Universidad de Navarra | CIMA LAB DIAGNOSTICS

Exploring the Impact of Comprehensive Genomic Profiling in Solid Tumor Testing

Gorka Alkorta-Aranburu
galkorta@unav.es

CIMA LAB Diagnostics - Clínica Universidad de Navarra

Plataforma de Genómica - Instituto de Investigación Sanitaria de Navarra (IdiSNA)

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Disclaimer

- Thermo Fisher Scientific and its affiliates are not endorsing, recommending, or promoting any use or application of Thermo Fisher Scientific products presented by third parties during this seminar.
- Information and materials presented or provided by third parties are provided as-is and without warranty of any kind, including regarding intellectual property rights and reported results.
- Speaker was provided an honorarium by Thermo Fisher Scientific for this presentation.
- Parties presenting images, text and material represent they have the rights to do so.

Clínica Universidad de Navarra (CUN, Spain)

- A small academic and research private hospital:
  - **Personalized Medicine:**
    - Patient have a reference doctor to whom the other professionals who collaborate in the comprehensive diagnosis and treatment will report.
  - **Up-to-date-technology and research:**
    - A non-profit organization that reinvest in promoting implementation of newest technologies as well as research and clinical trials
    - This allows us to offer new opportunities and new responses to our patients.
  - **Comfort and results:**
    - Streamline the process of consultations, treatments and tests for its patients.
    - Diagnostic speed, efficiency in the appointment and accessibility to results and treatment.
      - A single hospital in two different locations, but molecular diagnostics centralized in Pamplona.

https://www.cun.es/en/

The Role of CIMA LAB Diagnostics

- CIMA LAB Diagnostics is the genetic and phenotypic diagnostic laboratory of the Clínica Universidad de Navarra (CUN).

https://www.unav.edu/web/cimalab

- Collaborate side by side with clinicians in order to improve the quality of medical care

The Role of CIMA LAB Diagnostics

- … by making the newest biomarkers available to medical professionals to support diagnosis.
  - Quality Control & Assurance (QC & QA)
  - Turnaround time (TAT)
  - Newest technology.

- How? With the implementation of Comprehensive Genome Profiling (CGP), which has positively impacted our communication with clinicians directly benefitting patient care

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Agenda

- **Exploring the Impact of Comprehensive Genomic Profiling (CGP) in Solid Tumor Testing**
  - Section 1: **The impact of CGP** in everyday cases
    - What is CGP?
    - Three examples:
      - Patients with inherited pathogenic sequence changes
      - Patients with a chance to enter a clinical trial
      - Improved chances by providing MSI, TMB, mutation signatures, HRD, …
    - Final thoughts
  - Section 2: **The impact of CGP** in how we work in CIMA LAB Diagnostics in clinical research
    - Two examples:
      - Sample preparation to be molecularly tested by the pathologist
      - Biomarker identification by the molecular laboratory.

