Técnicas ómicas en el diagnóstico de enfermedades raras

Día 1

El proyecto iGenCo y la plataforma RD-Cat GPAP

Día 2

Uso y análisis del transcriptoma y el epigenoma

16 -17 Noviembre

Inscripciones gratuitas:

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Agenda – Técnicas Ómicas en el Diagnóstico de Enfermedades Raras

	DÍA 1 – 16 de noviembre 2022
15:00	Introducción al proyecto iGenCO **Sergi Beltran**, CNAG-CRG
15:30	Plataforma RD-Cat: módulo de datos fenotípicos - teoría (30 min) + práctica (1h) **Leslie Matalonga**, CNAG-CRG y **Gemma Bullich**, CNAG-CRG
17:00	**COFFEE BREAK**
17:30	Plataforma RD-Cat: módulo de datos genómicos - teoría (30 min) + práctica (1h) **Leslie Matalonga**, CNAG-CRG y **Gemma Bullich**, CNAG-CRG

Organizan:

ISGlobal Instituto de Salud Global Barcelona

cnag centre nacional d'anàlisi genòmica centro nacional de análisis genómico

CRG Centre for Genomic Regulation

Con el apoyo de:

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Agenda – Técnicas Ómicas en el Diagnóstico de Enfermedades Raras

	DÍA 2 – 17 de noviembre 2022
15:00	Uso del transcriptoma y epigenoma en el diagnóstico de enfermedades raras **Juan Ramón González**, ISGlobal
15:30	Análisis del transcriptoma - teoría (30 min) + práctica (30 min) **Marc Dabad**, CNAG-CRG y **Gerard Múñoz**, Hospital Clínic, IDIBAPS, CIBERER
17:00	COFFEE BREAK
17:30	Análisis del epigenoma - teoría (30 min) + práctica (30 min) **Laura Balagué**, ISGlobal, **Natàlia Carreras**, ISGlobal y **Xavier Escribà**, ISGlobal

Organizan:

Con el apoyo de:

Centro Nacional de Análisis Genómico (CNAG-CRG)

- ✓ Created in 2010
- ✓ Funded by MCIU and Generalitat de Catalunya
- ✓ Competitive grants & contractual research provide additional funds
- ✓ Since 2015 it is integrated with the CRG
- ✓ 100 people, directed by Ivo Gut

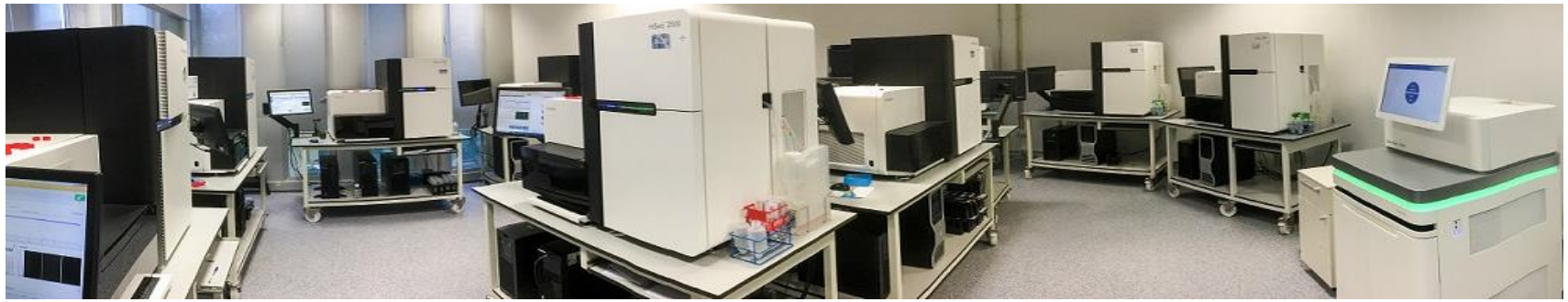
Mission

- ✓ To carry out projects in genome analysis that will lead to significant improvements in people's health and quality of life, in collaboration with the Spanish, European and International research and clinical community.

Vision

- ✓ To be a high quality sequence analysis center and to be a world reference center for genomic analysis.

The CNAG-CRG's Genomehenge 2022



Sequencing capacity	>10,000 Gbases/day = 100 human genomes/day at 30x
Sequencing instruments	7 Illumina sequencers (3 NovaSeq6000, 1 HiSeq2500, 2 HiSeq4000, 1 MiSeq) 1 GridIon, 1 Promethion (Oxford Nanopore Technologies)
Single cell/ Spatial Genomics	10x Chromium Controller, 10x Chromium Connect Vutara microscope
Computing	8,500 cores 10 PB disk + 3 PB tape

CNAG-CRG Quality

- ✓ SGS Certification ISO 9001: 2015
- ✓ ENAC ISO 17025 : 2005 Accreditation
- ✓ BBMRI-ERIC Expert Centre
- ✓ Oxford Nanopore Technologies Certified Service Provider
- ✓ Genomic Quality Assessment Programs (GenQA)
- ✓ Coordination of an interlaboratory comparison program for Whole Genome Sequencing (Proficiency testing. ISO/IEC 70243)
- ✓ Preparation of standarized guidelines for the International Organization of Standarization (ISO)

