Pharmacogenomics and pharmacometabonomics

B.Sc. Pharmacology & Translational Medical Science, yr 2

Marc-Emmanuel Dumas, Ph.D.
Biomolecular Medicine, Dept Surgery and Cancer
Sir Alexander Fleming Building, room 360
South Kensington Campus
m.dumas@imperial.ac.uk

Learning objectives

- Pharmacogenetics and pharmacogenomics
- Methods available
  - DNA microarrays
  - Proteomics
  - Metabonomics
- Genotype/phenotype associations
- Pharmacometabonomics
- Personalized healthcare

Pharmacogenetics and pharmacogenomics

Definitions

- **PHARMACOGENETICS:** "The study of genetically determined inter-individual differences in therapeutic response to drugs and susceptibility to adverse effects"

⇒Restricted to one or few genes of interest

⇒Mendelian segregation

- **PHARMACOGENOMICS:** "Use of genome-based techniques in drug development"

⇒Not restricted to one or few genes

⇒Use of high-throughput technologies

Pharmacogenetics vs. Pharmacogenomics

- **PHARMACOGENETICS:** Study of variability in drug response determined by **single genes**.

- **PHARMACOGENOMICS:** Study of variability in drug response determined by **multiple genes** within the genome.

Pharmacogenetics and Pharmacogenomics

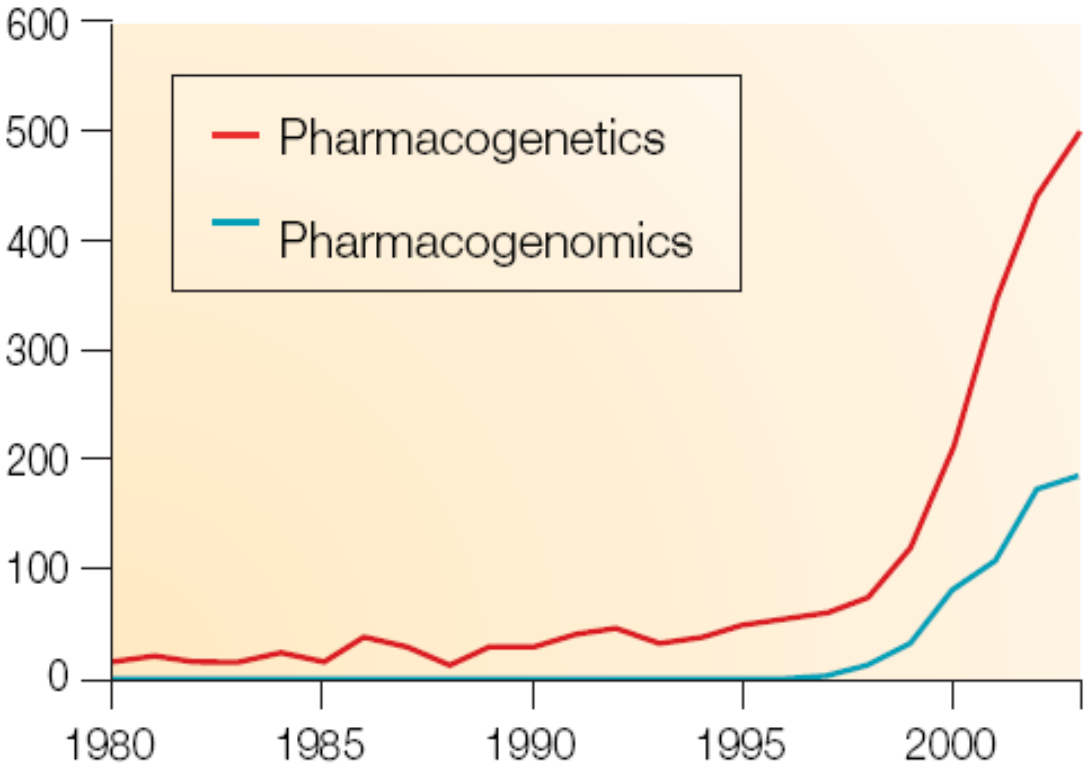


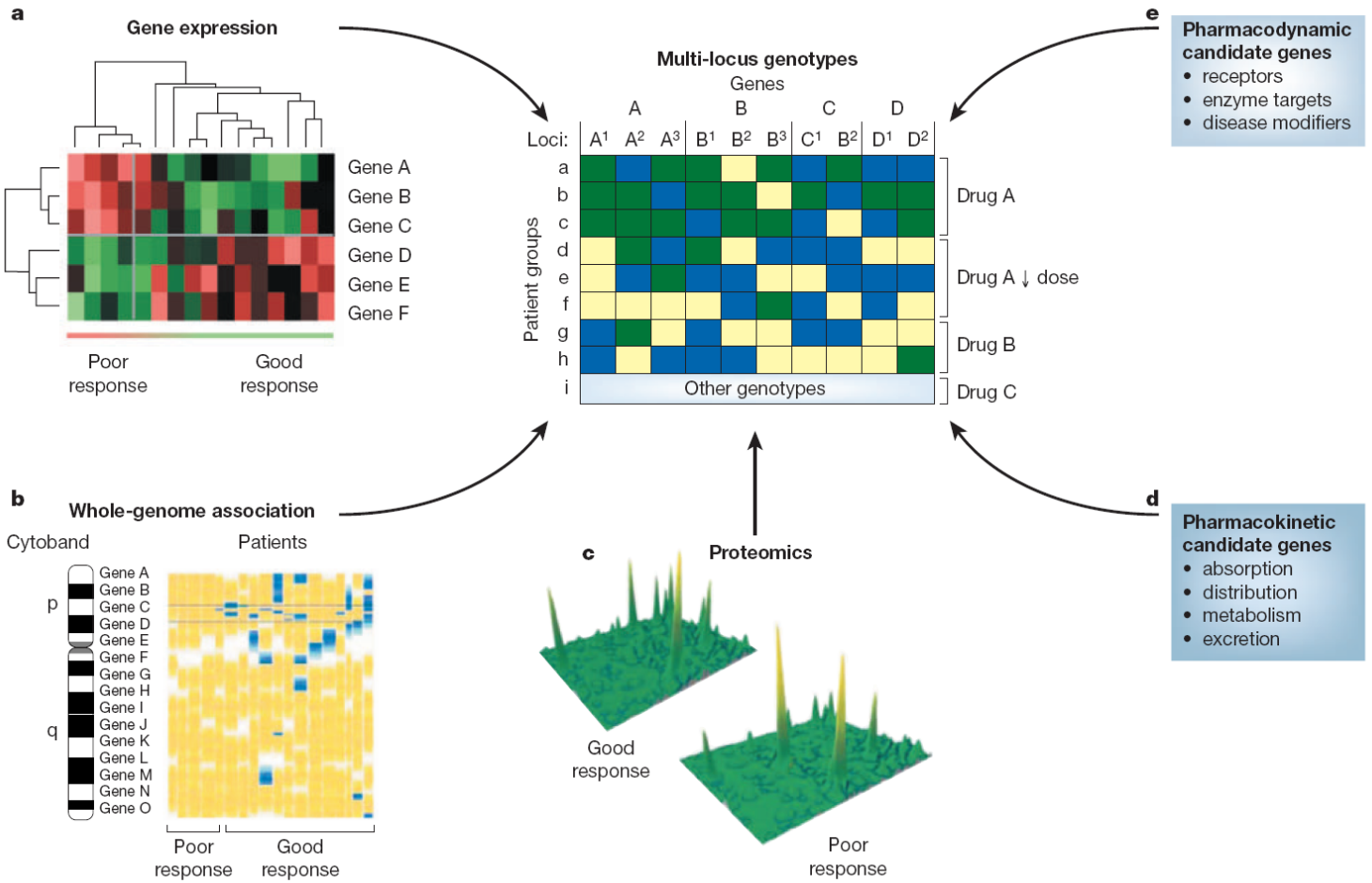
Figure 1 | **Appearance of the terms pharmacogenetics and pharmacogenomics in publications in PUBMED (National Library of Medicine).** Vogel first used the term pharmacogenetics (PGx) in 1959 (REF. 13). Publications on PGx have increased sharply in the last 5 years with the emergence of molecular genetics and genotyping technologies in clinical investigations. The term 'pharmacogenomics' first appeared in 1998.

Genomics

- **GENOME:** the entirety of an organism's hereditary information. includes both the genes and the non-coding sequences of the DNA (RNA in the case of retroviruses).
- **TRANSCRIPTOME:** set of all RNA molecules, including mRNA, rRNA, tRNA, and other non-coding RNA produced in one or a population of cells.
- **PROTEOME:** Entire set of proteins expressed by a genome, cell, tissue or organism.

