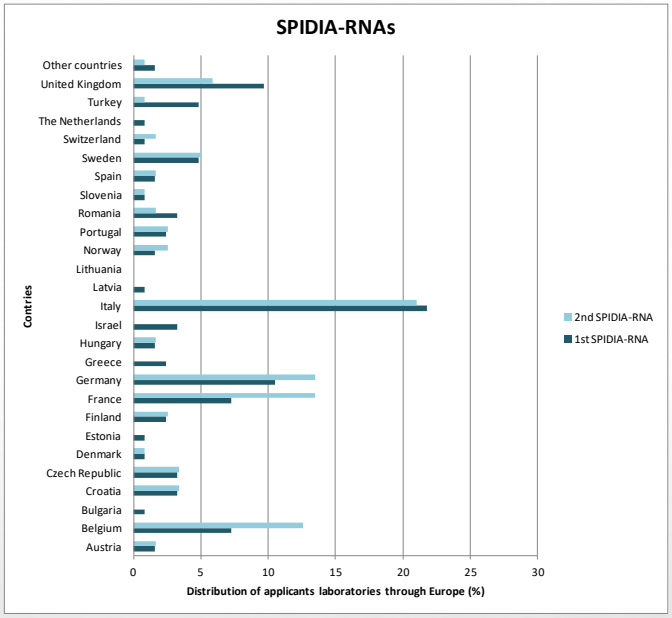
SPIDIA blood EQAs: participant laboratories

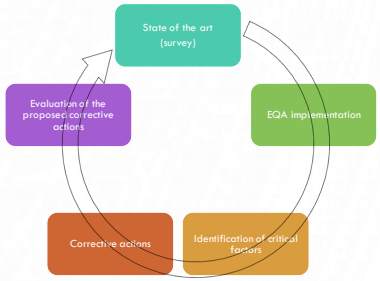
2 runs of EQAs for blood-DNA/DNA-plas and blood-RNA

	SPIDIA – DNA/DNAplas		SPIDIA - RNA	
	applicants	effective	applicants	effective
1st EQA	197[(a)]	183	124	93
2nd EQA	188[(b)]	174	122	109

[(a)] 130 labs took part to the SPIDIA-DNA, only; 67 labs to both SPIDIA-DNA and SPIDIA-DNAplas EQAs
[(b)] 127 labs took part to the SPIDIA-DNA, only; 61 labs to both SPIDIA-DNA and SPIDIA-DNAplas EQAs