ENAC
Entidad Nacional de Acreditación

SYSTEM CERTIFICATION STANDARD SGS

BBMRI-ERIC EXPERT Centre TRUSTED PARTNER

Oxford NANOPORE Technologies

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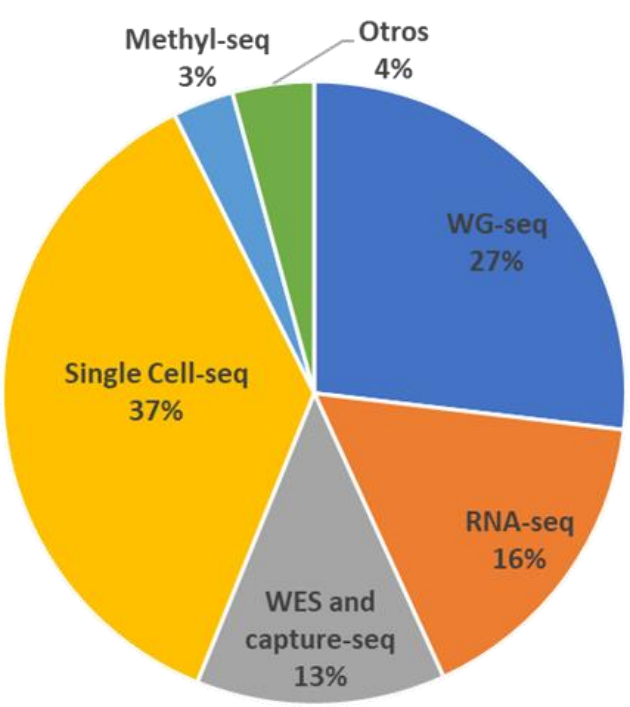
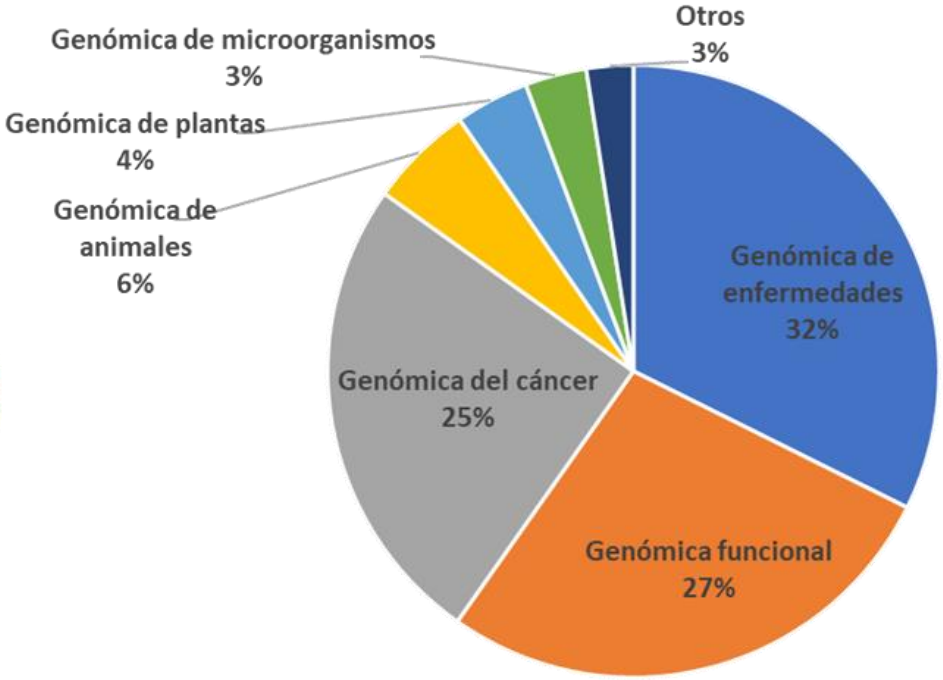
centro nacional de análisis genómico

CRG
Centre for Genomic Regulation

The CNAG-CRG Activity

2021 691projects,
221 collaborators
30,828 samples processed
396 Tb of sequence produced





CNAG-CRG Strategic Areas

Personalized Medicine

GA4GH
Global Alliance for Genomics and Health

European 1+MG initiative

ISCIII IMPaCT projects

EU projects
Screen4Care
Genomed4all

Rare Diseases

IRDiRC
International Rare Diseases Research Consortium

EU Research projects
Solve-RD
EJP-RD

RD-Connect
Genome Phenome Analysis platform

Cancer

ICGC
International Cancer Genome Consortium

EU Research projects
EUCanCan
IMMUCan

Cancer-Connect
Genome Phenome Analysis platform

Single-Cell Analysis

HCA
Human Cell Atlas

Chan Zuckerberg Initiative

EU projects
BCLL@las
Doctis
Espace

Genome in Action

IHEC
International Human Epigenome Consortium

EU projects
LifeTime
3Domics

NIH project
Center for genomics imaging

Biodiversity

BGE
Biodiversity Genomics Europe

ERGA
European Reference Genome Atlas

Catalan Biogenome Project

Introducción al proyecto iGenCO

Técnicas Ómicas en el Diagnóstico de Enfermedades Raras
Barcelona, 16-17 Noviembre 2022