- **METABOLOME:** Complete set of small-molecule metabolites (such as metabolic intermediates, hormones and other signalling molecules, and secondary metabolites) to be found within a biological sample, such as a single organism.

Pharmacogenomics



Evans and Relling Nature 2004

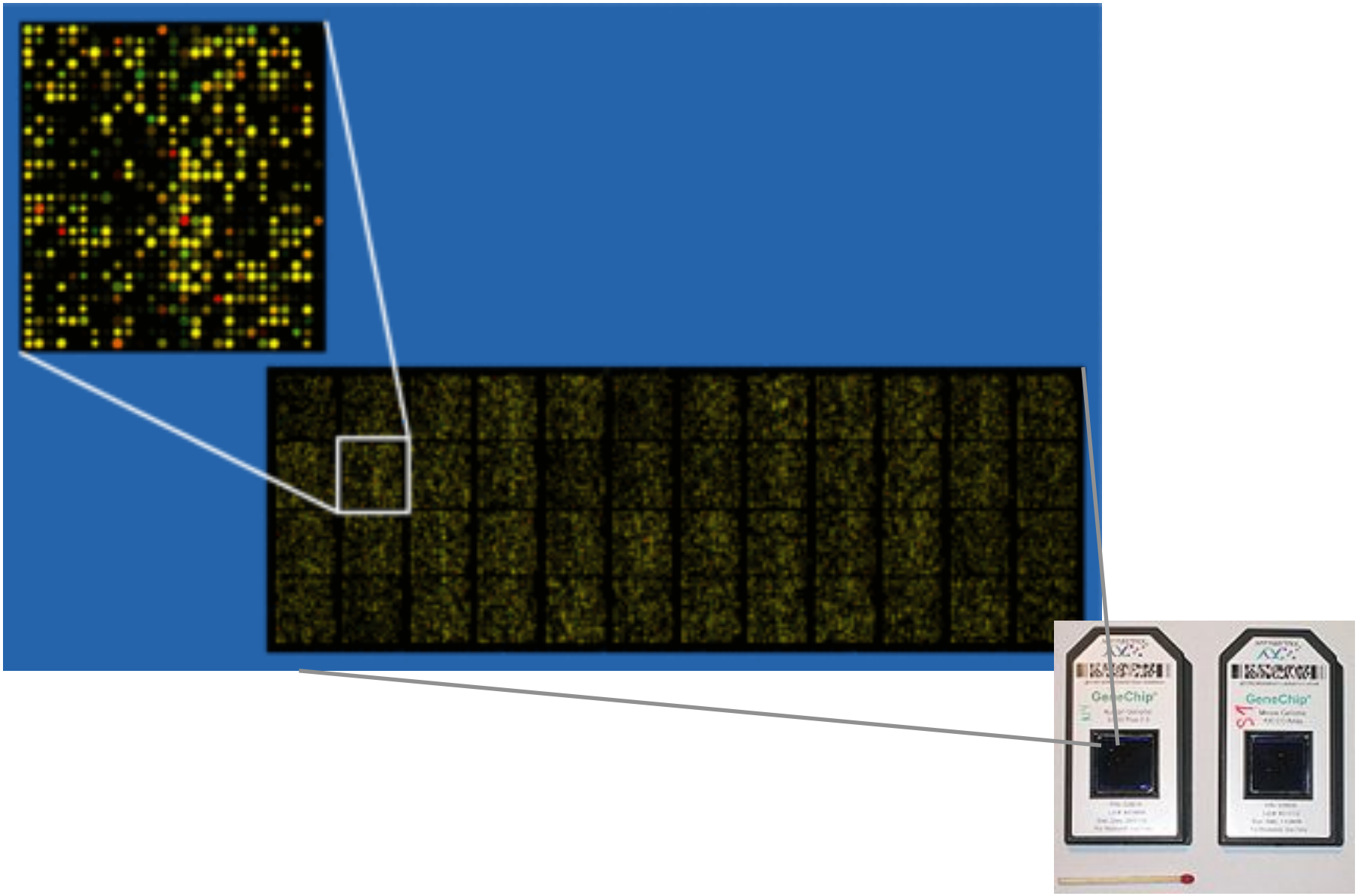
Methods

How can we study 30,000 genes?