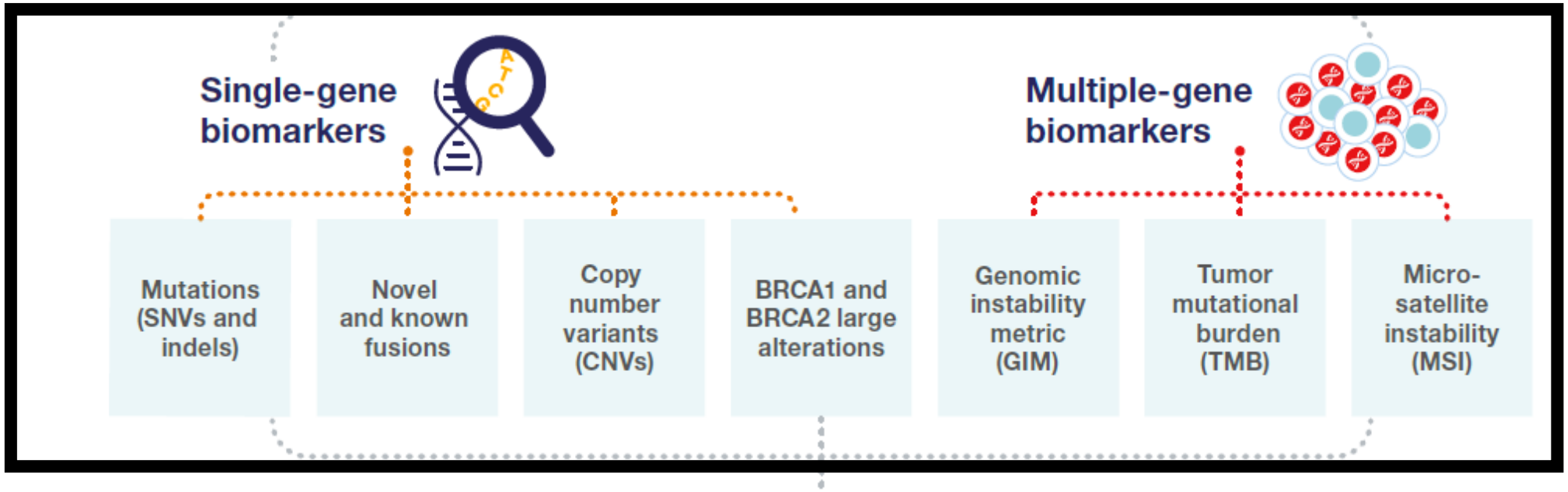
Universidad de Navarra | CIMA LAB DIAGNOSTICS

Agenda

- **Exploring the Impact of Comprehensive Genomic Profiling (CGP) in Solid Tumor Testing**
  - Section 1: **The impact of CGP** in everyday cases
    - What is CGP?
    - Three examples:
      - Patients with inherited pathogenic sequence changes
      - Patients with a chance to enter a clinical trial
      - Improved chances by providing MSI, TMB, mutation signatures, HRD, …
    - Final thoughts

Section 1: Comprehensive Genomic Profiling (CGP)

- A single test based on NGS technology that provides both single- as well as multiple-gene biomarkers; **reducing the need for more sample, time and cost.**



Section 1: CGP impacting everyday cases

Example 1. Patients with-inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change

Hindawi Publishing Corporation
BioMed Research International
Volume 2015, Article ID 341723, 11 pages
http://dx.doi.org/10.1155/2015/341723

Review Article

Hereditary Ovarian Cancer: Not Only *BRCA* 1 and 2 Genes

Angela Toss,[1] Chiara Tomasello,[1] Elisabetta Razzaboni,[1] Giannina Contu,[2] Giovanni Grandi,[2] Angelo Cagnacci,[2] Russell J. Schilder,[3] and Laura Cortesi[1]

Section 1: CGP impacting everyday cases

Example 1. Patients with inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change
- Other cancer types too
  - Pancreatic cancer patients too

https://www.cancer.gov/about-cancer/treatment/drugs/pancreatic

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 1. Patients with inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change
- Other cancer types too
  - Pancreatic cancer patients too
  - Olaparib, for example.

Drugs Approved for Pancreatic Cancer

Abraxane (Paclitaxel Albumin-stabilized Nanoparticle Formulation)
Afinitor (Everolimus)
Capecitabine
Erlotinib Hydrochloride
Everolimus
5-FU (Fluorouracil Injection)
Fluorouracil Injection
Gemcitabine Hydrochloride
Gemzar (Gemcitabine Hydrochloride)
Infugem (Gemcitabine Hydrochloride)
Irinotecan Hydrochloride Liposome
Lynparza (Olaparib)
Mitomycin
Olaparib

https://www.cancer.gov/about-cancer/treatment/drugs/pancreatic

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 1. Pati

- Patients wit
  - up to 10
- Other cance
  - Pancreat
  - Olaparib

Olaparib is approved to treat:

- **Breast cancer** that is HER2 negative and has certain germline mutations in the *BRCA1* or *BRCA2* gene. Olaparib is used after surgery in adults with:
  - High-risk early-stage breast cancer that has been treated with chemotherapy before or after surgery.
  - Metastatic cancer that has been treated with chemotherapy before or after the cancer spread.
- **Ovarian epithelial, fallopian tube,** or **primary peritoneal cancer.** Olaparib is used as maintenance therapy in adults who are having a complete or partial response to platinum chemotherapy. It is used:
  - As the first maintenance therapy in patients with advanced cancer that has certain germline or somatic mutations in the *BRCA1* or *BRCA2* gene.
  - With bevacizumab as the first maintenance therapy in patients with advanced cancer that has genomic instability and/or certain germline or somatic mutations in the *BRCA1* or *BRCA2* gene.
  - In patients with recurrent cancer.
- **Pancreatic cancer.** Olaparib is used as maintenance therapy in adults with metastatic cancer that has not progressed after first-line therapy with platinum chemotherapy and has certain germline mutations in the *BRCA1* or *BRCA2* gene.
- **Prostate cancer** that has spread to other parts of the body and is castrate resistant (has not responded to treatments that lower testosterone levels). Olaparib is used:
  - Alone in adults with germline or somatic mutations in certain genes involved in the homologous recombination repair pathway whose cancer has gotten worse after treatment with enzalutamide or abiraterone.
  - With abiraterone and prednisone or prednisolone in adults with certain mutations in the *BRCA1* or *BRCA2* gene.

Olaparib is also being studied in the treatment of other types of cancer.

BACK TO TOP

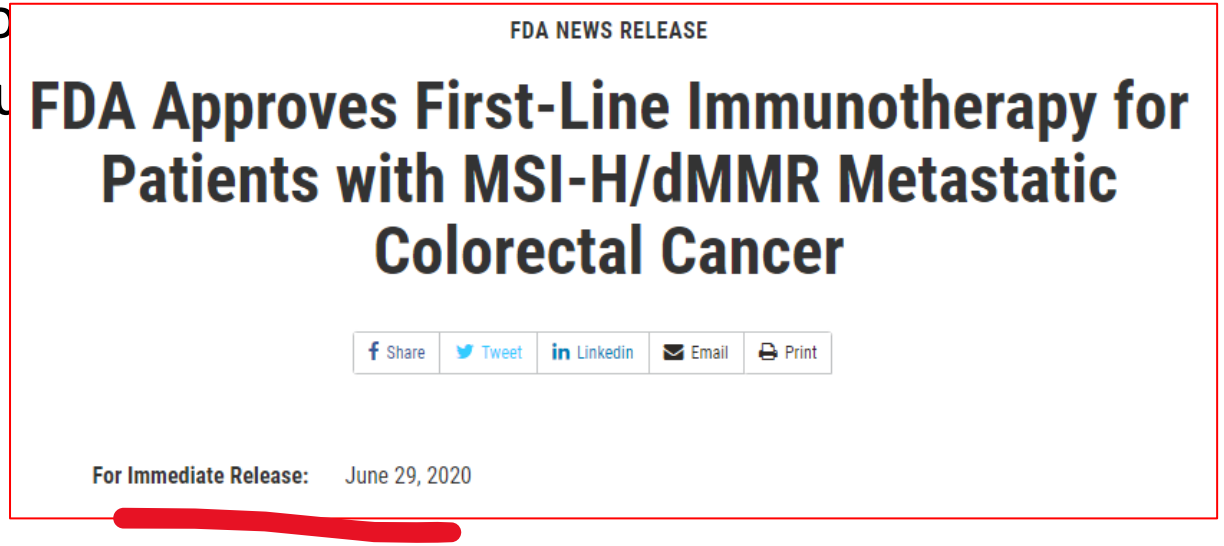
https://www.cancer.gov/about-cancer/treatment/drugs/olaparib

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 1. Patients with inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change
- Other cancer types too
  - Pancreatic cancer patients to
    - Olaparib, for example (bu
  - Colon cancer
    - MSI, for example,



FDA NEWS RELEASE

FDA Approves First-Line Immunotherapy for Patients with MSI-H/dMMR Metastatic Colorectal Cancer

Share | Tweet | Linkedin | Email | Print

For Immediate Release: June 29, 2020

https://www.fda.gov/news-events/press-announcements/fda-approves-first-line-immunotherapy-patients-msi-hdmmr-metastatic-colorectal-cancer

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 1. Patients with inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change
- Other cancer types too
  - Pancreatic cancer patients too
    - Olaparib, for example (but also for prostate cancer etc).
  - Colon cancer
    - MSI, for example,

But, we are talking about identification of patients that have inherited a pathogenic sequence change, so MSI?

