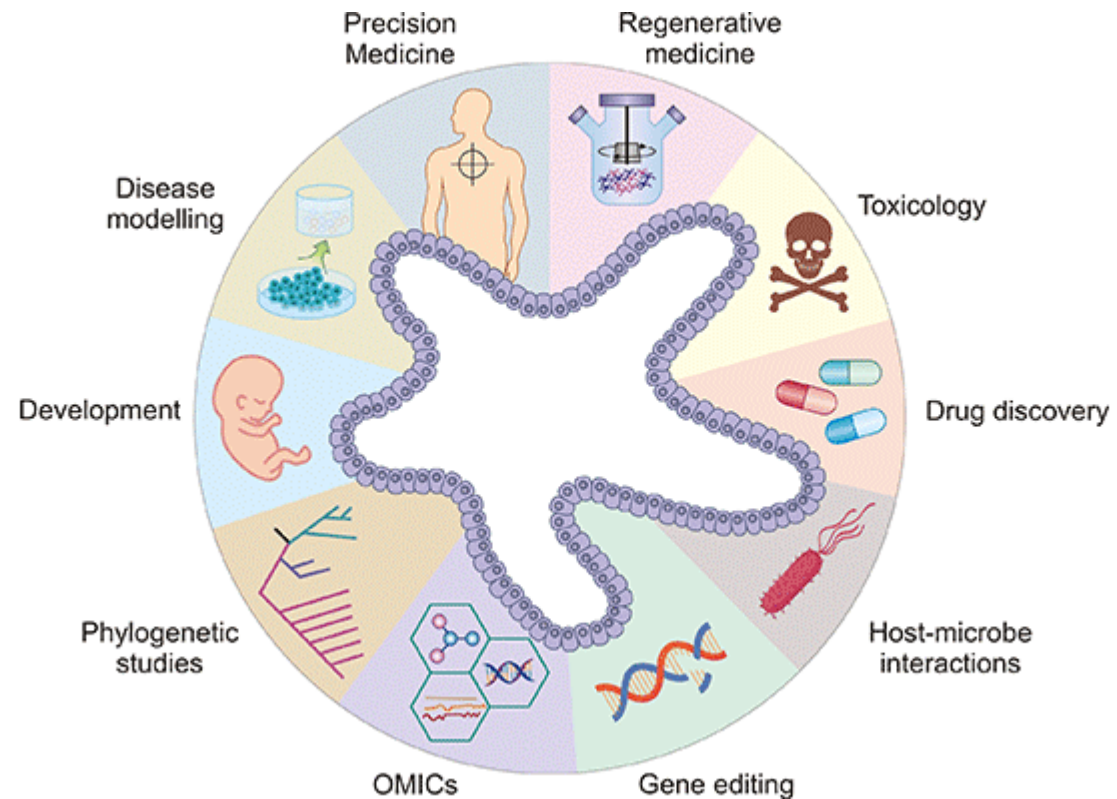


Core Facility Genomics (CFG)



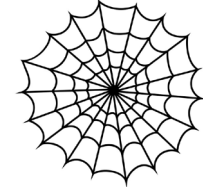
Location Vumc, RDC, ZH 2 J 055 and 057

Location AMC, G2-105.3

Typical CFG Workflow

(1) Intake

- Research questions / experiment design
- Planning / costs
- Data analysis



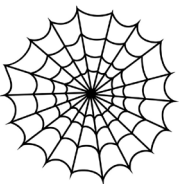
(2) Registration, sample delivery

- User and experiment (type) registration
- Sample delivery

(3) Technical Processing

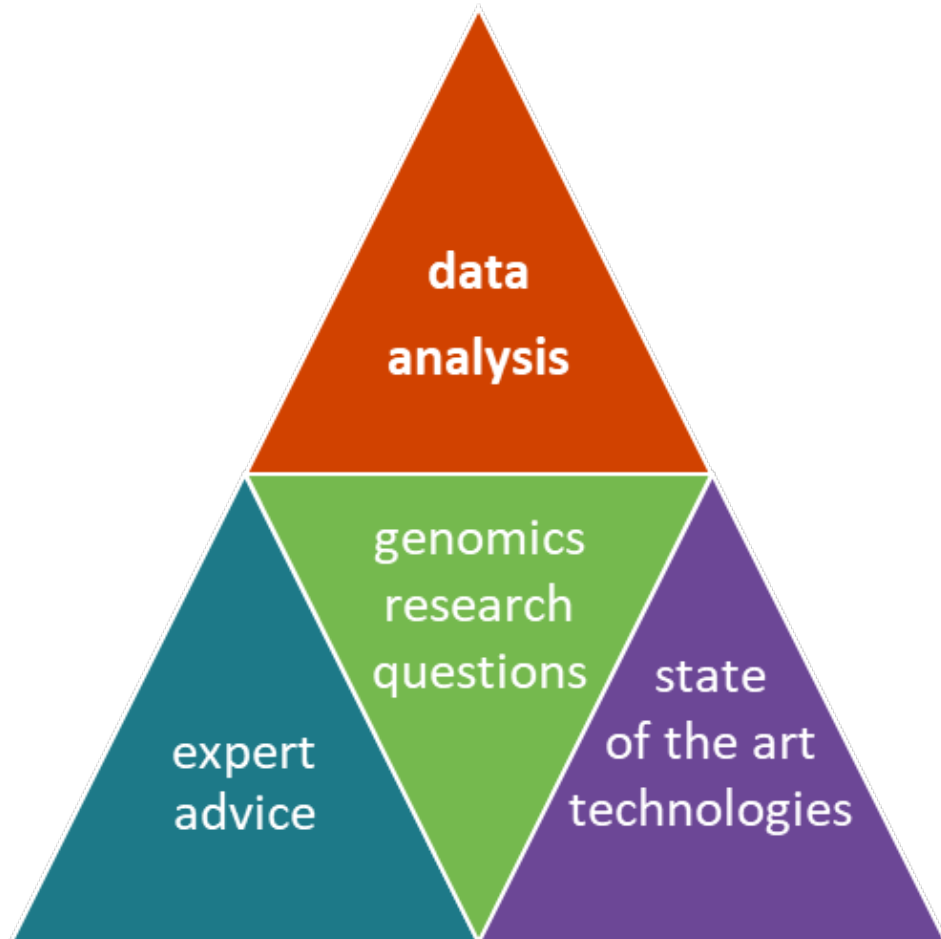
(4) Data

- Data transfer
- Data Archiving
- **Data analysis (HPC)**



The Core Facility Genomics, leverage the quality of your research data, on time and cost effective.

Consultancy team CFG



Scientific and technical consultants
4 lead technicians
17 technicians (partially part-time)
1 support staff
Bion-inf team

Core Facility Genomics (CFG)



CFG CONTACT

email - cfg@amsterdamumc.nl

website - <https://cfg.amsterdamumc.nl>

Location Vumc, RDC, ZH 2 J 055 and 057

CFG lab: 06 46126777

Location AMC, G2-105.3

CFG lab: 06 22297916



Topics

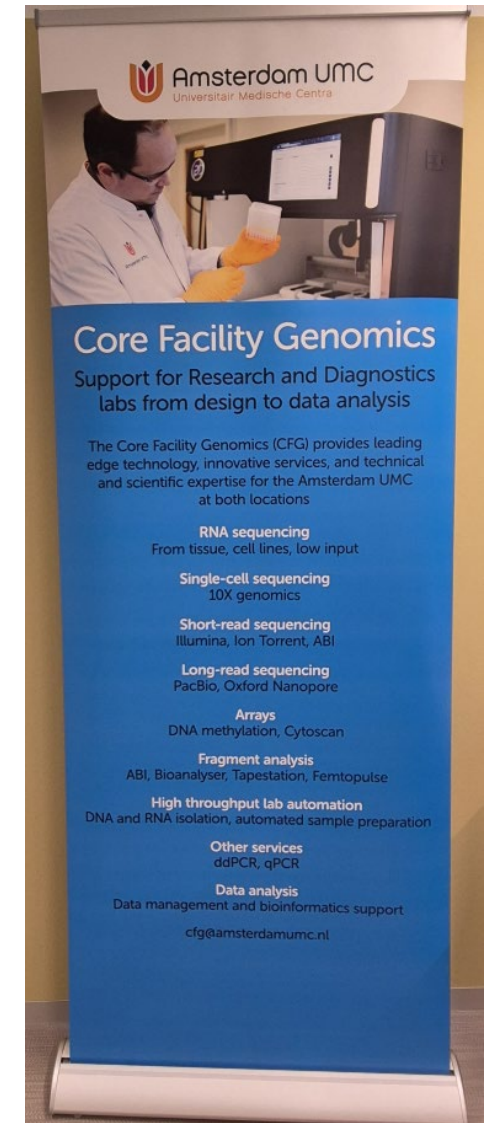
- CFG locations, user registration and CFG equipment
- DNA/RNA isolation
- Next Gen Sequencing Platforms
- Single cell/spatial
- Amsterdam UMC Core Facilities

CFG locations



RDC	
Pre-PCR lab	ZH2J055
Post-PCR lab	ZH2J057
Offices	ZH2J078a

AMC	
Pre-PCR lab	K2-252
Post-PCR lab	G2-105.3
Offices	K2-218



CFG user Registration

- Before use of labs, equipment and services
- Central registration in CFG LIMS
 - activate badge access
 - enable direct communication for CFG
 - access to equipment reservation system in CFG LIMS
 - billing of costs if applicable
 - communication of CFG house rules



House rules CFG lab

House rules of CFG can be found in K2 and via CFG website: [CFG - Huisregels voor gebruikers](#)
(and are sent in email after registration).