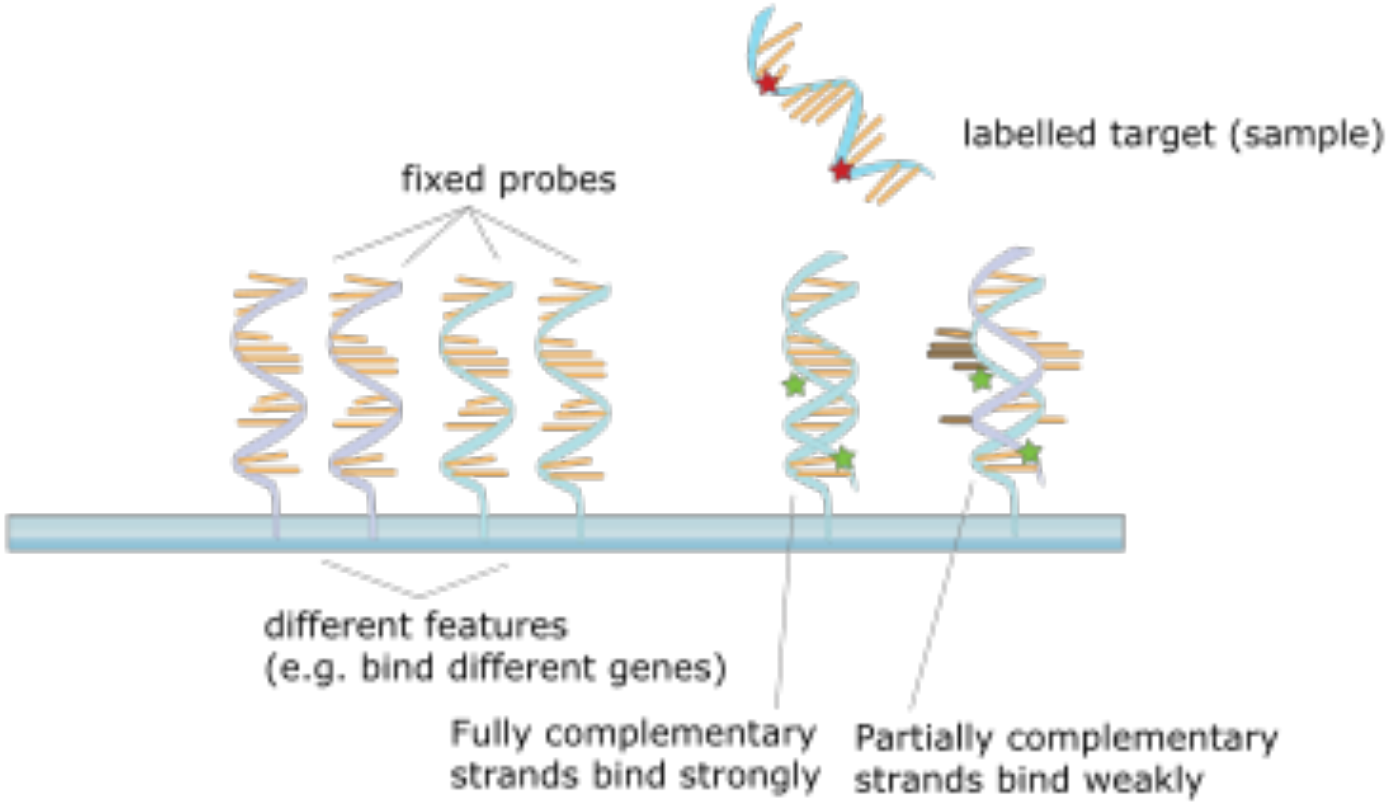
- **DNA microarrays**
- Multiplexed technology (measures made in parallel)
- Evolution of classical Southern blots
- Arrays of microscopic spots of DNA, attached to array surface
- Hybridization of labelled cDNA or DNA on surface when sequences match

- Possibility to detect SNP polymorphisms (genotyping), mRNA (transcriptomics), etc…

