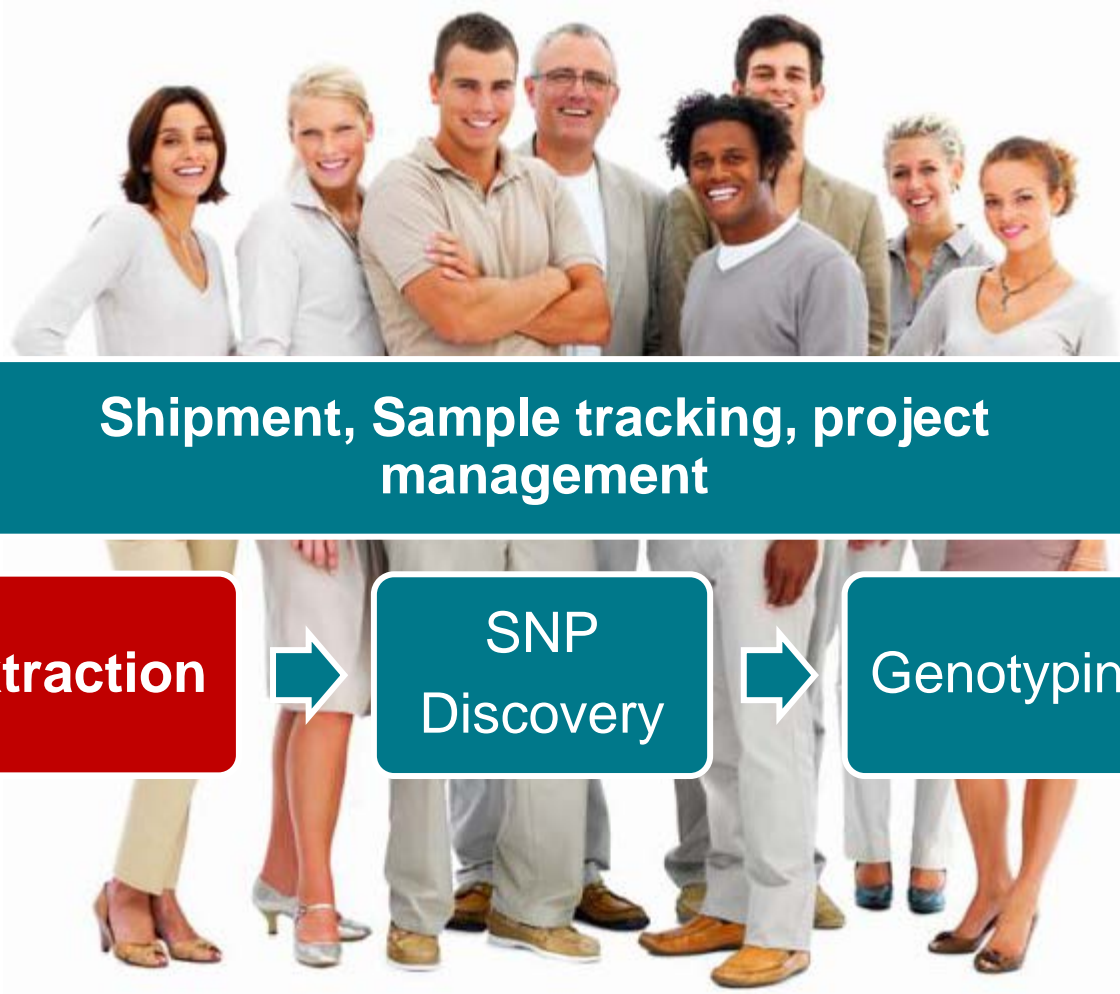




# Solutions for advancing cohort studies



# Cohort study project support services



**Shipment, Sample tracking, project management**

**Extraction**

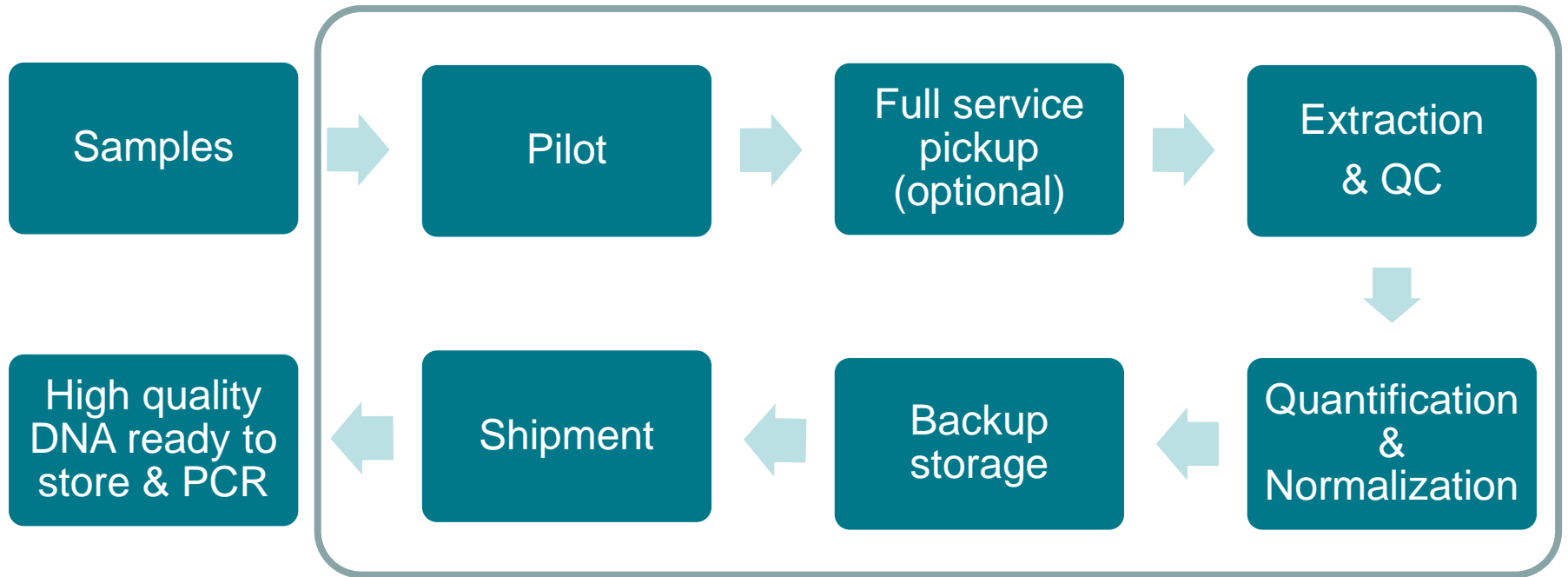


**SNP  
Discovery**



**Genotyping**

# Extraction module





# Extraction service package

|                       | Description  | Remarks  |
|-----------------------|--|--|
| <b>Volume</b>         | Up to 10 mL  |  |
| <b>Yield</b>          | Expected around 30 µg/ µL                                | Dependent on amount & quality of the received samples        |
| <b>Sample type</b>    | Blood, saliva, others                                    |  |
| <b>Quantification</b> | Full spectrophotometer graph (incl. OD 260/280) provided | Other quantification methods optional                        |
| <b>Normalisation</b>  | Normalised fraction shipped to customer                  | Aliquot volume and concentration determined in collaboration |
| <b>Backup</b>         | Aliquots stored at LGC Genomics (optional)               | Fraction determined in collaboration                         |
| <b>QC</b>             | 3 SNP genotyping (KASP) QC incl. gender test             | SNP data delivered to customer                               |
| <b>Certification</b>  | Options for full ISO certified service                   | DIN EN ISO 9001:2008   |
| <b>Plate type</b>     | Matrix 2D barcode storage racks included                 |  |
| <b>Shipment</b>       | All-in shipment arrangements                             |  |



# Pilot and pick-up

1. Pilot study (12 - 48 samples)
2. Full service pick-up of samples (bulk or batch)



Samples

Pilot

Full service  
pick-up

Extraction  
& QC

Quantification  
&  
Normalisation

Backup  
storage

Shipment

High quality  
DNA ready to  
store & use



# DNA extraction

## 3. DNA extraction

- Extraction volumes: up to **10 mL** (higher volumes possible)
- Sample types: whole blood, buffy coats, serum, tissue, saliva or buccal swabs (other matrices possible)