- Pre- and post-lab areas are separated. No post-PCR materials in the pre-lab area.
- UV cabinets have dedicated pipettes for additional compartmentalization and clean working.
- A CFG employee is available for questions between 8:30 and 17:00; due to the large building, you can call 06 46126777 if necessary.
- Lab coat is mandatory.
- One glove policy according to AmsterdamUMC policy.
Do not touch pipettes, computers, doors, etc. with a gloved hand.

Always leave the lab in a better condition than you found it.

House rules CFG lab

House rules of CFG can be found in K2 and via CFG website: [CFG - Huisregels voor gebruikers](#) (and are sent in email after registration)

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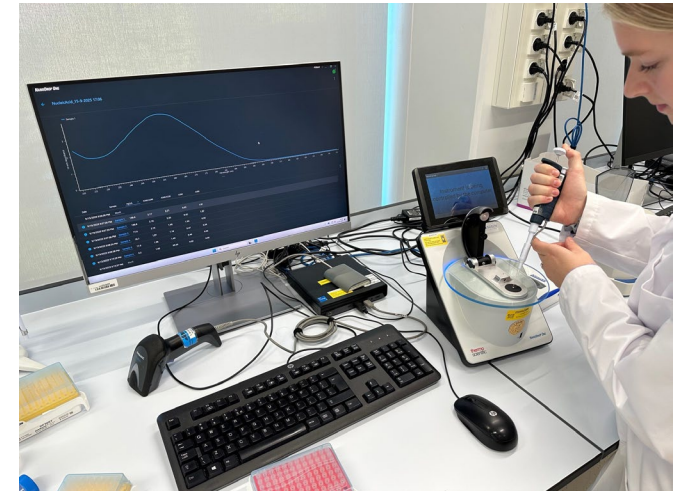
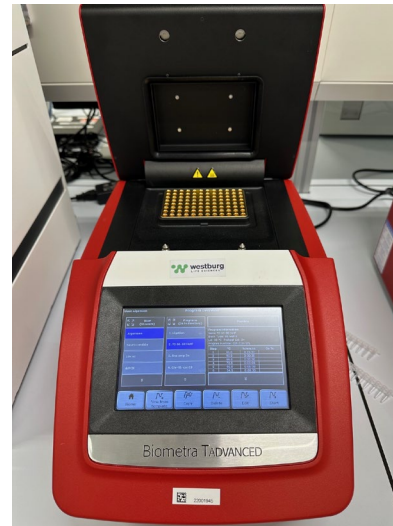
Equipment at the CFG

- Equipment provenance
 - Equipment used at the Human Genetics
 - Equipment acquired by the CFG
 - Equipment from other labs
- Equipment usage types
 - Free open use
 - Paid use (maintenance, purchase and / or running costs)
 - Use by CFG only. (paid services)



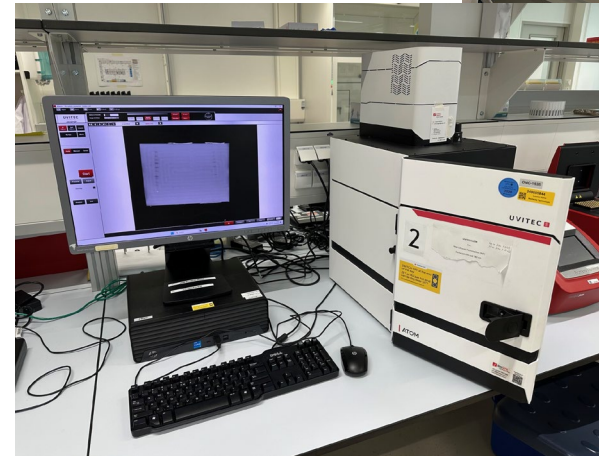
Equipment at the CFG

- Machines for general use:
 - Nanodrop
 - Thermocyclers 96 en 384 well
 - Qiaxcell (RDC)
 - 2d Tube readers
 - Centrifuges
 - Plate sealers
 - Tissue lyser (AMC)
 - Magna lyser (AMC)



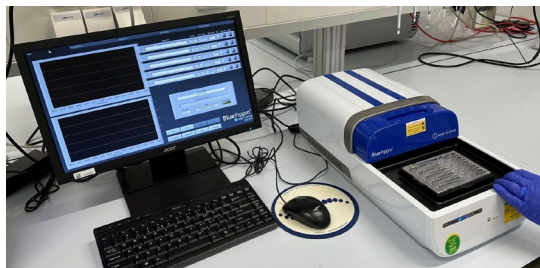
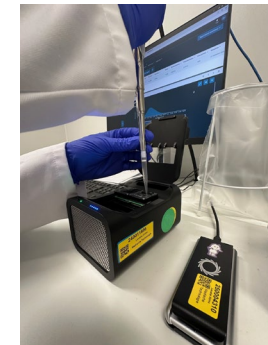
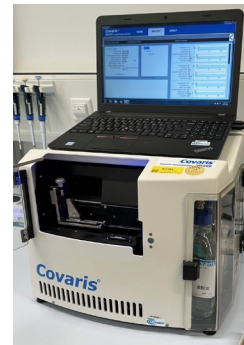
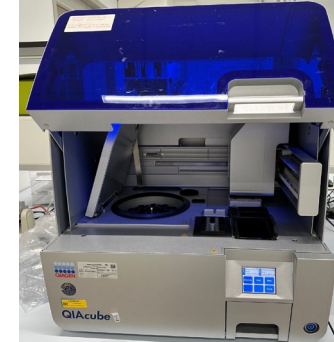
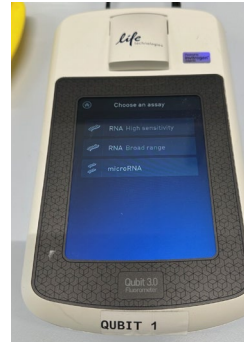
Equipment at the CFG

- Machines for general use:
 - Plate readers
 - Tecan (RDC)
 - Spectramax (RDC)
 - Fluorstar omega and optima (AMC)
 - Imagers
 - Las4000 (AMC, fluorescence and chemiluminescence)
 - Uvitec for fluorescence (NIR/IR) and chemiluminescence (ECL ed.) western blots (RDC)
 - Auto blot and image system WES (AMC)



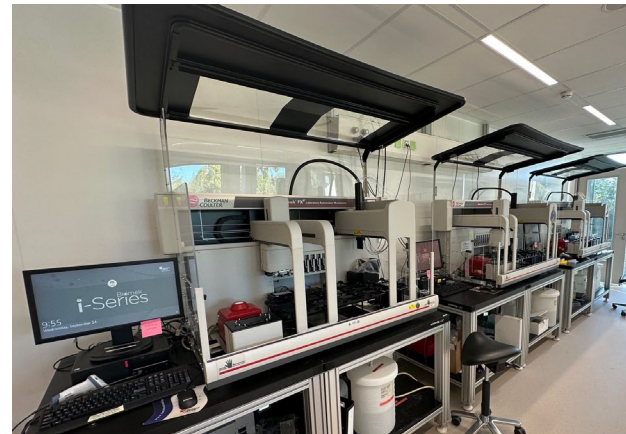
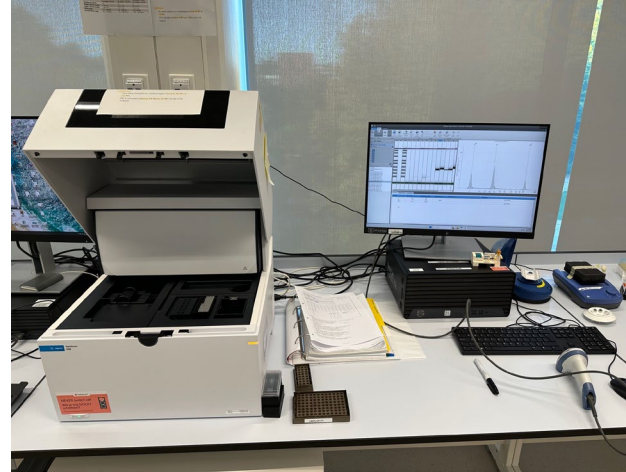
Equipment at the CFG