Sergi Beltran
Centre Nacional d'Anàlisi Genòmica, CNAG-CRG, Barcelona

cnag CRG Centre for Genomic Regulation

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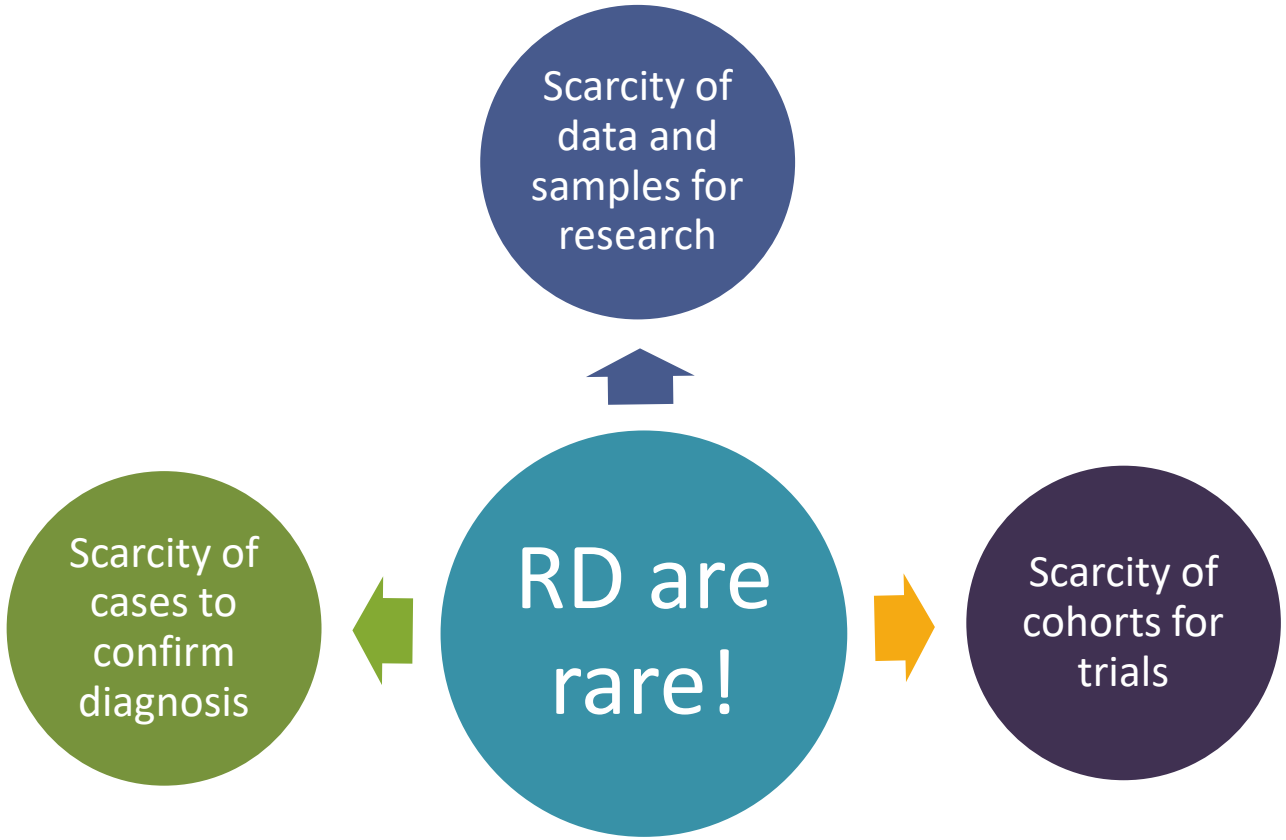
Rare diseases and genomics

Unmet needs and bottlenecks in RDs

Lack of diagnosis

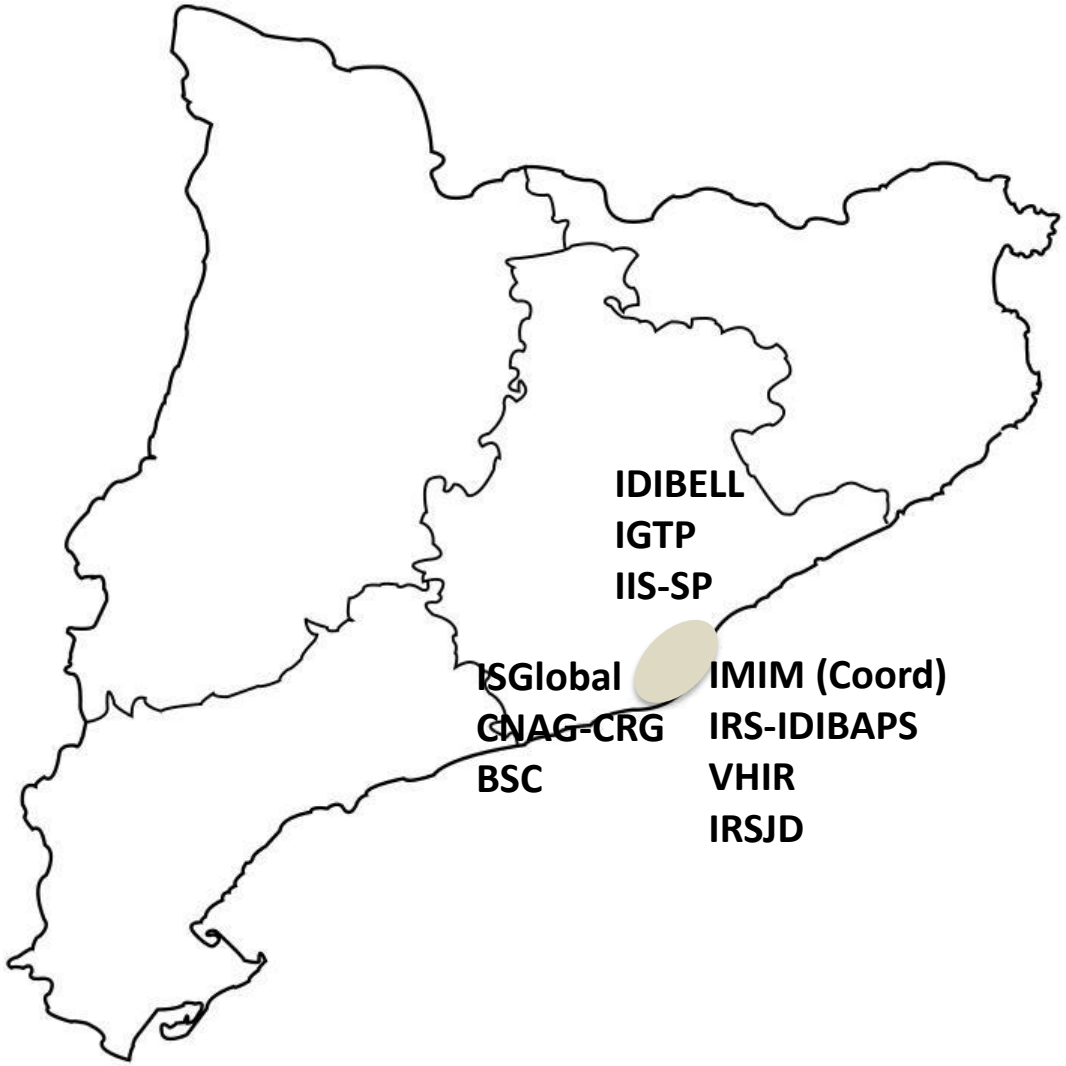
Lack of therapies



Data sharing is essential

Undiagnosed Rare Disease Program of Catalonia

2017-2019



Coordinator: Luis Pérez Jurado (IMIM)

16 Groups
- 7 Hospitals (IIS)
- 3 CERCA centers
- FEDER (Patients)

MAIN OBJECTIVE:

Enable the Catalan Health System to **provide personalised genomic medicine as a fully integrated service for patients with RDs**, initially as a pilot project for RDs with neurologic involvement.