**DNAgenOTEK**  
Certified partner

Samples

Pilot

Full service pickup

Extraction & QC

Quantification & Normalization

Backup storage

Shipment

High quality DNA ready to store & use

# QC - Unique KASP SNP genotyping QC



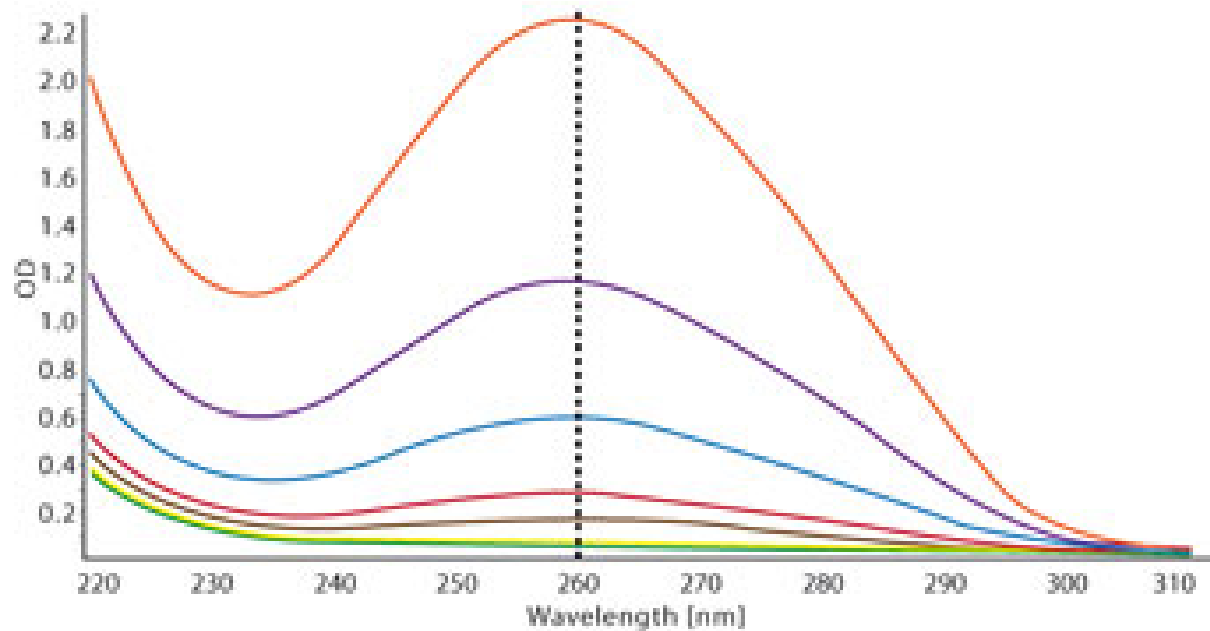
## 4. Genotyping QC

- 3 SNP Genotyping QC standard. Includes M/F assay for sample validation
- 24 SNP Genotyping panel  
(also applicable for validation of e.g. NGS samples)



# Quantification

## 5. Quantification of DNA / RNA concentration (UV measurement) (other quantification methods optional)





# Normalisation



## 6. Normalisation to desired concentration and volume (typically of a <math><1\text{ml}</math> fraction of total sample)