- Machines for paid use:
 - Qubit
 - ddPCR
 - 10x Single Cell
 - Blue Pippin
 - Qiacube
 - LC480
 - Covaris
 - Nanopore sequencing



Equipment at the CFG

- Use by CFG only:
 - Tapestation
 - ABI 3730xl
 - Biomek FXp robotics
 - Hamilton Star robotics
 - Illumina sequencers
 - Qiagen DNA / RNA isolators
 - Autogen DNA isolator
 - Promega DNA / RNA isolators
 - Speedvac
 - Thermofisher array platform
 - Megaruptor



Additional info / services

- Automation of laboratory process (pipetting robots)
- Bulk purchase of chemistry and kits
- AVG compliant outsourcing
 - GSA / EPIC array
 - PACBIO long read sequencing
 - Femtopulse
- See our website for full list of current services

cfg.amsterdamumc.nl

DNA and RNA isolation

1. Intake
2. Registration (LIMS)
3. Isolation (Devices)
4. DNA release verification (criteria)
5. Test, sending, storage (@biobank AUMC)



Afdeling Humane Genetica
Core Facility Genomics (CFG)

Postadres

Amsterdam UMC, locatie VUmc
Afdeling Humane Genetica
Research & Diagnostiek Centrum ADORE; ZH 0 J 003
Antwoordnummer 7700
1000 SN AMSTERDAM
Telefoon: 020-4440747

afleveradres voor koeriers (kantooruren 8.30-17.00):
Research & Diagnostiek Centrum ADORE; ZH 0 J 003
Van der Boechorststraat 6B
1081 BT AMSTERDAM

E-mail: cfg@amsterdamumc.nl
website: <https://cfg.amsterdamumc.nl>
Telefoonnummer: 06-46126777

Coördinator research project: email en telefoonnummer

Gegevens aanvrager:

Naam aanvrager:

zh/installing:

afdeling:

adres:

plaats:

uw referentie:

telefoonnummer/sein:

Materiaal

Monsters die niet zijn voorzien van een deugdelijke identificatie worden geweigerd.

× buizen (< ml) EDTA bloed voorzien van **identifiser** verzenden per post bij **kamertemperatuur** (niet onder gecontroleerde omstandigheden).

Datum afname:

**Projectnaam/ID
Afspraken isolatie**

Aanvullende afspraken project specifiek

Methode:

FlexStar 5 ml/Qiasymphony 2 ml protocol Neem over van
submission formulier.



barcode project

In te vullen door het laboratorium; hieronder niet schrijven en geen stickers plakken

Ingeschreven door (paraaf):.....

Studienaam

A
Tot Minerva live is,
dit vakje gebruiken
om aan te geven dat
CORE inschrijving
doet.

Aanvraagformulier Biobanking

Extern ID nr. barcode
sticker

Geboorte datum enkel op
specifiek verzoek
onderzoeker

Geslacht enkel op
specifiek verzoek
onderzoeker

Per persoon een aanvraagformulier volledig invullen



DNA/RNA isolation, source materials

- EDTA Blood (fresh/frozen) DNA
- PAX blood (RNA)
- Buffy coat
- Saliva (oragene)
- Blood spot ("heel prick")
- Cell-free fetal DNA
- Tissue (chorion villi, skin biopsy etc)
- Urine
- FFPE (paraffin)
- ???

Qiasymphony (2x)

DNA and RNA extraction by Magnetic beads

- 1-96 samples/run (200/day)
- Fast
- Fully automated
- Max. 2ml blood as source (DNA)
- RNA possible
- DNA ready to use



Autogen flex+

DNA extraction by precipitation

- 30 samples/run (90/day)
- Slow
- Fully automated
- Default 5 ml blood as source (max 10 ml)
- DNA should be dissolved (o/n)



Maxwell (3x)

DNA and RNA extraction by Magnetic beads

- 1-16 samples/run
- Semi-automated
- Low input
- DNA ready to use
- RNA possible



DNA/RNA release verification

- OD measurement (standard for DNA)
- Qubit measurement (optional for DNA, standard for RNA)
- Tapestation (optional for DNA, standard for RNA)

- Criteria DNA
 - > DNA back to applicant
 - > DNA for inhouse testing, sending
 - > storage at [AmsterdamUMC biobank](#)
- >30µg (from 1,8 ml blood)
- OD260/280 1,6 -2.0

DNA Storage 4° C / RNA storage -80° C (2D barcode tubes)



Next Gen Sequencing Platforms

- iSeq100
- NextSeq2000
- NovaSeqXplus
- MinIon
- P2 solo
- REVIO

Illumina

- short reads
- clonal amplification



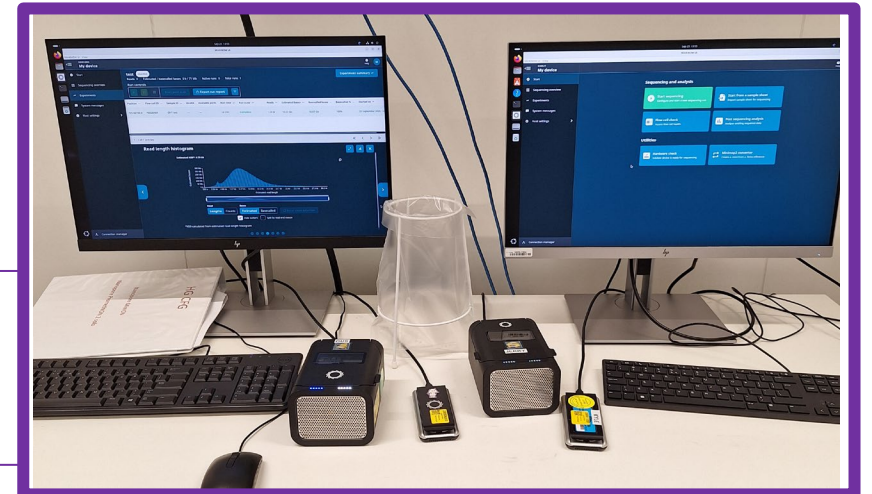
PacBio

- long reads
- single molecule



Oxford Nanopore

- long reads
- single molecule

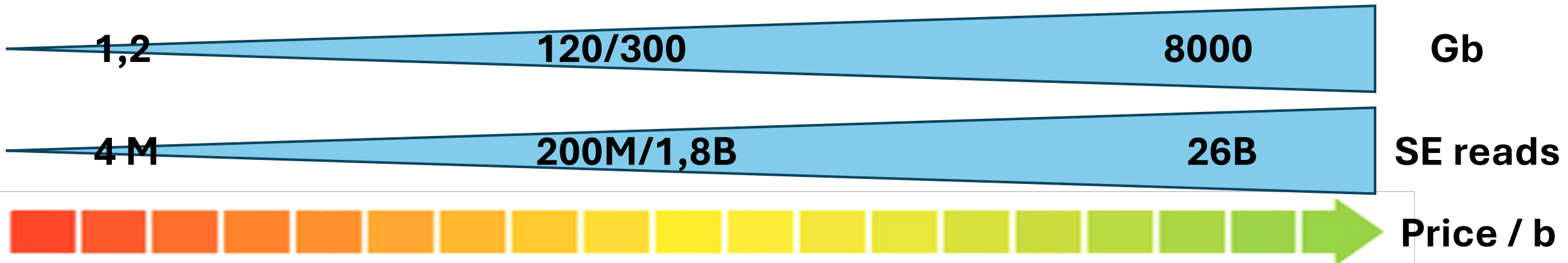


Illumina Next Gen Sequencing

iSeq100

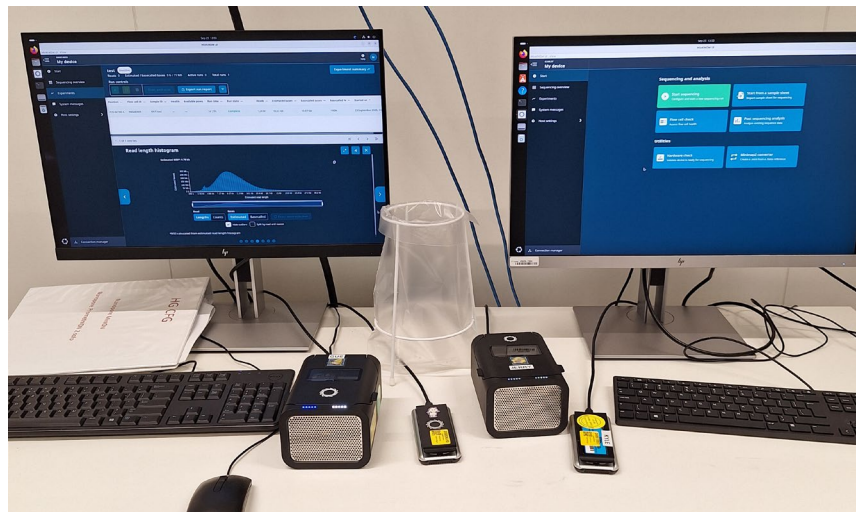


NextSeq2000



Oxford Nanopore

	flongle	MinION	PromethION 2
channels per flow cell	126	512	2675
number flow cell per device	1	1	2
max output per flow cell	2,8 Gb	50 Gb	290 Gb
max output per device	2,8 Gb	50 Gb	580 Gb
run time	1 min -16 h	1 min -72 h	1 min -72 h