PERIS 2016 2020

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Undiagnosed Rare Disease Program of Catalonia (pre iGenCO results)

URDCat *2017-2019*

Prioritised by clinical committee

934 neurological RD cases proposed
- Undiagnosed after routine tests
- With or without existing NGS data (panels/WES/WGS)

GPAP URDCAT

Genomics (panels/WES/WGS)
SNV/InDels coding regions CNVs
Relatedness, ROH*
*Matalonga L *et al* 2020 – J Mol Diag

Reanalysis (panels/WES/WGS – 323 cases):
61 of 323 cases diagnosed (18.9%)
Bullich G *et al* 2022 J Mol Diag.

New generated data (WES/WGS - 735 cases):
161 of 495 analysed cases diagnosed (32.5 %)

Transcriptomics (pilot n=29)
DROP protocol: Aberrant splicing, MAE, Aberrant expression
Yépez VA *et al* 2021 – Nature Protocols

Transcriptome:
3 of 29 cases diagnosed (10.3%)

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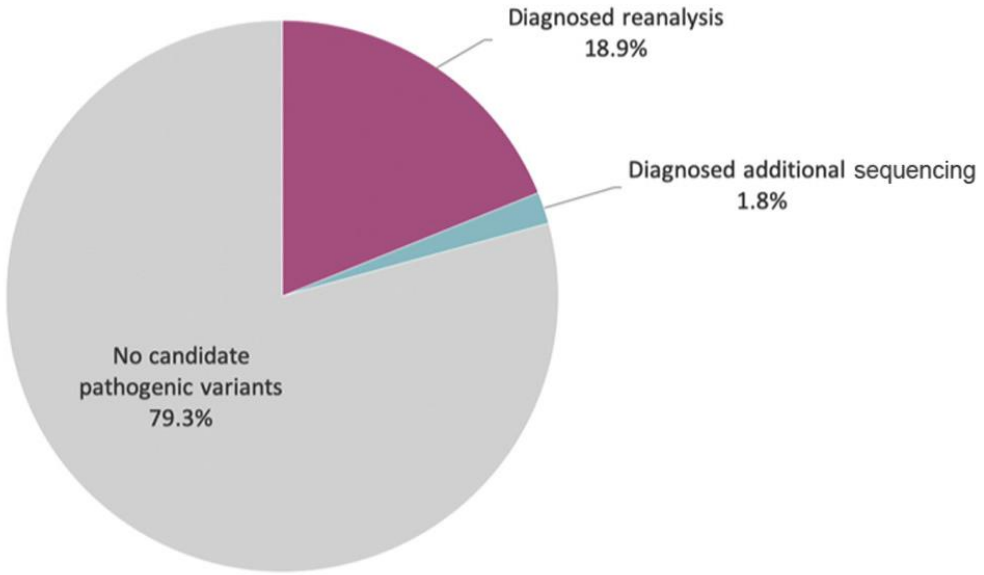
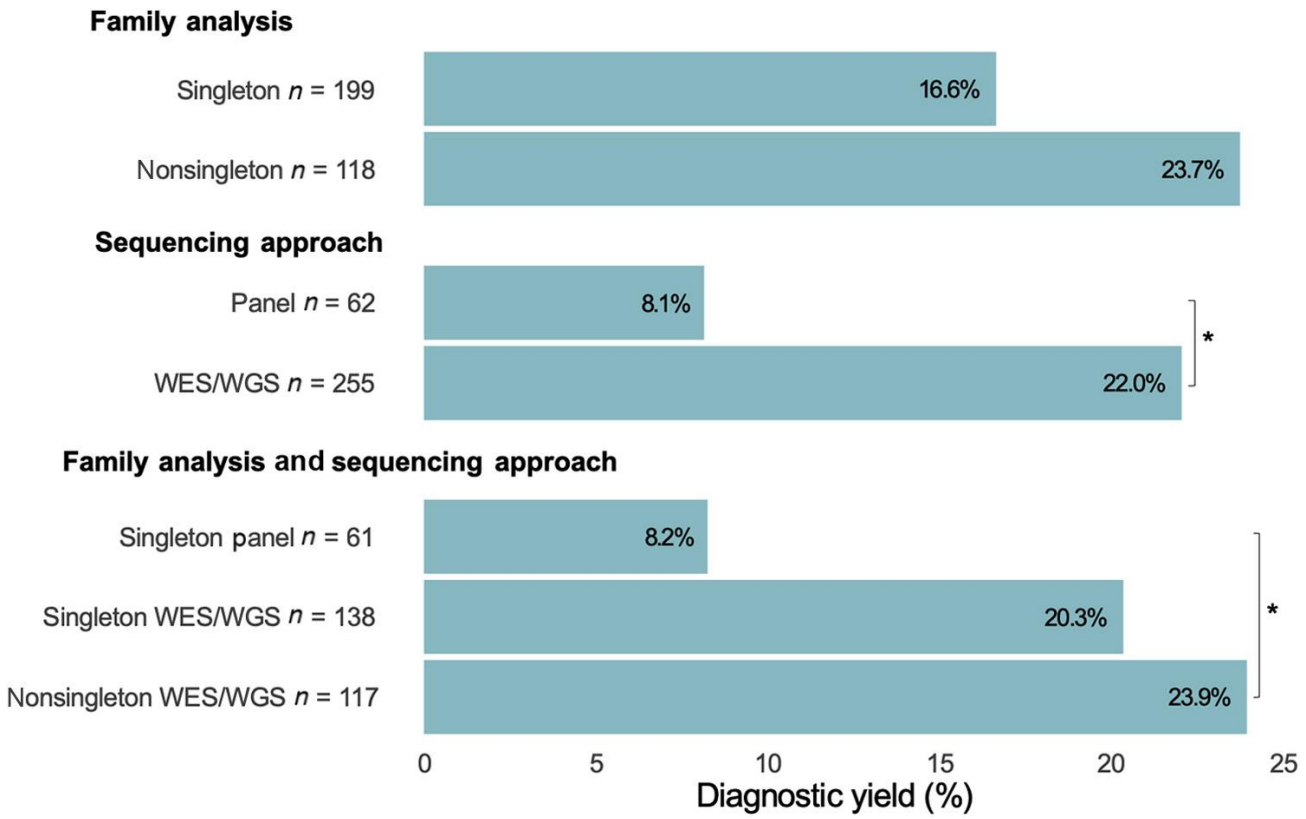
Reanalysis results