Microarray technology

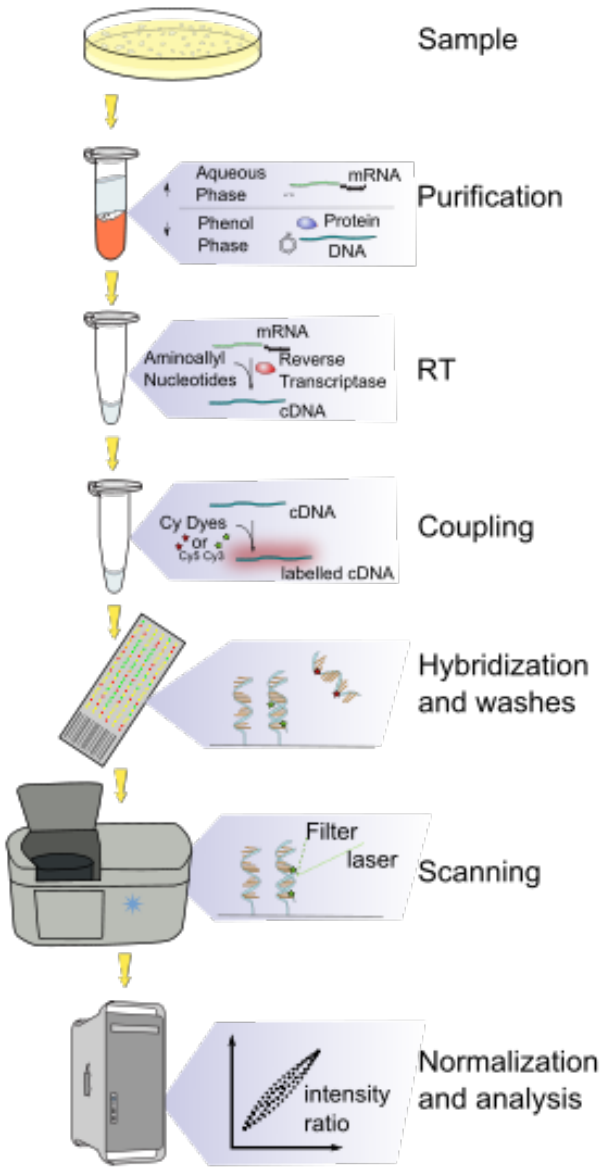


Principle of DNA arrays

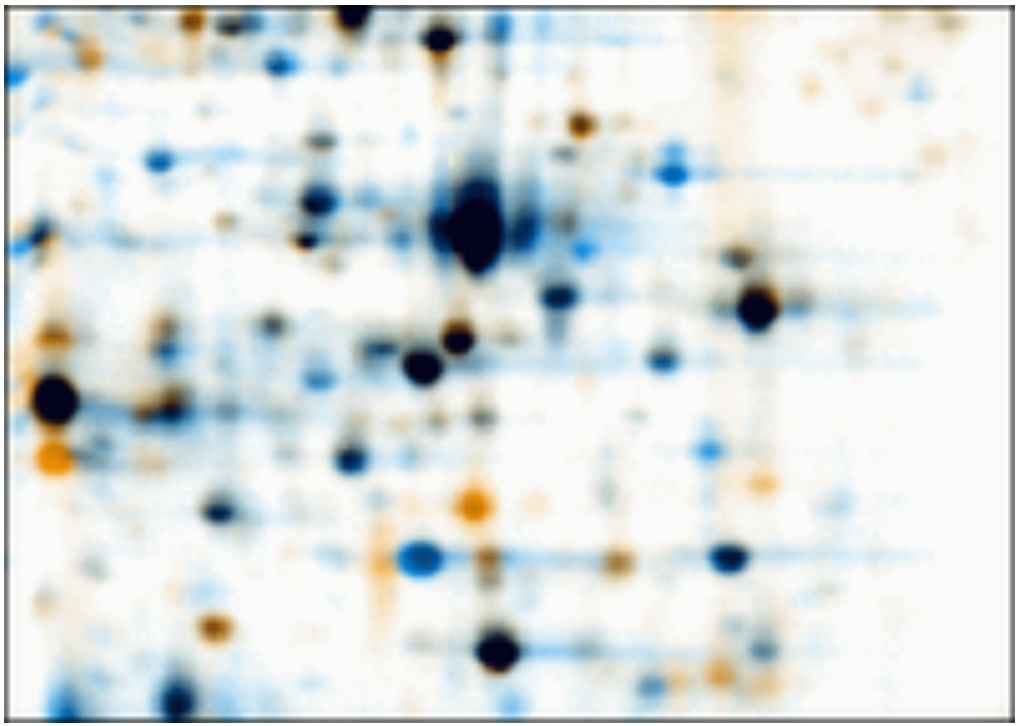


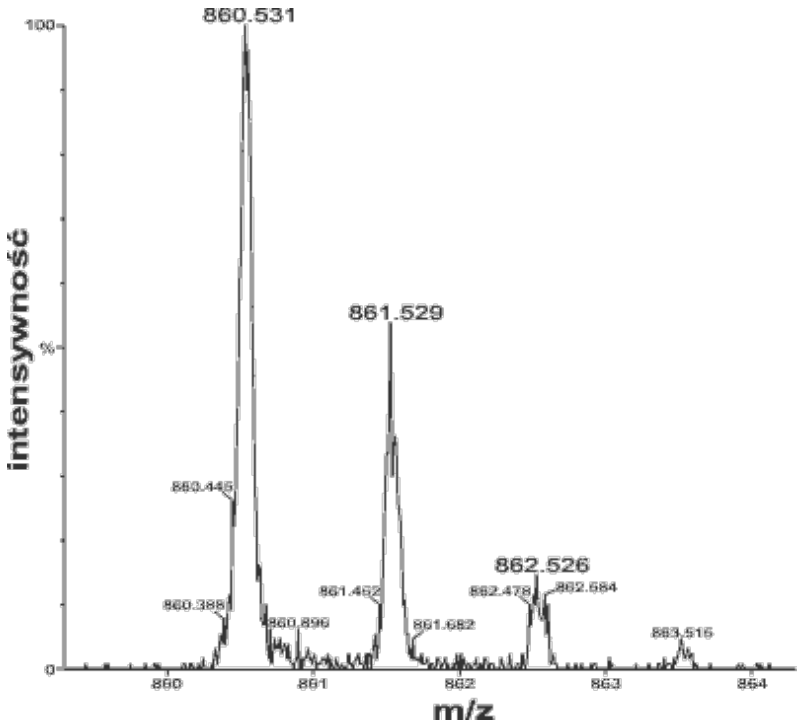
Microarray experiment

- Extraction of samples, purification prior to amplification