# Backup



## 7. Backup of aliquots (typically 10%) in walk-in-freezer storage facility UK



Samples



Pilot



Full service pickup



Extraction & QC



Quantification & Normalization



Backup storage



Shipment



High quality DNA ready to store & use

# Packaging and shipment

## 8. Shipment in industry standard 2D barcoded storage racks (Matrix)



Samples



Pilot



Full service pickup



Extraction & QC



Quantification & Normalization



Backup storage



Shipment



High quality DNA ready to store & use

# Enhance productivity..., do more with less

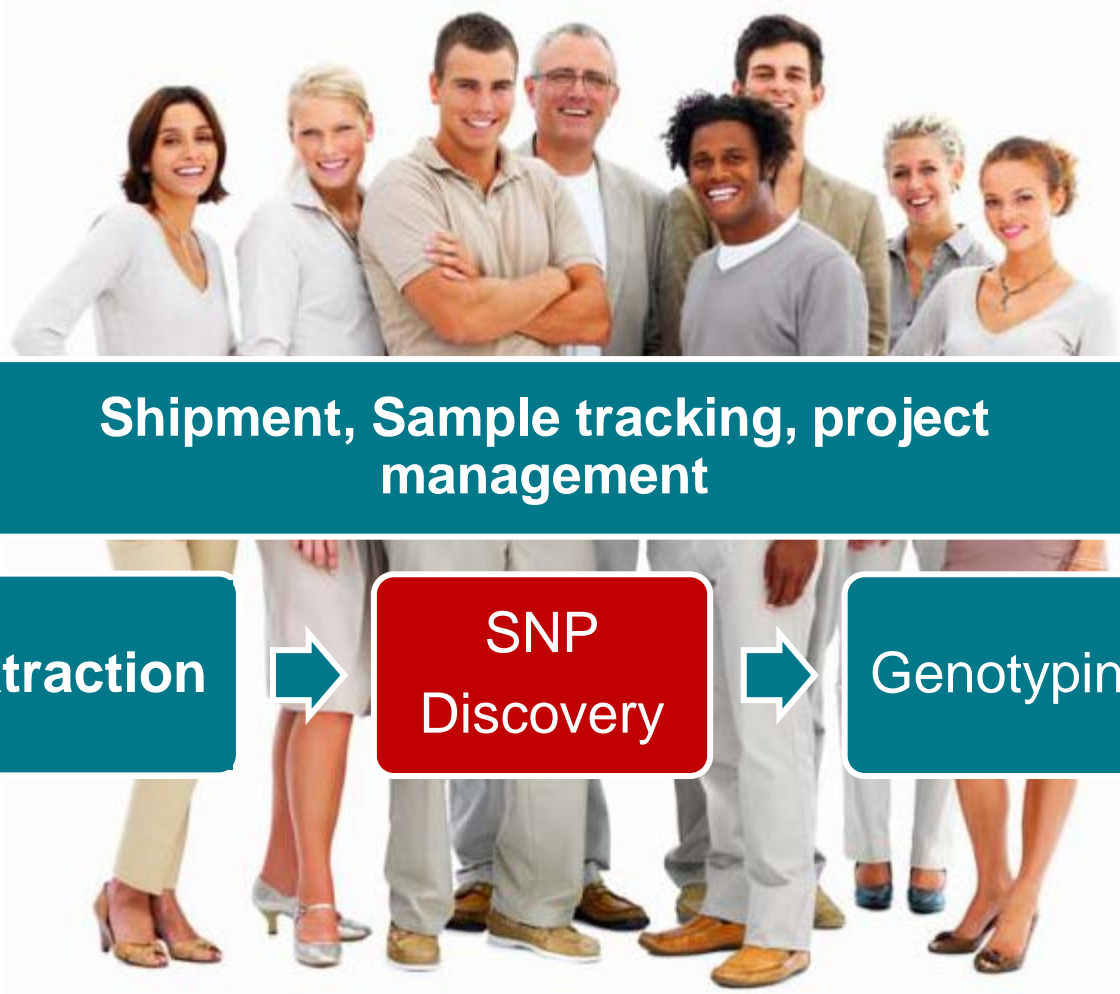


- Cost savings can be used to increase sample number
- Real customer example:

|               | In-house (core facility) | LGC Genomics |
|---------------|--------------------------|--------------|
| Sample number | 973                      | 4206         |