2017-2019

Molecular results overview



Diagnostic yield by type of family analyses and sequencing strategy

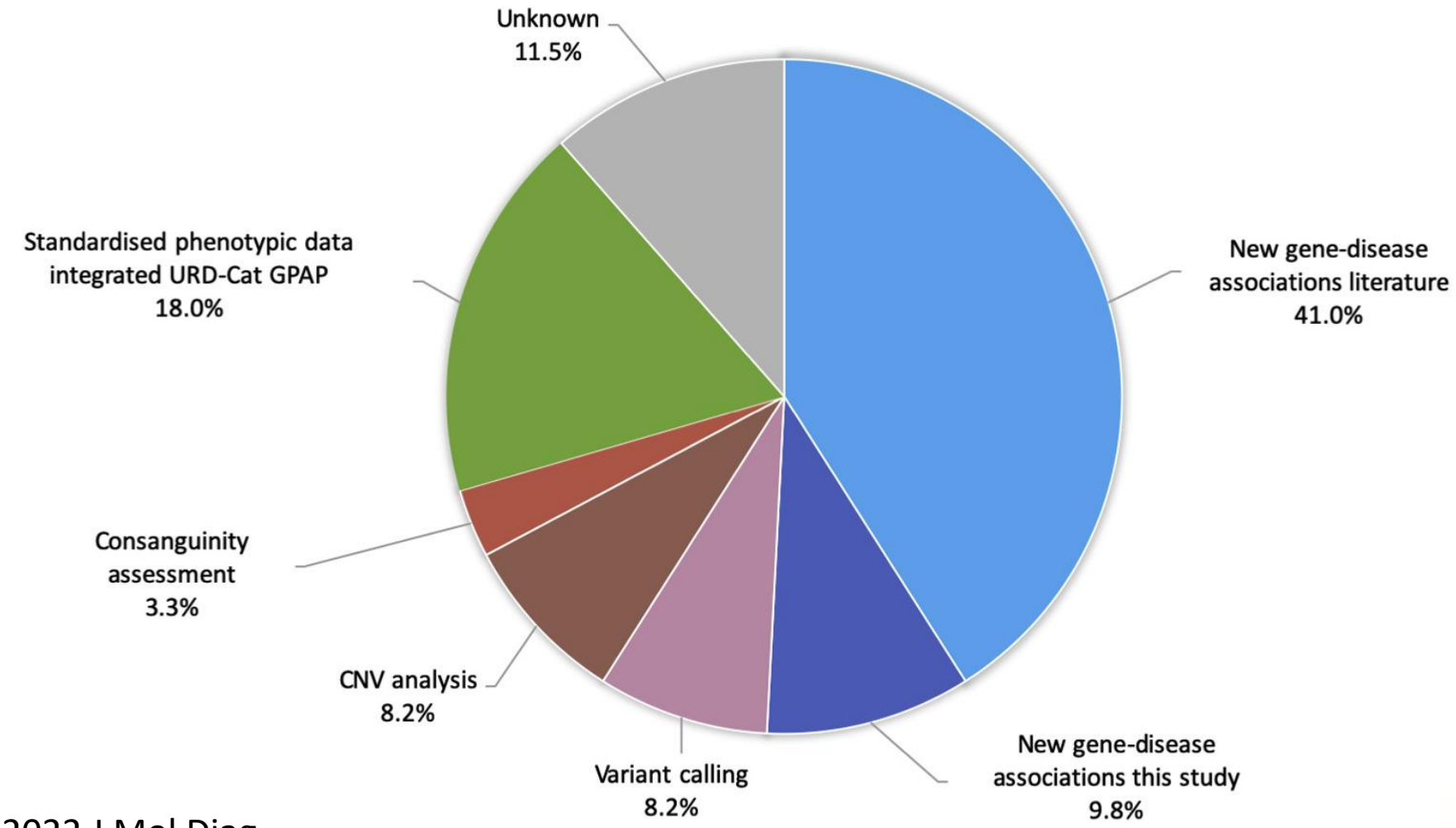


Bullich G *et al* 2022 J Mol Diag.

Reanalysis results

2017-2019

Reasons for increasing diagnostic rate with data reanalysis.



Bullich G *et al* 2022 J Mol Diag.

Undiagnosed Rare Disease Program of Catalonia (pre iGenCO results)

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GPAP URDCAT

Genomics (panels/WES/WGS)
SNV/InDels coding regions CNVs
Relatedness, ROH*
*Matalonga L *et al* 2020 – J Mol Diag

222 index cases diagnosed
658 index cases undiagnosed

Reanalysis (panels/WES/WGS – 323 cases):
61 of 323 cases diagnosed (18.9%)
Bullich G *et al* 2022 J Mol Diag.