REVIO - PacBio

- SMRT cell 8M
- 8 million ZMWs
- 4.000.000 reads
- 30uur

Samenwerking
RadboudUMC





Single strand

- No amplification, no PCR bias
- No copying errors
- Long reads
- Structural variants
- Modifications stay intact
- Very low signal

Differences

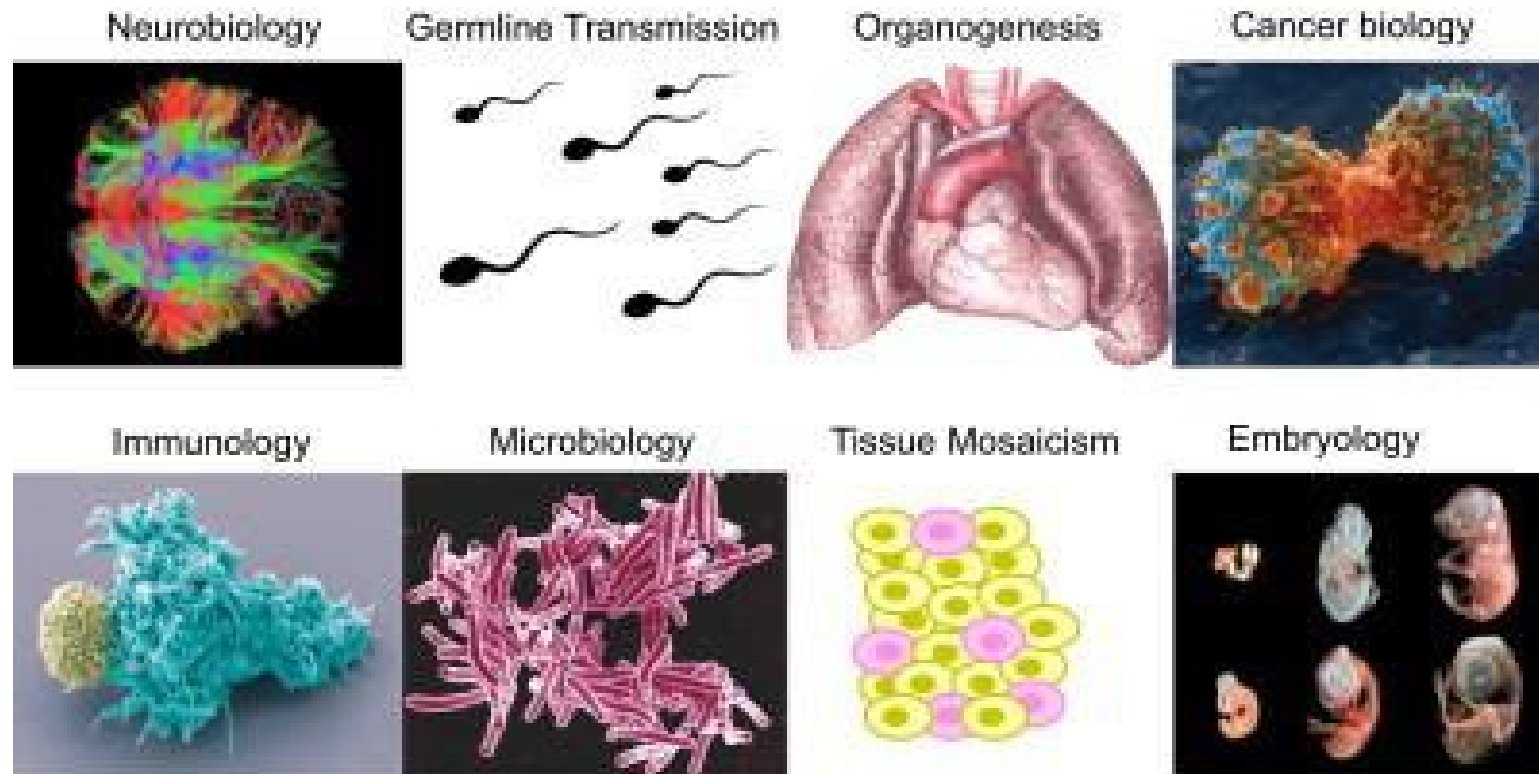


Clonal Amplification

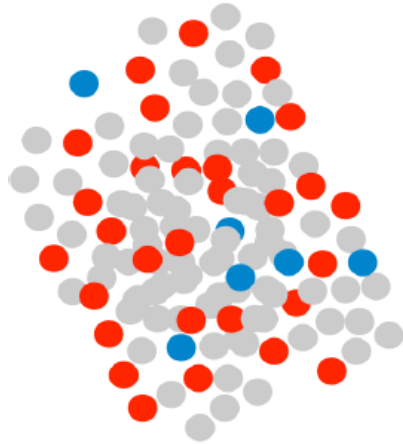
- Strong signals
- PCR errors are averaged
- Loose modifications
- PCR bias/errors
- Short reads
- High throughput

Single cell

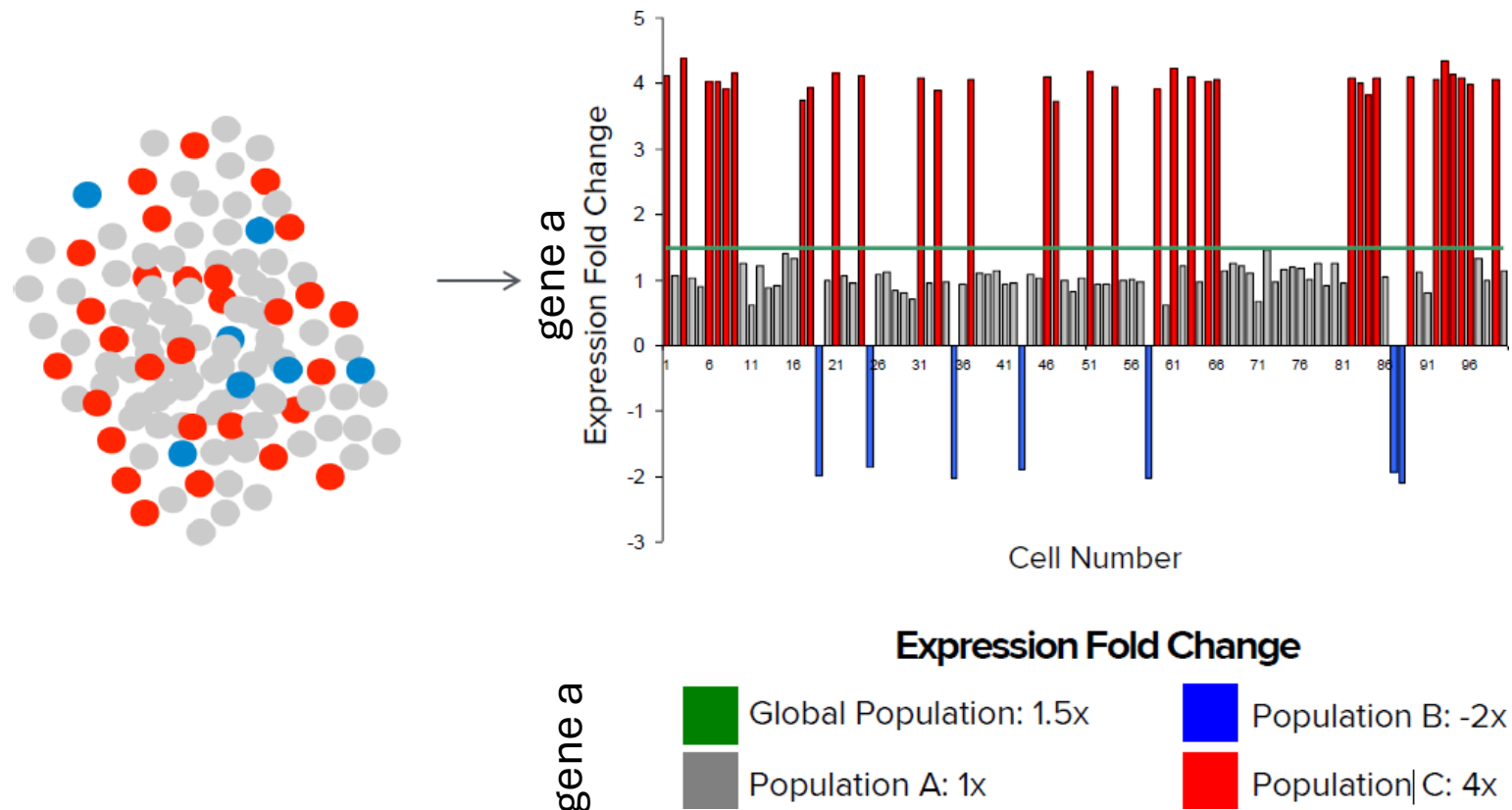
Single cell biology impacts most areas of research