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 1. Patients with inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change
- Other cancer types too
  - Pancreatic cancer patients too
    - Olaparib, for example (but also for prostate cancer etc).
  - Colon cancer
    - MSI, for example.

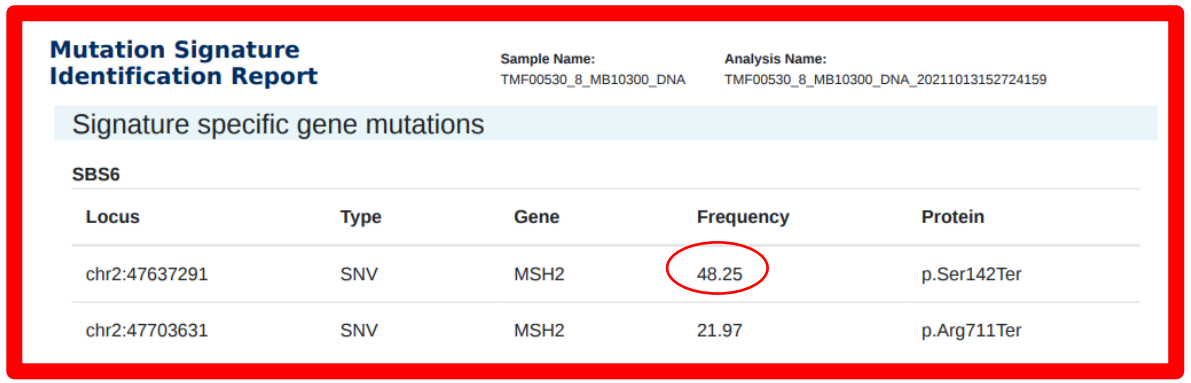
Analysis	Sample	MSI Status	MSI Score	MSI Coverage	MSI Algorithm version	MSI QC		
TMF00530_8_MB10300_c2922_2021-10-13-17-25-32-352	TMF00530_8_MB10300_DNA	MSI-High	56.59	139923	MSI_IR 2.0.2			

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 1. Patients with inherited pathogenic sequence changes

- Patients with ovarian cancer
  - up to 10% with confirmed inherited pathogenic sequence change
- Other cancer types too
  - Pancreatic cancer patients too
    - Olaparib, for example (but also for prostate cancer etc).
  - Colon cancer
    - MSI, for example.
    - However, CGP also sequenced *MSH2*.



Mutation Signature Identification Report

Sample Name: TMF00530_8_MB10300_DNA

Analysis Name: TMF00530_8_MB10300_DNA_20211013152724159

Signature specific gene mutations

SBS6

Locus	Type	Gene	Frequency	Protein
chr2:47637291	SNV	MSH2	48.25	p.Ser142Ter
chr2:47703631	SNV	MSH2	21.97	p.Arg711Ter

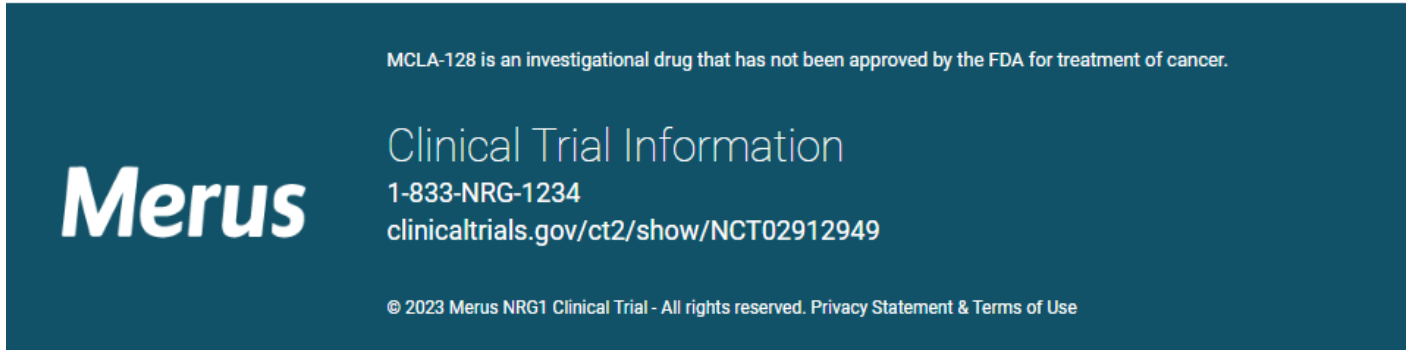
Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 2. Patients with a chance to enter a clinical trial