New generated data (WES/WGS - 735 cases):
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CRG Centre for Genomic Regulation

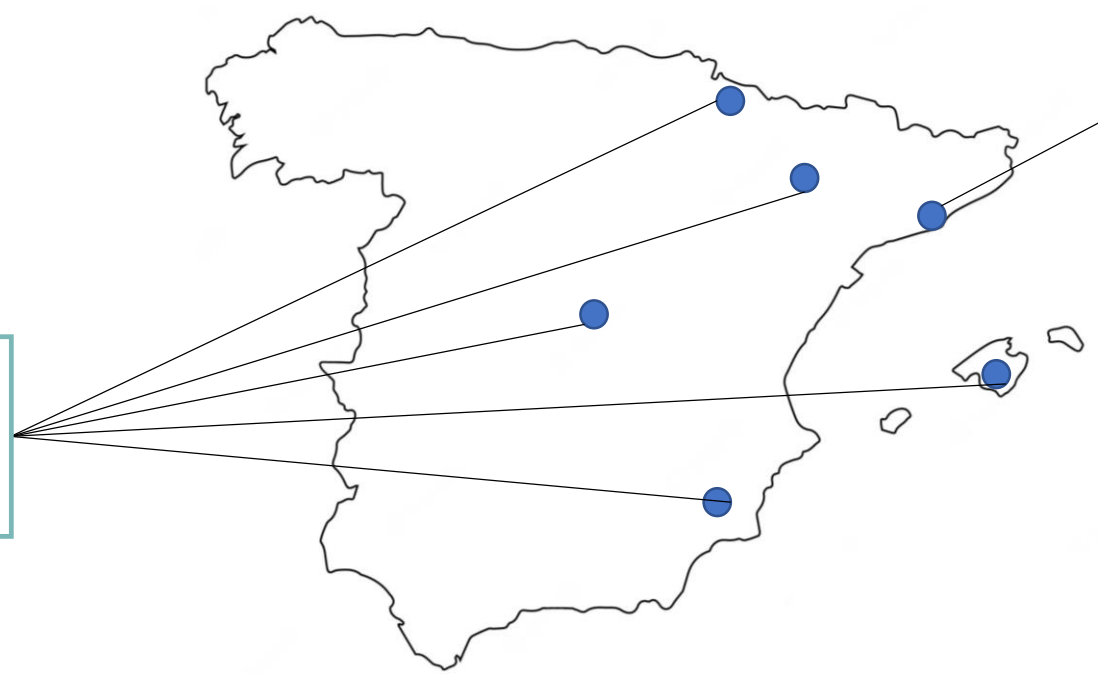
iGenCO 2021-2024

In-depth genomics and cross-omics analysis

Collaborative project: Sergi Beltran (coordinator) — Juan Ramón González (incl. Luís Pérez-Jurado)

Funded by: Fundació La Marató 3

Data submitting centres:



URDCat consortium

ciberer — Centro de Investigación Biomédica en Red — Enfermedades Raras

iGenCO
2021-2024

In-depth genomics and cross-omics analysis

cnag
centre nacional d'anàlisi genòmica
centro nacional de análisis genómico

CRG
Centre for Genomic Regulation

Global aim

To develop innovative solutions to facilitate **in-depth genomic and cross omics analysis** of molecularly undiagnosed rare disease (RD) patients to enable their **diagnosis** and **novel gene discovery**

Specific aims:

- Develop standardised pipelines for **in-depth genome-phenome analysis** of different genetic variant types in the existing genome and exome data. Inclusion of additional variant types and annotations for non-coding regions in the RD-Cat platform (Work Package 1).
- Enable **automated periodic identification** of **new candidate variants** and implement international data discovery and patient matchmaking functionalities in the RD-Cat platform (Work Package 2).
- Generate **transcriptomics and/or epigenomics** data to guide the identification of new pathogenic genes and variants in undiagnosed patients after in-depth genome-phenome analysis (Work Package 3).

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SLIDES REMOVED

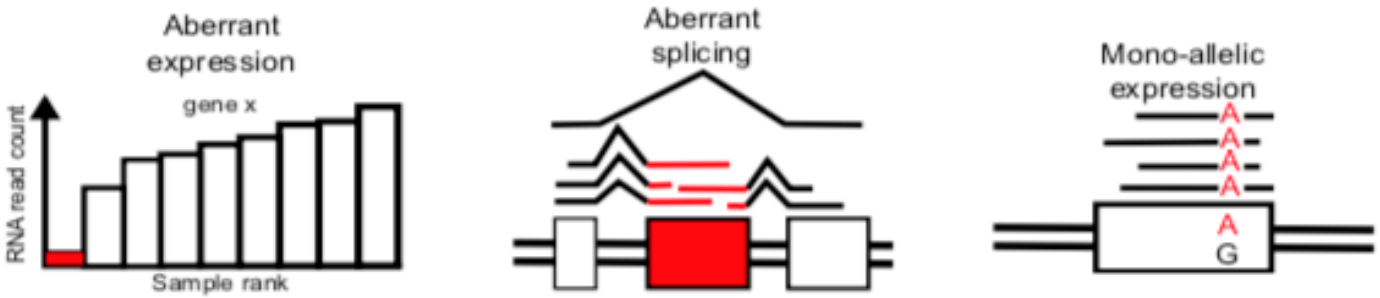
Methods: transcriptomes

Sequencing:

- paired-end reads (2x150bp) directional mRNA

Analysis:

- DROP protocol (Gagneur lab: Yépez VA *et al* 2021 – Nature Protocols): detection of RNA Outliers Pipeline
- Detects aberrant expression levels, aberrant splicing and mono-allelic expression:



Kremer *et al* 2017 – Nature Communications

- Controls – 2 options depending on tissue availability:
  - matched-tissue reference RNA-Seq from public repository
  - Rest of the patients from the analysis cohort