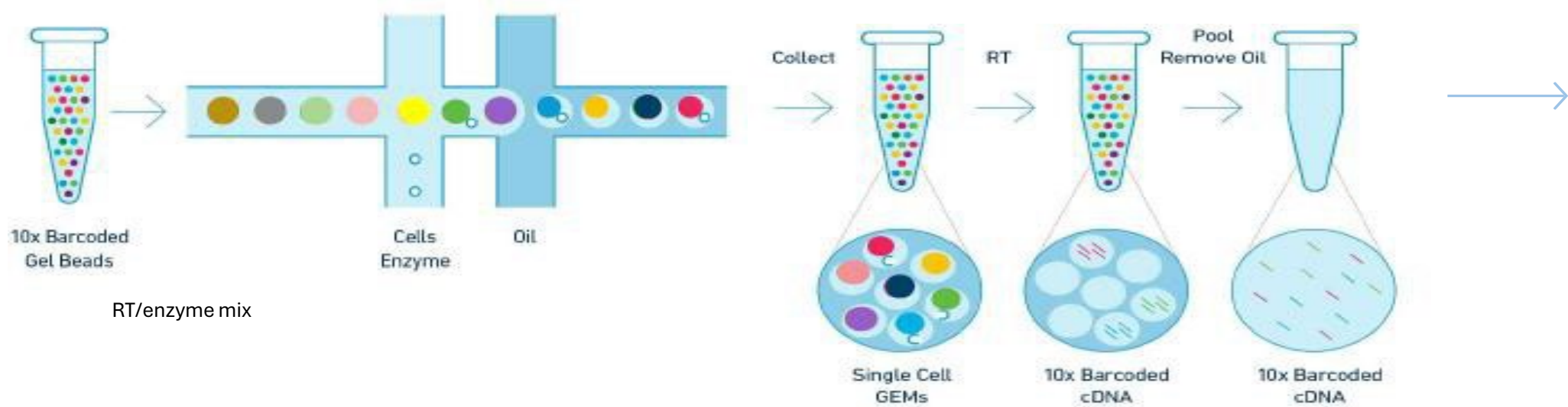
Individual cells behave differently from the average of many cells



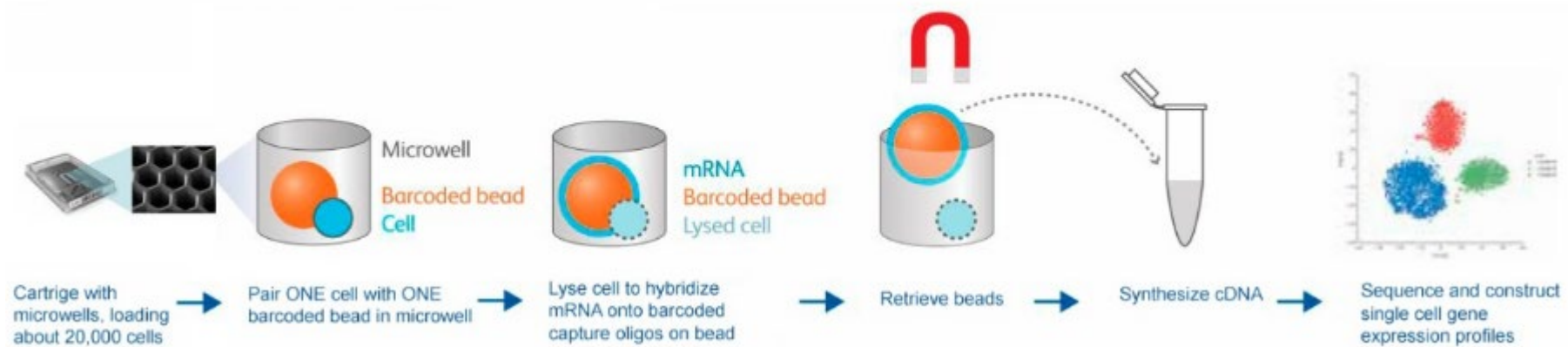
Individual cells behave differently from the average of many cells



10X Microfluidics droplet encapsulation



BD Rhapsody protocol



What cell?

transcript copies?

Which gene?

Single Cell data analysis

Top differentially expressed genes per cell cluster
(UMI counts/cell)

Tissues

Gene name	Cluster 1	Cluster 2	Cluster 3
Abc1	28.52	0.03	14.7
Xyz2	4.56	8.33	30.85
Fgh3	8.94	17.44	1.27

Which cells and how many per type are present?
What are they expressing?