- Amplification usually made by polymerase chain reaction (PCR)
- Hybridization
- Detection of signal made by scanner and image analysis
- Statistical analysis of data



Proteomics

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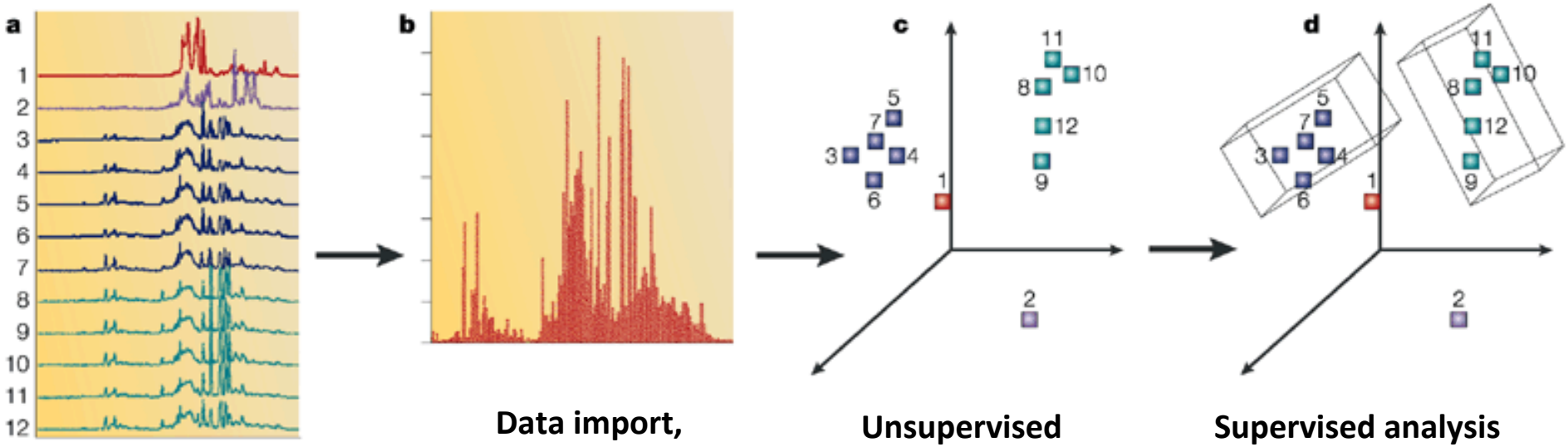
Metabolomics