# Cohort study project support services



Shipment, Sample tracking, project management

Extraction

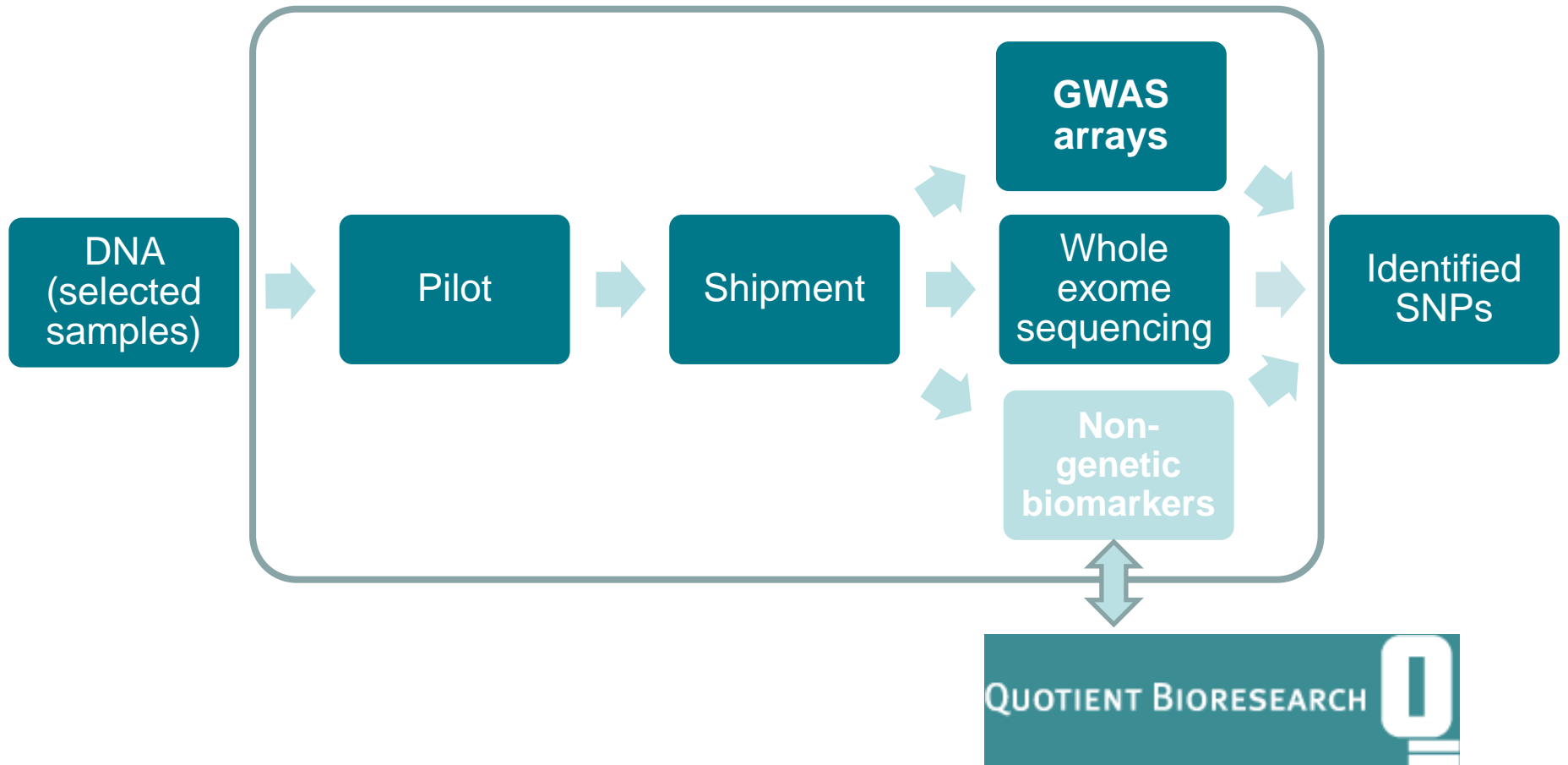


SNP  
Discovery



Genotyping

# SNP discovery module

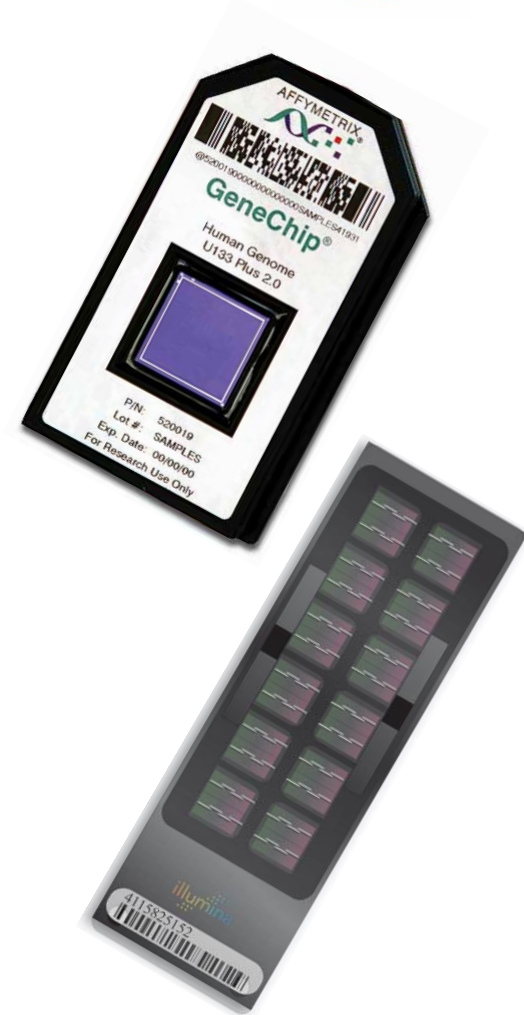


# SNP Discovery – Array technology



## Array based SNP discovery

- Affymetrix GeneChip® DNA analysis solutions
  - 10,000 to 1.8 million markers
  - starting material only 100-500 ng of DNA
- Illumina Infinium & Infinium HD bead arrays
  - 50,000 to 5 million SNP markers