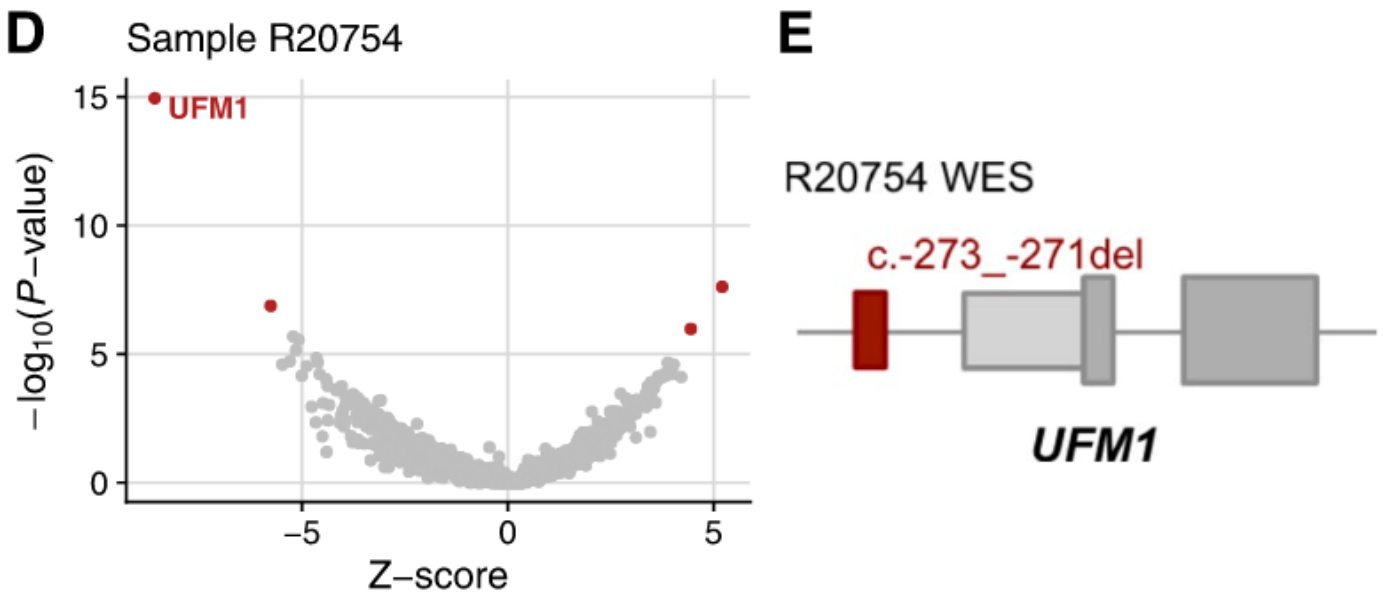
Yépez et al. Genome Medicine (2022) 14:38
https://doi.org/10.1186/s13073-022-01019-9

Genome Medicine

RESEARCH Open Access

Clinical implementation of RNA sequencing for Mendelian disease diagnostics

Vicente A. Yépez[1,2,3†], Mirjana Gusic[1,4,5†], Robert Kopajtich[1,4], Christian Mertes[2], Nicholas H. Smith[2], Charlotte L. Alston[6,7], Rui Ban[4,8], Skadi Beblo[9], Riccardo Berutti[1,4], Holger Blessing[10], Elżbieta Ciara[11], Felix Distelmaier[12], Peter Freisinger[13], Johannes Häberle[14], Susan J. Hayflick[15], Maja Hempel[16], Yulia S. Itkis[17], Yoshihito Kishita[18,19], Thomas Klopstock[20,21,22], Tatiana D. Krylova[17], Costanza Lamperti[23], Dominic Lenz[24], Christine Makowski[25], Signe Mosegaard[26], Michaela F. Müller[2], Gerard Muñoz-Pujol[27], Agnieszka Nadel[1,4], Akira Ohtake[28,29], Yasushi Okazaki[18], Elena Procopio[30], Thomas Schwarzmayr[1,4], Joél Smet[31], Christian Staufner[24], Sarah L. Stenton[1,4], Tim M. Strom[1,4], Caterina Terrile[4], Frederic Tort[27], Rudy Van Coster[31], Arnaud Vanlander[31], Matias Wagner[1,4], Manting Xu[4,8], Fang Fang[8], Daniele Ghezzi[23,32], Johannes A. Mayr[33], Dorota Piekutowska-Abramczuk[11], Antonia Ribes[27], Agnès Rötig[34], Robert W. Taylor[6,7], Saskia B. Wortmann[1,33,35], Kei Murayama[36], Thomas Meitinger[1], Julien Gagneur[1,2,3*] and Holger Prokisch[1,4,8*]



Slide by Frederic Tort and Antònia Ribes (IDIBAPS)

iGenCO **Methods: epigenomes** (PI: Juan Ramón González, ISGlobal)

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Methylation profiling:

- Infinium MethylationEPIC BeadChip kit (Illumina)

Analysis:

- Epivariants: DNA methylation defects at a single locus

  De novo epi-variations are significantly enriched in Neurodevelopmental disorders and congenital anomalies, while RNA-Seq analysis shows that epi-variations often have an impact on gene expression comparable to loss-of-function mutations (Barbosa M *et al* 2018 – Nature Communications).

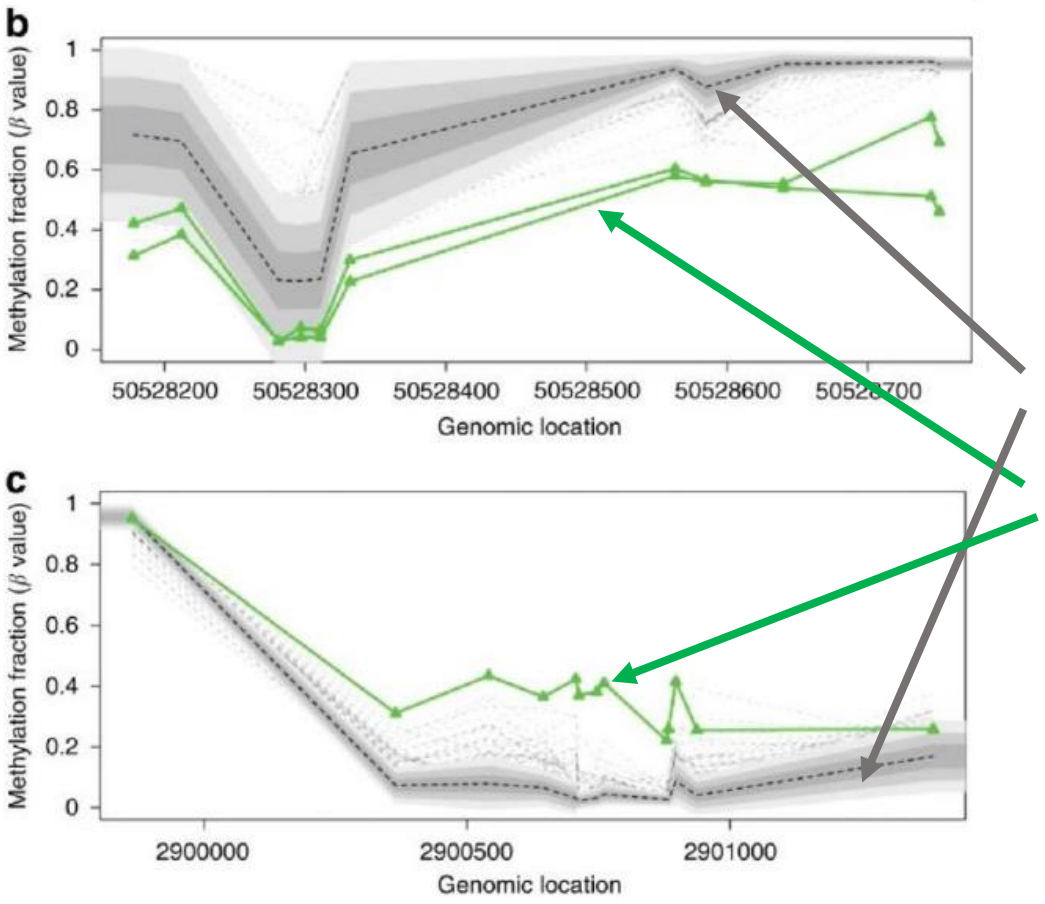
- Episignatures: Syndrome-specific DNA methylation changes across multiple loci