Spectral acquisition

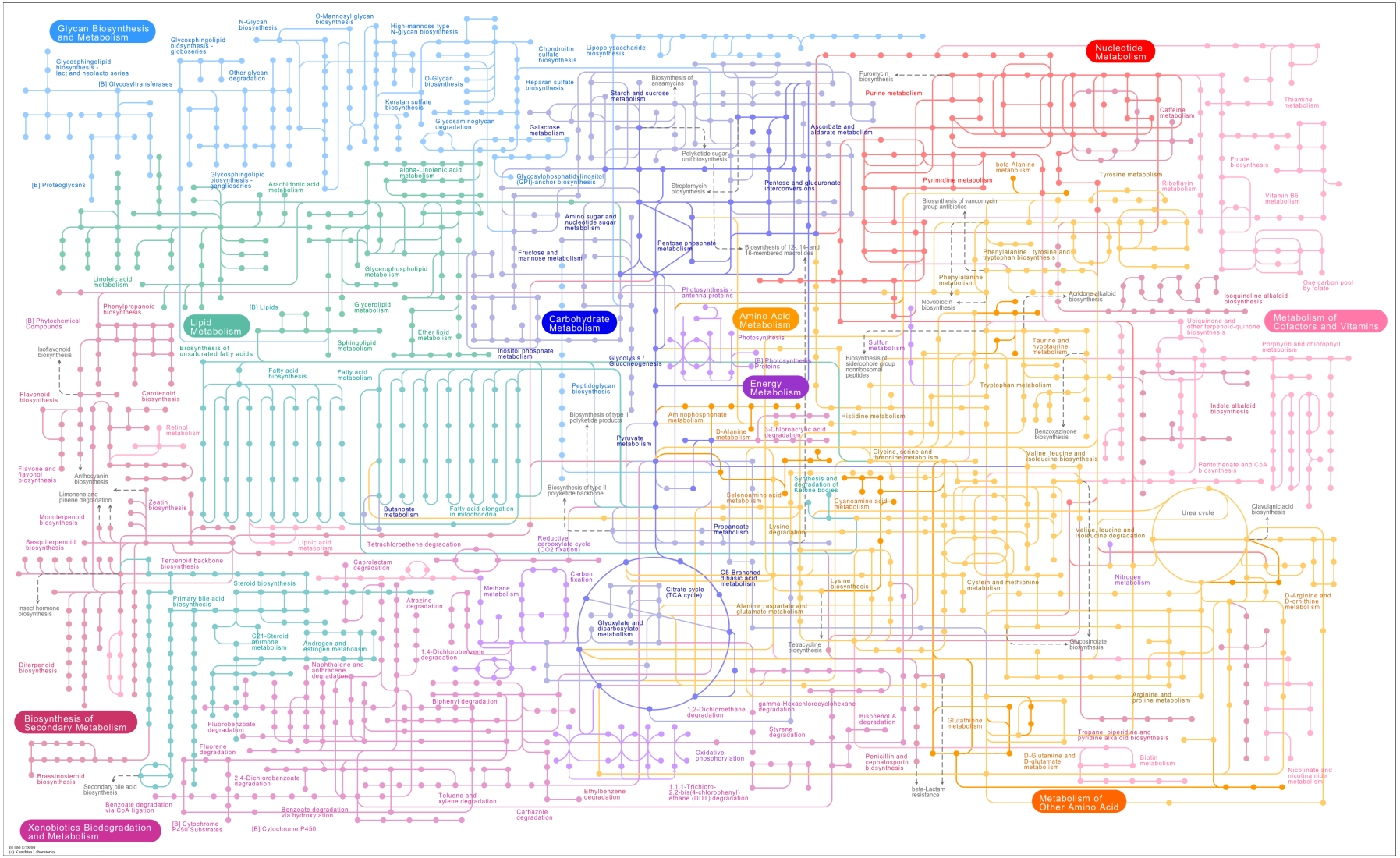
Data import, preprocessing and databasing

Unsupervised analysis

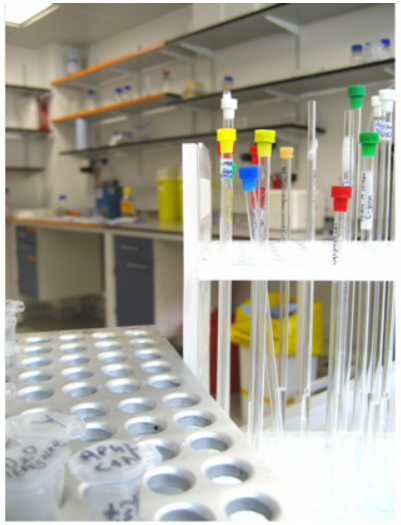
Supervised analysis confidence intervals

Nicholson JK, Connelly J, Lindon JC, Holmes E Nat Rev Drug Discov. 2002 Feb;1(2):153-61.