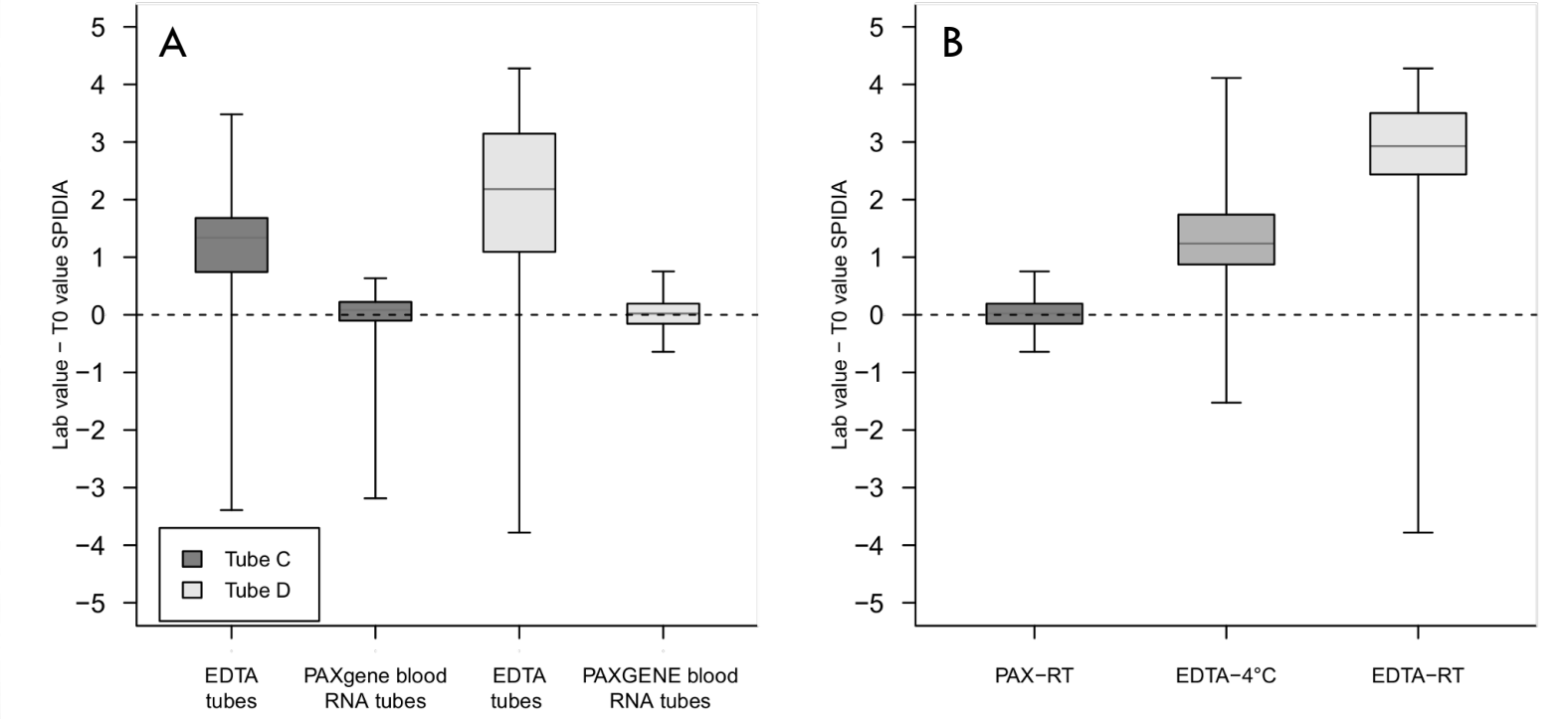






SPIDIA blood EQAs: some results

Participants were asked to extract RNA from Tube C (RNA C) immediately upon arrival of the tubes and from Tube D (RNA D) 24 h after Tube C. PAXgene Tube D was stored at RT while EDTA Tube D was stored either at RT or at 4°C according to a randomized scheme.



Overall distribution of IL8 according to blood collection tube (panel A) and to storage temperature/collection tube (panel B). The box horizontal sides identify the 25th and 75th centile, the horizontal line inside the box the median, the two whiskers correspond to minimum and maximum, and the dashed line indicates the T0 value zero.

Malentacchi F, et al. PLoS One. 2014;9(11):e112293

SPIDIA: the achieved goals

Development of **9 CEN Technical Specifications (CEN/TS) for pre-analytical workflows** in Europe, within the CEN/Technical Committee 140 for "In vitro medical devices":

- ✓CEN/TS 16826-1, snap frozen tissue – Part 1: Isolated RNA
- ✓CEN/TS 16826-2, snap frozen tissue – Part 2: Isolated proteins
- ✓CEN/TS 16827-1, FFPE tissue – Part 1: Isolated RNA
- ✓CEN/TS 16827-2, FFPE tissue – Part 2: Isolated proteins
- ✓CEN/TS 16827-3, FFPE tissue – Part 3: Isolated DNA
- ✓CEN/TS 16835-1, venous whole blood – Part 1: Isolated cellular RNA
- ✓CEN/TS 16835-2, venous whole blood – Part 2: Isolated genomic DNA
- ✓CEN/TS 16835-3, venous whole blood – Part 3: Isolated circulated cell free DNA from plasma
- ✓CEN/TS 16945, metabolomics in urine, serum and plasma

http://www.spidia.eu/publications/technical-specifications/

III. Concluding remarks

It is important in the –omics era to have:

- reliable biomarkers easily translatable in the clinic
- guidelines for the whole biomarker determination process from sample collection to biomarker detection and analysis

SPIDIA4P aim to implement comprehensive portfolio of 21 pre-analytical CEN/TS to be developed into global ISO/IS documents

THANK YOU!