- For example, *NRG1* gene fusion
- We did have a case, just because the solid biopsy had been characterized with **CGP** (161 gene panel)
- We entered the clinical trial as a molecular diagnostic lab and many patients benefitted from **CGP** testing (161 and >500 genes panels)

Do you or someone you know have cancer with an NRG1 gene fusion? Here you'll find information about Merus' clinical trial for MCLA-128, an experimental medicine, for cancer patients with solid tumors with an NRG1 Fusion. You can also learn about the science behind MCLA-128.



Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: CGP impacting everyday cases

Example 3. Improved chances by providing MSI, TMB, mutation signatures, HRD, …

- MSI, but inherited *MSH2* pathogenic sequence change - colon cancer.
- TMB high case, but MSS and *POLE* pathogenic sequence change - glioblastoma.

Other studies; for example, A.B. Schrock et al., 2017

"Mutations of the DNA polymerase epsilon (POLE) can lead to a hypermutated tumor phenotype, in the absence of microsatellite instability (MSI). Exceptional responses to ICPIs in POLE-mutated endometrial adenocarcinoma (EA), colorectal (CRC), and glioblastoma (GBM) are described."

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Section 1: Final thoughts

Section 1. CGP is impacting patient care

- Providing more comprehensive information from each solid (or liquid) biopsy tested
- However,
  - ACCURACY
    - Proficiency testing programs as well as accreditations need to be mandatory
    - Case by case variant validation using techniques such as dPCR will be key (false positives).
  - SPEED
    - Shorter turnaround time is definitively desired.
  - COST
    - Less expensive is desired too.
  - MORE COMPREHENSIVE
    - For example, genomic markers (TMB, MSI, HRD, …)

Universidad de Navarra | CIMA LAB DIAGNOSTICS

Agenda

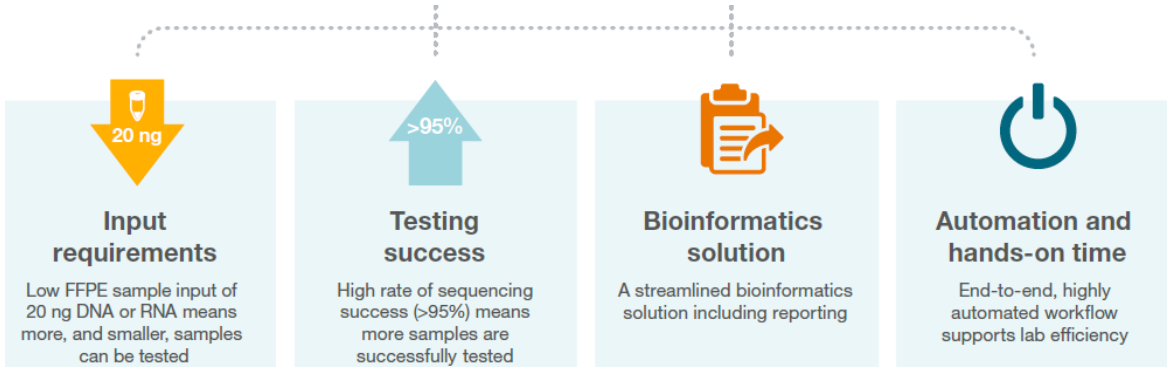
- **Exploring the Impact of Comprehensive Genomic Profiling (CGP) in Solid Tumor Testing**