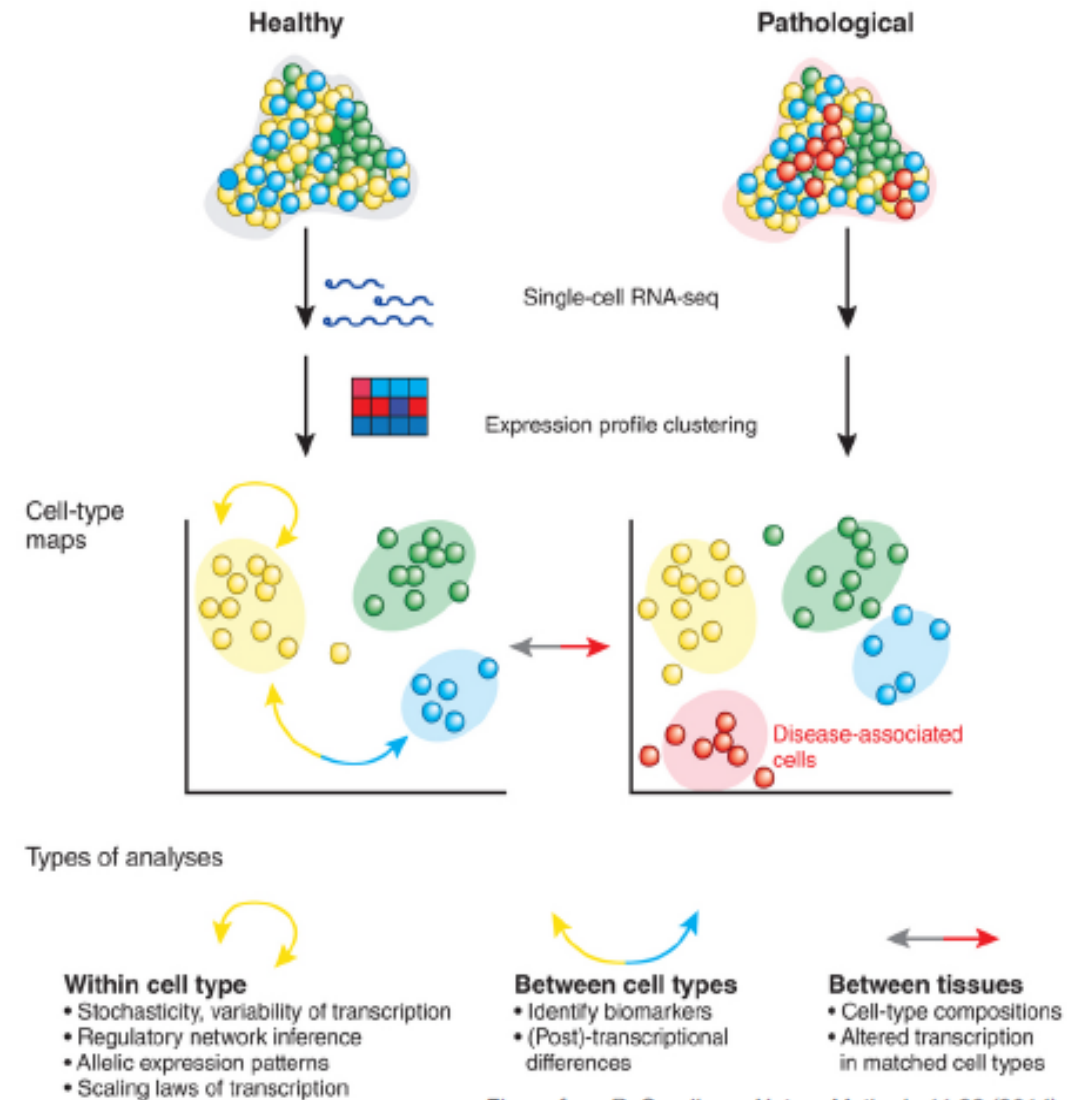


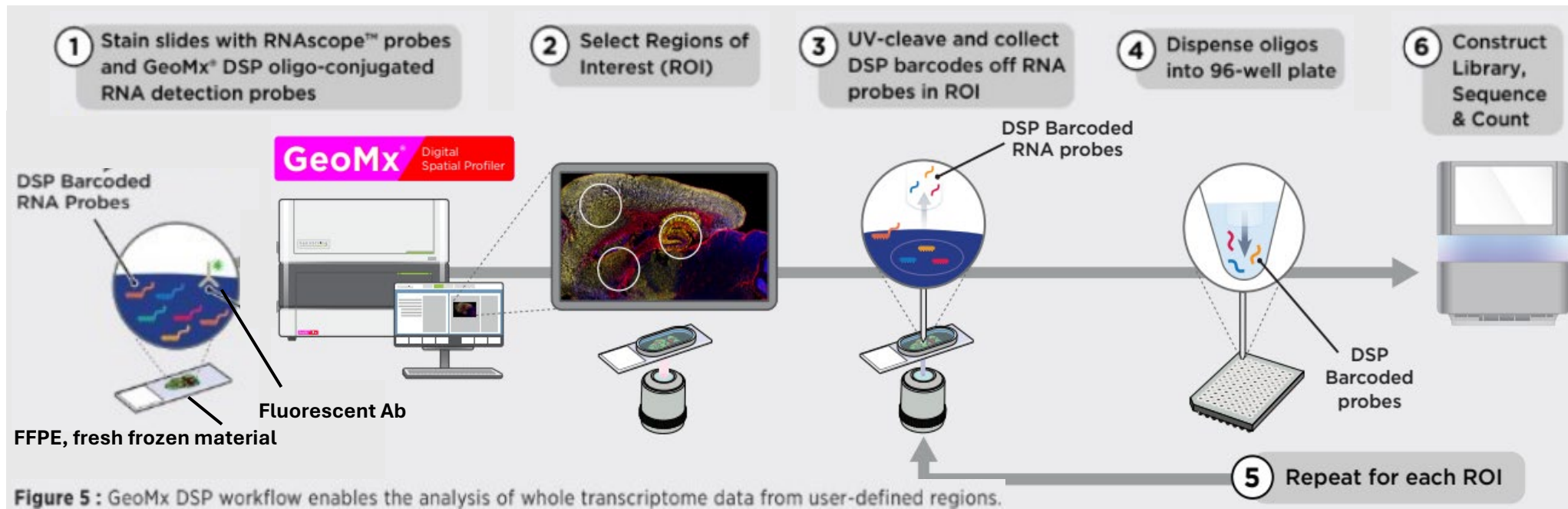
Figure from R. Sandberg, *Nature Methods* 11:22 (2014)

		10X Genomics		BD Rhapsody	
WTA, poly-A capture	singleplex	✓	Live cells, nuclei, fixed PBMC's	✓	Live cells, nuclei, fixed PBMC's
	multiplex	✓ (HT-Ab or OCM)		✓ (HT Ab)	
Targeted RNA	singleplex	✓	Fixed cells or nuclei, WTA BC Probe Panel*	✓	Targeted Probe Panel on cDNA*
	multiplex	✓		✓	
V(D)J		✓ (BCR and/or TCR)*		✓ (BCR and/or TCR)*	
ATAC		✓		✓	
CITEseq		✓		✓	
Multiome WTA & ATAC		✓		✓	

*Human & Mice

Spatial Transcriptomics

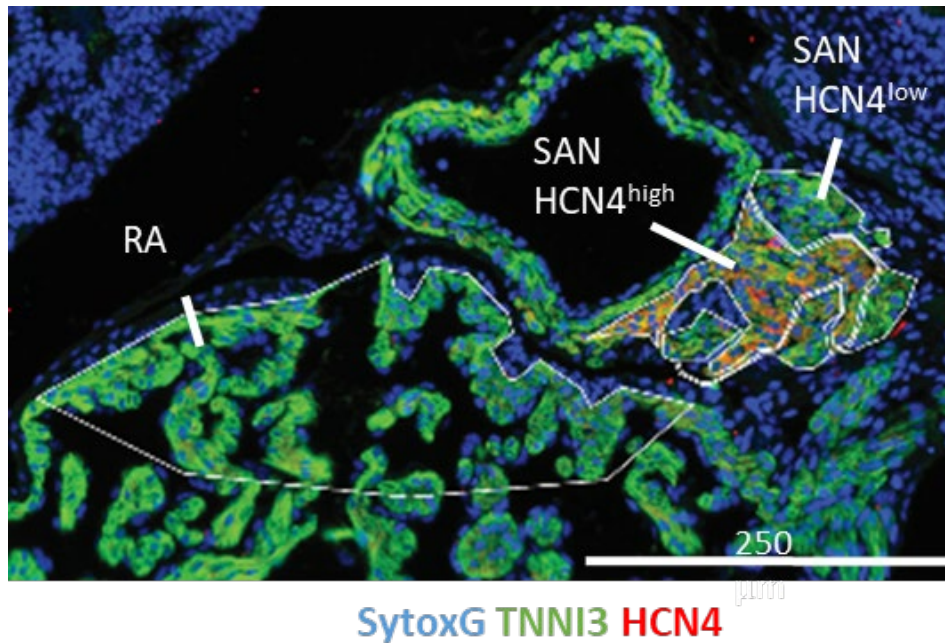
GeoMx DSP Nanostring, a targeted approach