Extraction



SNP  
Discovery



SNP  
Genotyping

# SNP discovery - NextGen sequencing



## Roche GS FLX TITANIUM & Illumina HiSeq 2000 technology



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- Whole exome enrichment
  - SureSelect Human All Exon v4+UTRs – 71Mb
  - Illumina TruSeq human all exome – 61MB
- Exome sequencing
  - HiSeq 2000



Extraction



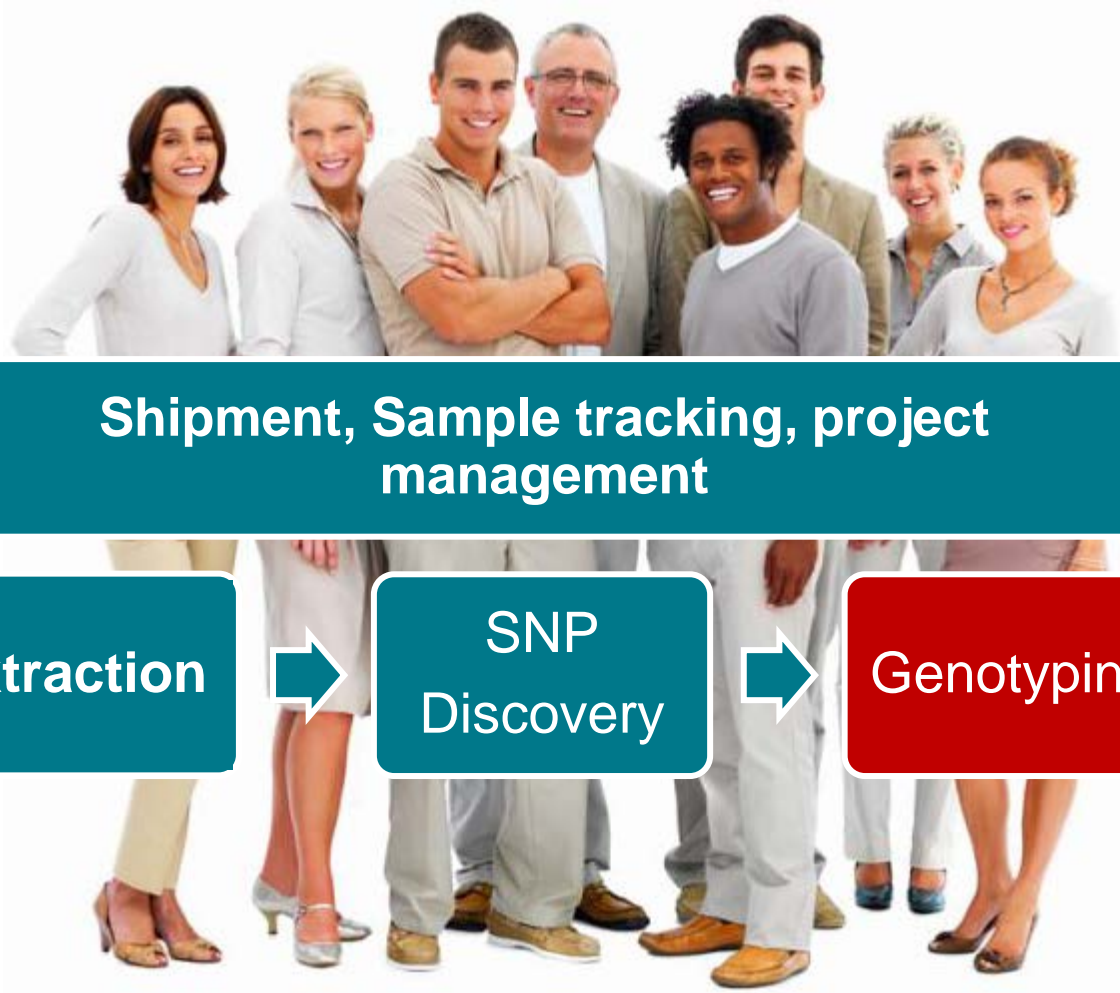
SNP  
Discovery



SNP  
Genotyping



# Cohort study project support services



Shipment, Sample tracking, project management