- Section 2: **The impact of CGP** in how we work in CIMA LAB Diagnostics in clinical research
  - Two examples:
    - Sample preparation to be molecularly tested by the pathologist
    - Biomarker identification by the molecular laboratory

Section 2: CGP in CIMA LAB

Oncomine Comprehensive Assay Plus (OCA Plus)

- From one sample, in one assay run, you can achieve CGP based on DNA and RNA analysis of >500 genes without having to compromise on:



- **165** genes with recurrent hotspot mutations
- **333** genes with focal CNV gains or loss
- **227** genes with full-coding DNA sequence (CDS)
- **46** genes in Homologous Recombination Repair pathway
- **49** Fusion driver genes
- **MET** exon skipping detection at DNA and RNA level
- **Others:**
  - **Cellularity** (Tumor fraction) calculation
  - Tumor Mutational Burden (**TMB**)
  - Microsatellite Instability (**MSI**)
  - Loss of heterozygosity (**gene LOH)**
  - Large genomic alterations in ***BRCA1*** and ***BRCA2***
  - Genomic Instability Metric **(GIM)**

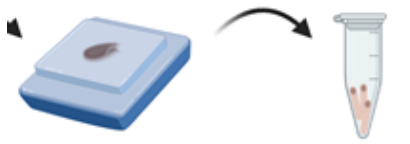
Section 2: CGP in CIMA LAB

Example 1. Impacting sample preparation for pathologist? NO.

- 52 gene panel - Oncomine Focus Assay (OFA)
- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

5-micron sections

5-8 sections



DNA

Please, careful with:

- The first time you implement the sample quantity requirements in collaboration with the pathologist, but after that no need to change.

5-8 sections

RNA

Two eppendorf tubes with between 5 and 8 (5-micron) FFPE sections (with higher than 50% tumor content) each

Modified from https://www.coriell.org/1/NIGMS/Additional-Resources/FFPE

Section 2: CGP in CIMA LAB Diagnostics

Example 1. Impacting sample preparation for lab technicians? NO.

- 52 gene panel - Oncomine Focus Assay (OFA)
- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

- OCA Plus analyzes both DNA as well as RNA
  - SNV, indels and CNVS – DNA sample
  - Fusions – RNA sample

Maxwell RSC (Promega)

Please, careful with:

- FFPE sample processing (and storage)
- Nucleic acid isolation (and storage)

Qubit (ThermoFisher)

TapeStation (Agilent)

Modified from https://www.coriell.org/1/NIGMS/Additional-Resources/FFPE

Section 2: CGP in CIMA LAB Diagnostics

Example 2. Impacting biomarker identification for lab technicians? NO MUCH!

- 52 gene panel - Oncomine Focus Assay (OFA)
- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

Please, careful with:

- Adapting the S5 to be able to use ION 550 sequencing chips
- Only 4 cases per chip.

Otherwise, no other change!

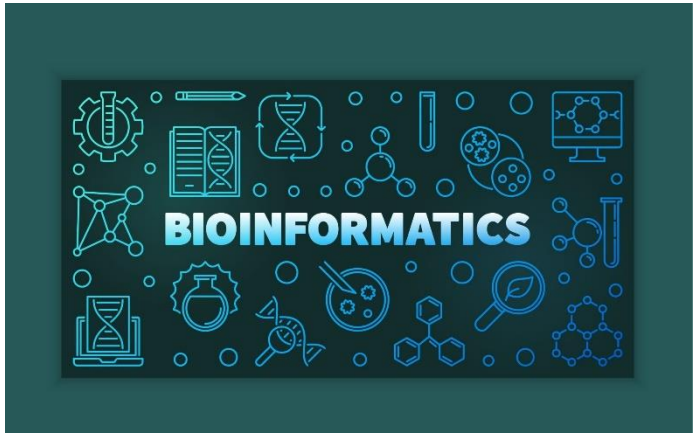
Section 2: CGP in CIMA LAB Diagnostics

Example 2. Impacting biomarker identification for bioinformaticians? YEP!