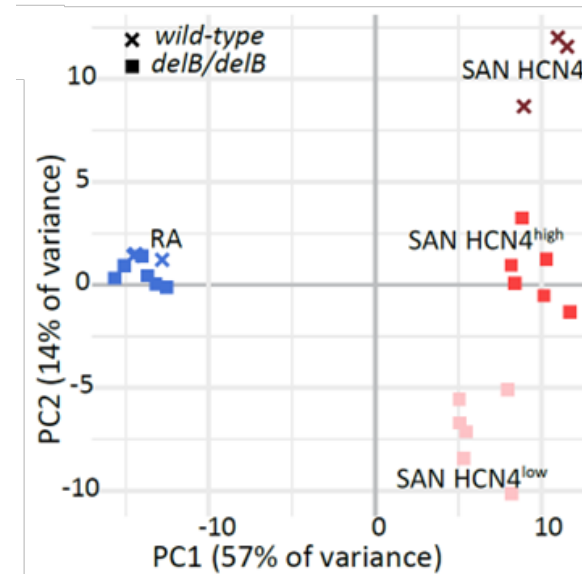
MCCF

CF
G

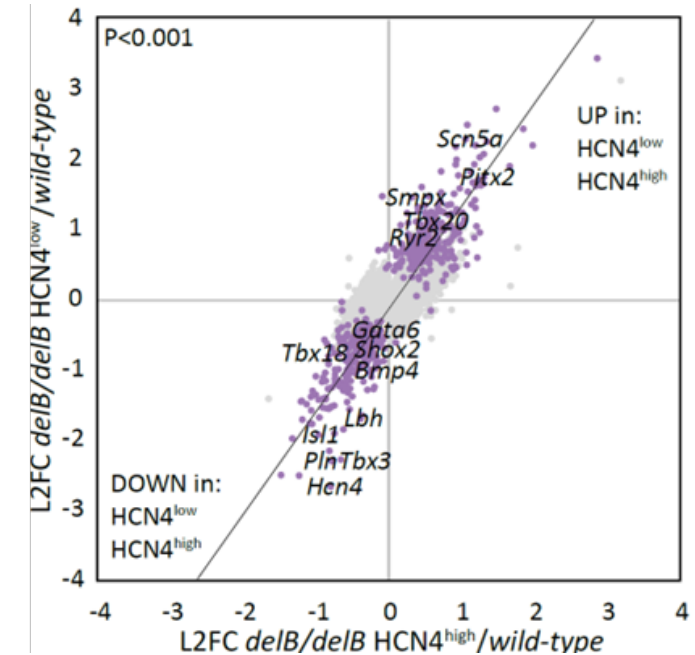
Example results Spatial Transcriptomics



GeoMX DSP image



Result after coupling the Illumina seq data to the ROIs



L. van de Maarel,; NanoString GeoMx DSP; Mouse Whole Transcriptome Atlas (22 k transcripts)

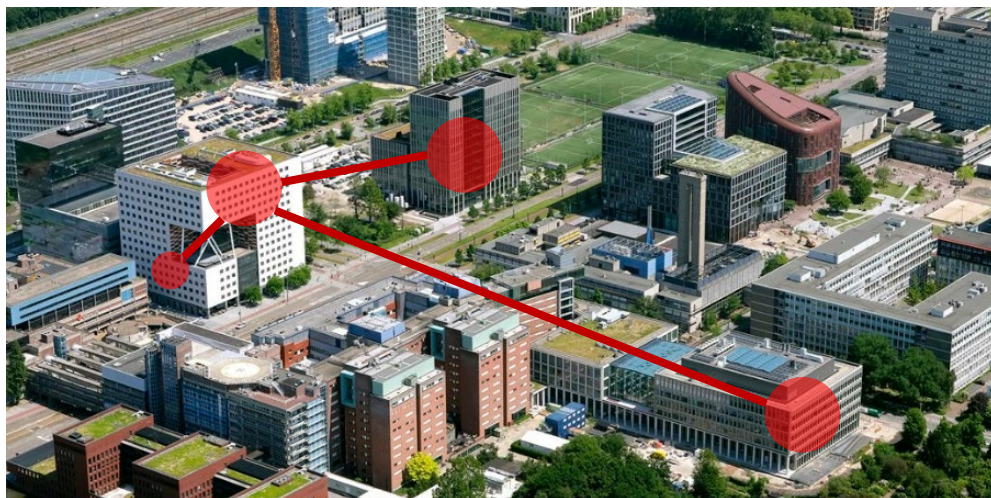
Amsterdam UMC Core Facilities

Core Facility	Microscopy & Cytometry CF	Proteomics	Genomics	Metabolomics	Microscopy & Flow Cytometry CF
Acronym	MCCF-VUMC	ProCoRe	CFG	CFM	MCCF-AMC
Director	Juan J. Garcia Vallejo	Connie Jimenez	Alex Postma	Fred Vaz	Eric Reits
High Performance Computing and Research Data Management Hub (ACDC) Katy Wolstencroft and Daoud Sie					
Embedding	Molecular Cell Biology & Immunology	Medical Oncology	Human Genetics	Clinical Chemistry	Medical Biology
Location	Multiple buildings (O2, RDC , SWT) (VUMC Location)	RDC Building (VUMC Location)	RDC Building (VUMC Location)	F0 (AMC Location)	L3-M3 (AMC Location)

Core facilities | Amsterdam UMC

<https://www.amsterdamumc.org/en/research/core-facility.htm>

MCCF-VUMC



Central hub + 3 satellites
Strong collaboration with VU
12 operators, 36 instruments
>500 users, >70000 h/year



FOCUS



immuno



onco



neuro

Cellular heterogeneity (blood/tissue)

Throughput

Multiplexity

(Functional) imaging

Experimental Set Up

- Choosing the right technique
- Panel design and optimization
- Existing panels (via ITC)

Data Acquisition

- Training 1:1
- Operator assisted
- Troubleshooting

Data Analysis

- Bioinformatics support
- Established pipelines
- Commercial packages

Community

- Educational activities
- Focus technology groups
- Connecting projects



Satellite
@ RDC

Imaging Flow Analysis



Attune CytPix

ML2 Sorting



BD Melody

Biomarker assays



MESO QuickPlex
SQ120MM

Upright IF



Leica DM6000

Live Cell IF/BF



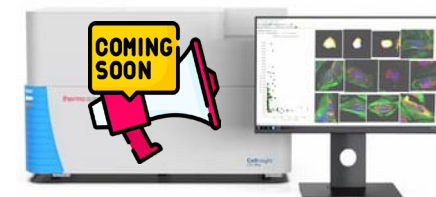
ECHO CellCyte X

Live cell CLSM



Nikon A1R

High-content
screening



CellInsight HCS

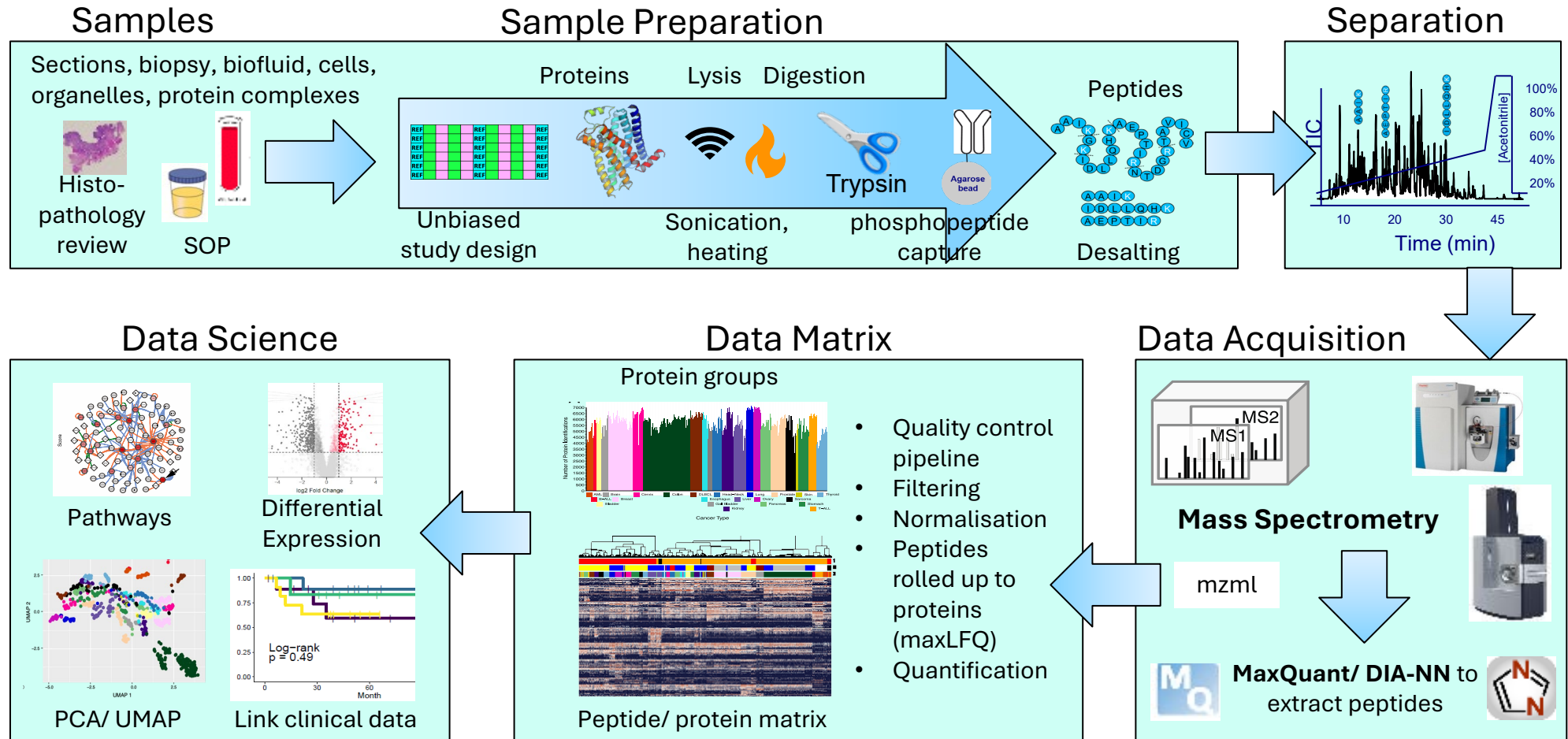
Proteomics Core Resource

Multi-disciplinary core team: 5 PhD level scientists
Infra: 3 MS platforms; **Users** from 16 AUMC depts

Focus:



- Cancer
- Cardio
- Method/ Other
- Microbio
- Neuro
- Immuno



Input: Biological/ clinical samples; **Support:** wet lab, full service MS and dry lab

Output: User-friendly excel file with data QC, dedicated statistics, heat maps; **Turn-around time:** ~4-6 weeks; **Fair** data management; Annual **course**; **>200 ProCoRe publications** in PubMed