  Episignatures for 14 neurodevelopmental disorders and congenital anomalies syndromes (Aref-Eshghi E *et al* 2019 – AJHG).

- Epivariants and episignatures associated with unresolved cases will be used to find new candidate genes
- Controls: existing epigenomic profiles from 1200 well characterised individuals

New approach:

Epivariation or epimutations: DNA regions with aberrant methylation values



Barbosa, M. et al. (2018)

Distribution in normal Cases (i.e reference panel)

Undiagnosed cases

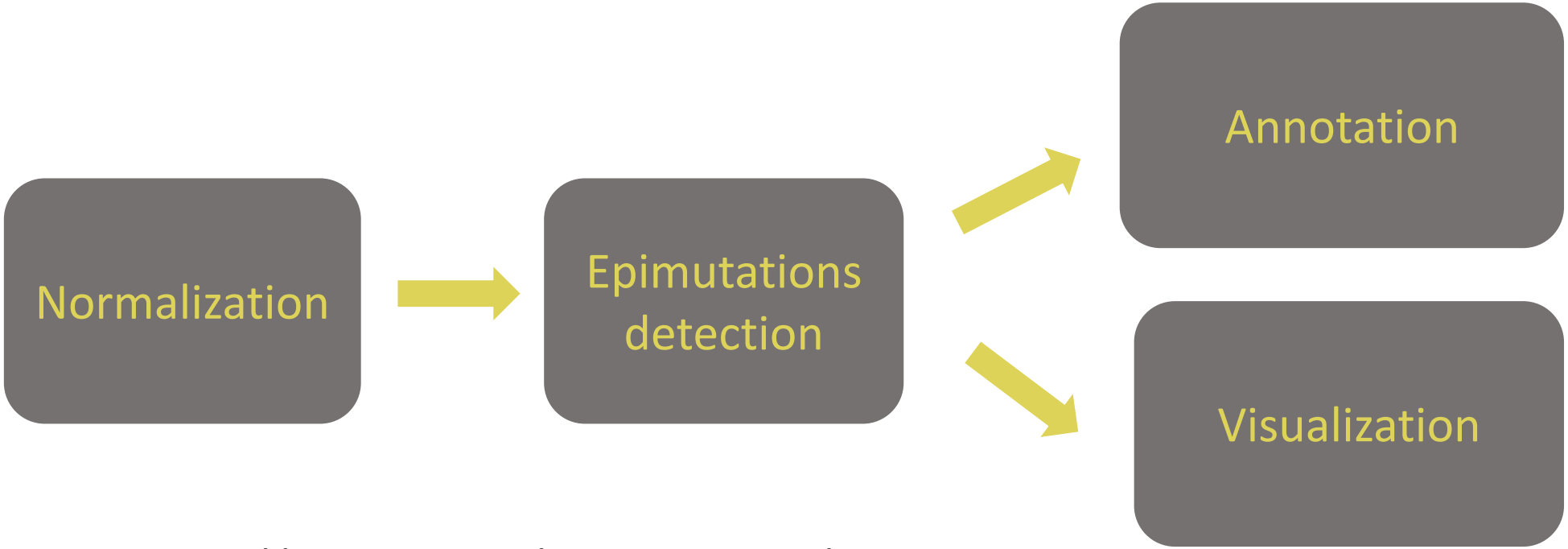
Recurrent hypomethylation at the promoter, 5' UTR, and first exon of *MOV10L1* in 2 unrelated probands.

Hypermethylation of *ZNF57* in one proband.

Epimutation detection

epimutacions bioconductor's package

- common repository of all the powerful methods for epimutation identification by using different outlier detection techniques



Devel version: https://github.com/isglobal-brge/epimutacions

SLIDES REMOVED

Acknowledgments

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Óscar Mira

Juan Ramón González
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Mariona Bustamante
Pol Cuscó
Alejandro Cáceres

Bea Morte
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H.U. Son Espases: Jordi Rosell
H. La Paz María Palomares
H.C.U. Lozano Blesa: Feliciano Ramos
H.C.U. Virgen Arrixaca: Encarna Guillén
H. Clínic: Susana Puig

IDIBAPS:
Antònia Ribes, Frederic Tort
Laia Rodríguez
Glòria Garrabou

IDIBELL-IGTP:
Aurora Pujol
Conxi Lázaro

IMIM:
Luis Pérez-Jurado

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Daniel Grinberg

HSP:
Pia Gallano

Parc Taulí:
Míriam Guitart

VHIR
Alfons Macaya
Eduardo Tizzano

Técnicas ómicas en el diagnóstico de enfermedades raras

Día 1

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Uso y análisis del transcriptoma y el epigenoma

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