- 52 gene panel - Oncomine Focus Assay (OFA)
- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

Please, careful with:

- data storage – significantly more raw data.
- the verification process - more complex



https://leverageedu.com/blog/application-of-bioinformatics/

Section 2: CGP in CIMA LAB Diagnostics

Example 2. Impacting biomarker identification for geneticists? YEP!

- 52 gene panel - Oncomine Focus Assay (OFA)
- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

Please, careful with:

- the amount of new information that has to be included in each report (including the material and method section with the assay limitations)
- the communication with other colleagues

Section 2: CGP in CIMA LAB Diagnostics

Example 2. Impacting biomarker identification for high-risk cancer research samples? YEP!

Pre-CGP implementation:

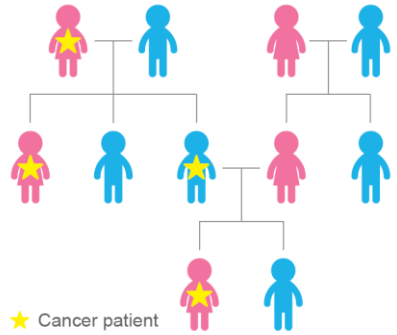
- Germline-testing from blood samples

However, CGPs do sequence the genomic region of relevant cancer predisposition genes.

- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

Post-CGP implementation:

- Careful with samples with inherited pathogenic sequence changes



https://www.sofivagenomics.com.tw/en/Product/2/4/item/19

Section 2: CGP in CIMA LAB Diagnostics

Example 2. Impacting biomarker identification for clinical research trials? YEP!

- 52 gene panel - Oncomine Focus Assay (OFA)
- 161 gene panel - Oncomine Comprehensive Assay (OCA)
- 500+ gene panel - Oncomine Comprehensive Assay Plus (OCA Plus)

Please, careful with:

- the amount of information received - ask if you have any question
- the requirements that clinical research trials have (clinical trial unit)

https://www.thetech.org/ask-a-geneticist/

Final conclusions

Section 1. CGP is impacting patient care

- Providing more comprehensive information from each solid (or liquid) biopsy tested

Section 2. CGP has impacted CIMA LAB Diagnostics in clinical research

- No much:
  - Sample preparation
  - DNA/RNA isolation
  - NGS library preparation and sequencing
- Significantly,
  - Biomarker identification

Section 2: CGP in CIMA LAB

Oncomine Comprehensive Assay Plus (OCA Plus)

- Positive control samples are being used to assess OCA Plus ability to detect:
  - SNV, indels,
  - CNVs
  - Fusions
  - TMB
  - MSI
  - ….

- But, what about HRD?

- **165** genes with recurrent hotspot mutations
- **333** genes with focal CNV gains or loss
- **227** genes with full-coding DNA sequence (CDS)
- **46** genes in Homologous Recombination Repair pathway
- **49** Fusion driver genes
- **MET** exon skipping detection at DNA and RNA level
- **Others:**
  - **Cellularity** (Tumor fraction) calculation
  - Tumor Mutational Burden (**TMB**)
  - Microsatellite Instability (**MSI**)
  - Loss of heterozygosity (**gene LOH)**
  - Large genomic alterations in ***BRCA1*** and ***BRCA2***
  - Genomic Instability Metric **(GIM)**

Section 2: CGP in CIMA LAB Diagnostics

Oncomine Comprehensive Assay Plus (OCA Plus)

- Positive control samples are being used to assess OCA Plus ability to detect:
  - SNV, indels,
  - CNVs
  - Fusions
  - TMB
  - MSI
  - ….

- But, what about HRD?

- HRD is the inability of a cell to effectively repair DNA double-strand breaks using the homologous recombination repair (HRR) pathway.
- Tumors with HRD sensitive to PARP inhibitors

- CGP assays can report Genomic Instability that quantifies genomic scarring associated with HRD, for example, OCA Plus provides: Genomic Instability Metric (GIM) values per analyzed sample.
  - Does GIM may reliably detect HRD status? Dr. Staebler´s talk

Exploring the Impact of Comprehensive Genomic Profiling in Solid Tumor Testing

Thanks

Gorka Alkorta-Aranburu, PhD

galkorta@unav.es

https://www.unav.edu/web/cimalab