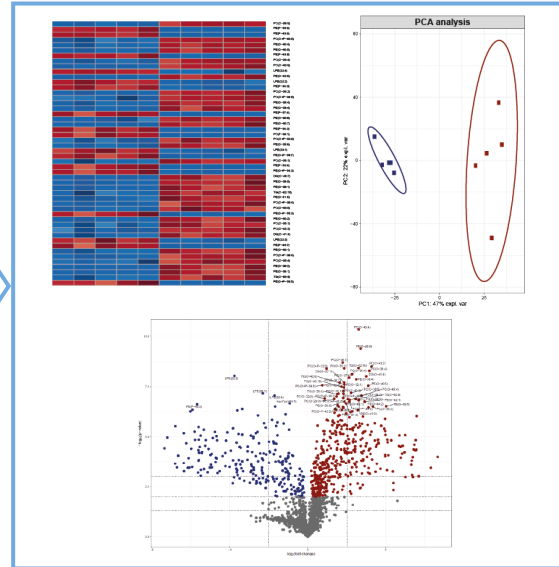
Core Facility Metabolomics

- Metabolomics
- Lipidomics
- Fluxomics
- Targeted assays

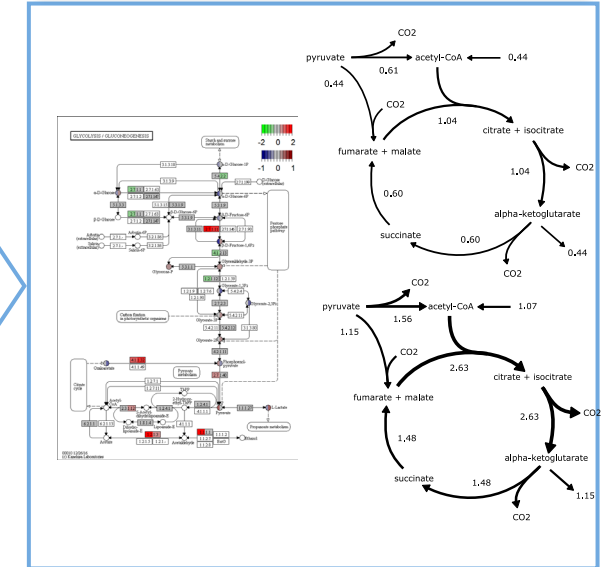
State-of-the-art Mass spectrometry



Bioinformatics/ data interpretation



Pathway analysis/ understanding metabolism



>20.000 features

>2500 lipid species

>300 polar metabolites

- Help with:
 - experimental design
 - data analysis
 - interpretation
 - data visualization

Including:

- Untargeted analysis
- Targeted analysis
- >1800 annotated

Including:

- Phospholipids
- Triglycerides
- Spingolipids

- Glycolysis, TCA cycle
- PPP, Nucleotides
- Amino acids

MCCF - AMC



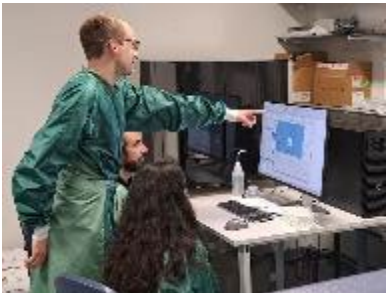
fluorescence microscopy facility:

- 3 operators, 2 data analysts (via NL-Biolmaging), expertise center Leica
- embedded in Leeuwenhoek Center Amsterdam Microscopy center (LCAM) with NKI and UvA, leading NWO Roadmap NL-Biolmaging and coordinating Dutch microscopy in Euro-Biolmaging



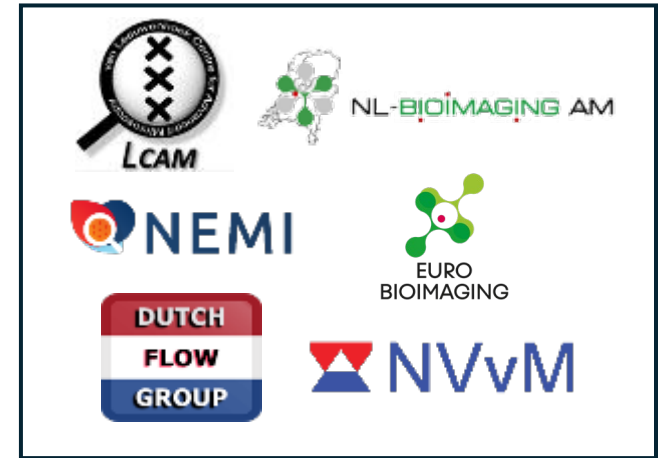
Electron Microscopy Center Amsterdam (EMCA) :

- Nicole van der Wel + 3 operators
- embedded in GWI Roadmap Netherlands EM initiative
- EM facility for AmsterdamUMC, NIN, Sanquin, NKI and ACTA



Flow cytometry:

- Kim Brandwijk + 2 operators, expertise center Sony



- UvA master track coordination Cell Biology & Advanced Microscopy
- 3 Doctor School courses on microscopy and data analysis
- Over 500 active users including biotech in 2024
- Current developments focus on visual omics to combine microscopy and omics core facilities
- Coordinating workgroups and demonstrations in the Dutch Microscopy Society
- Headed by Eric Reits

Advanced Compute & Data Core

***An integrated approach to
multi-omics and
bioimaging data & compute***

Genomics
Proteomics
Metabolomics
Flow Cytometry
Bioimaging



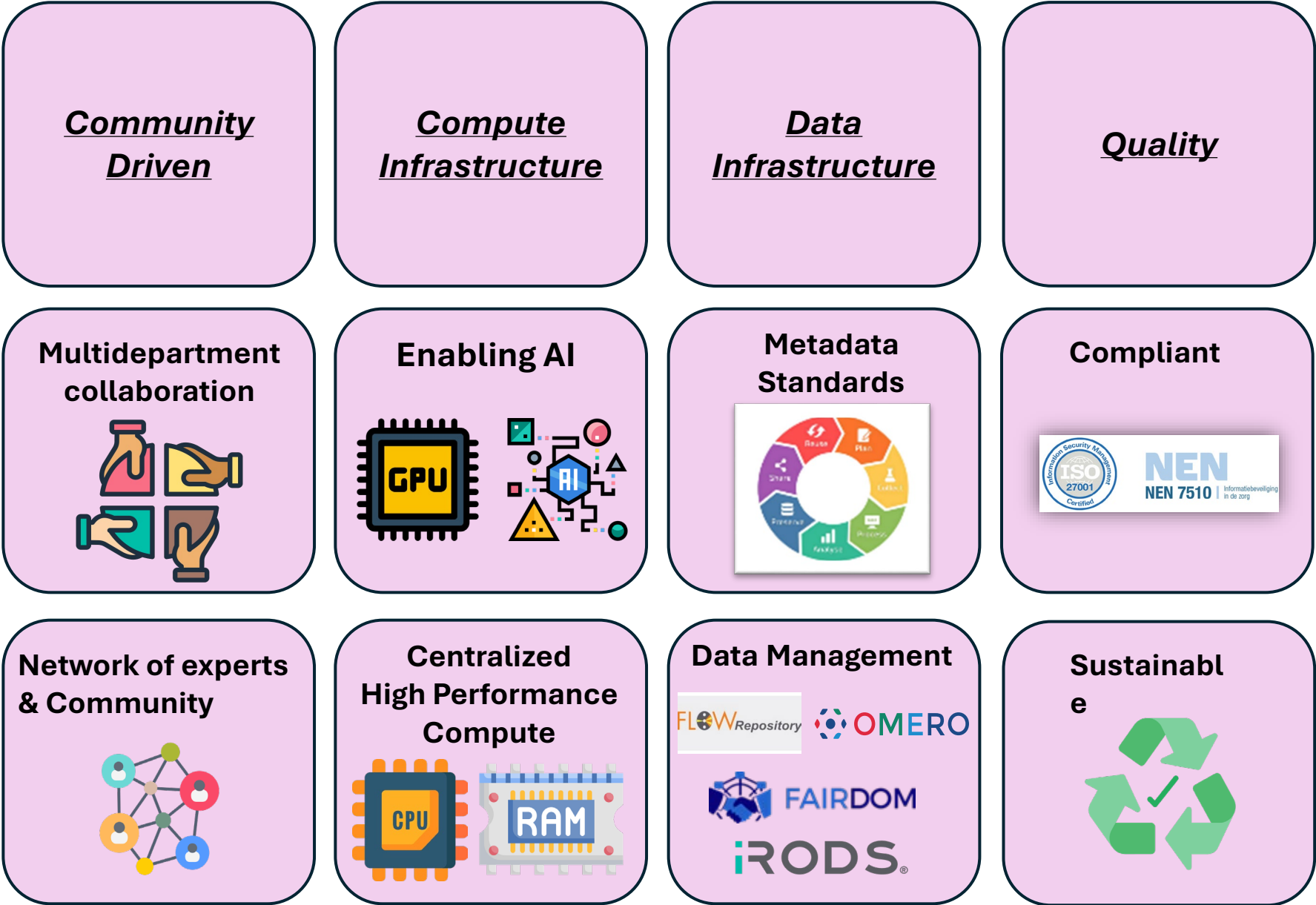


F_{indable}


A_{ccessible}


I_{nteroperable}


R_{eusable}

Discover the Core Facility Genomics with a lab tour

Contact

Cfg@amsterdamumc.nl

Intake to discuss project, experimental design, possibilities, alternatives costs.

Website

cfg.amsterdamumc.nl

Pricelist

[CFG: Cost and cost examples](#)

Protocols

[Extern via WebShare in CFG website](#)

[Intern via K2](#) <https://amsterdamumc.iprova.nl/>

CFG User meetings

[Previous CFG User Meetings](#) To subscribe for future meetings, please email Cfg@amsterdamumc.nl

Newsletters

[Newsletters Core Facility Genomics](#) To subscribe, please email CFG@amsterdamumc.nl