Extraction

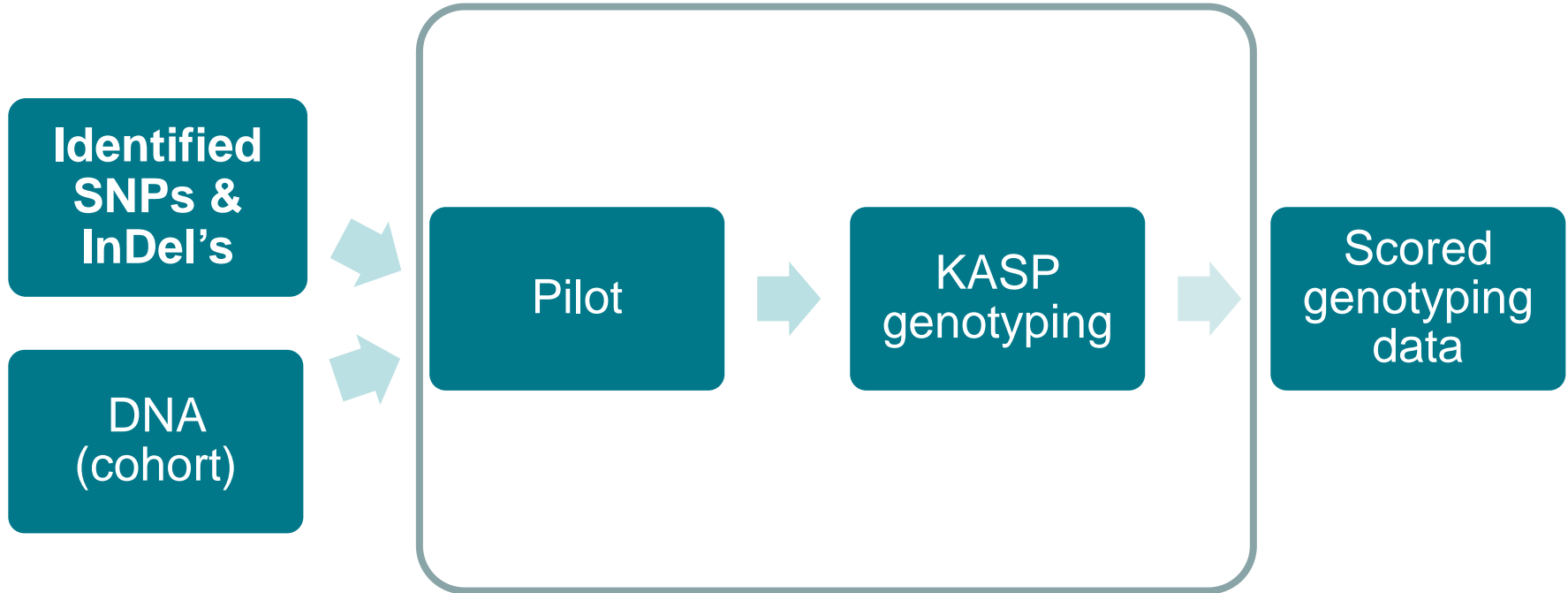


SNP  
Discovery



Genotyping

# Genotyping module



# KASP™ chemistry

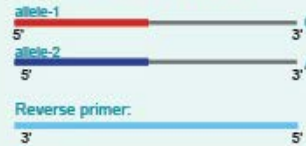


## 1) Assay components:

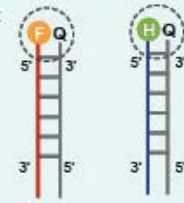
KASP uses three components: test DNA with the SNP of interest; KASP Assay mix containing two different, allele-specific, competing forward primers with unique tail sequences and one reverse primer; the KASP Master mix containing FRET cassette plus Taq polymerase in an optimised buffer solution.

### A) KASP Assay mix

Allele-specific forward primers:



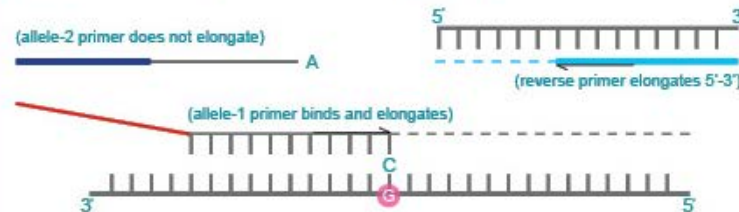
### B) KASP Master mix



### C) DNA template (sample)



## 2) Denatured template and annealing components – PCR round 1:



In the first round of PCR, one of the allele-specific primers matches the target SNP and, with the common reverse primer, amplifies the target region.

## 3) Complement of allele-specific tail sequence generated – PCR round 2:

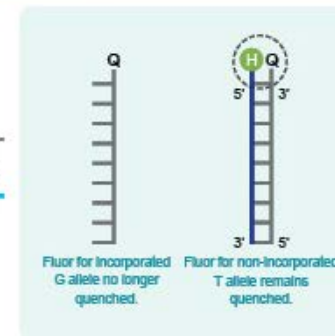


(Reverse primer binds, elongates and makes a complementary copy of the allele-1 tail.)

## 4) Signal generation – PCR round 3:



In further rounds of PCR, levels of allele-specific tail increase. The fluor labelled part of the FRET cassette is complementary to new tail sequences and binds, releasing the fluor from the quencher to generate a fluorescent signal.



| Legend                                   |   |
|--|---|
| <span style="color: red;">●</span>       | Allele-1 tail FAM-labelled oligo sequence |
| <span style="color: blue;">●</span>      | Allele-2 tail HEX-labelled oligo sequence |
| <span style="color: lightblue;">●</span> | Common reverse primer                     |
| <span style="color: orange;">F</span>    | FAM dye                                   |
| <span style="color: green;">H</span>     | HEX dye                                   |
| <span style="color: pink;">●</span>      | Target SNP                                |
| <span style="color: black;">Q</span>     | Quencher                                  |

# KASP genotyping in action



- We deliver over 1.5 Million PCR reactions / genotyping data point per day



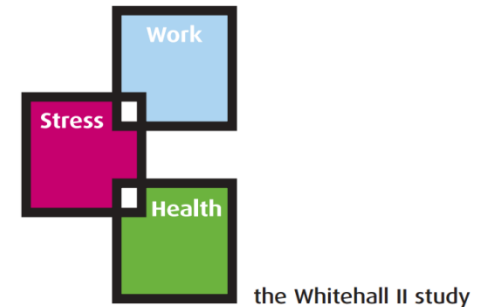
# Reference project

- **Name:**  
GEFOS (<http://www.gefos.org/>)
- **Location:**  
USA, Denmark, France, Germany, Greece, Italy, The Netherlands, Norway, Spain, Sweden and the United Kingdom
- **LGC Genomics solutions provided (completed):**  
85.000 DNA samples, 83 SNP's, 24 group world wide consortium
- **Reference:**  
Erasmus MC, Rotterdam



# Reference project

- **Name:**  
Whitehall II (<http://www.ucl.ac.uk/whitehallII>)
- **Location:**  
United Kingdom
- **LGC Genomics solutions provided (on-going):**  
NAP and SNP genotyping on 10.000 samples (several projects incl. DNA storage, sample tracking and sample handling)
- **Reference:**  
Prof. Dr. Aroon Hingorani (UCL):  
*“For cohort genotyping LGC Genomics is unbeatable”*



# Reference project

- **Name:**  
EPIGEN (<http://www.epigenchlamydia.eu>)
- **Location:**  
Denmark, The Netherlands, Spain and the United Kingdom
- **LGC Genomics solutions provided (on-going):**  
NAP & WGA 5.000 serum + SNP Discovery (450 samples, Aros AB)  
+ SNP Genotyping 10.000 samples
- **Reference:**  
Prof. Dr. Servaas Morre: *“Facilitating a complete solution was decisive in outsourcing to LGC” [vs. BGI]*





# How can we help you?

- Tell us about your research goals and needs
- Get a project specific price quote
- Start a free pilot

**[www.lgcgenomics.com](http://www.lgcgenomics.com)**