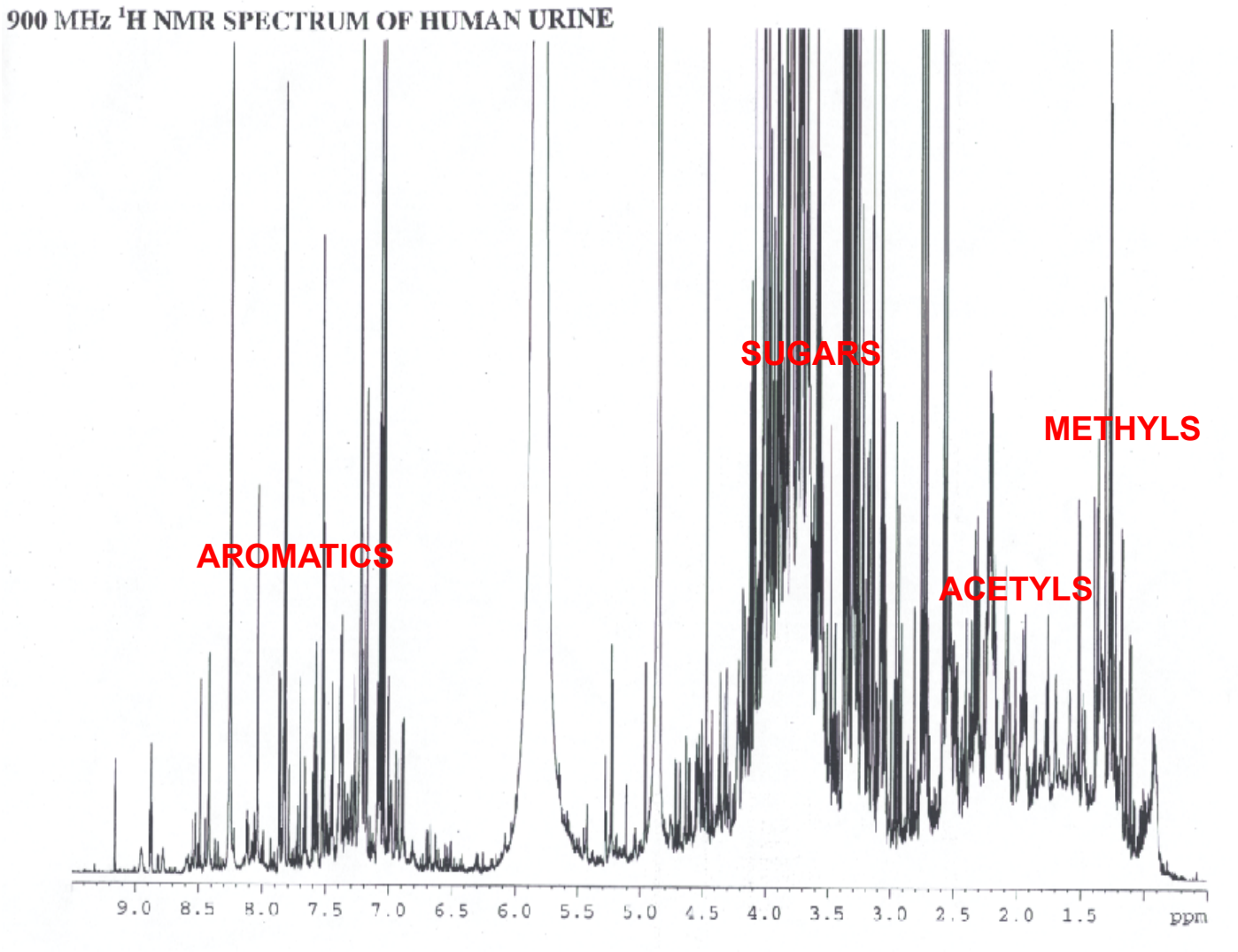
Metabolic pathways in one glance



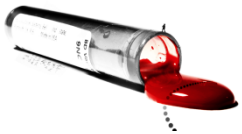
Metabolomic facility @ Imperial – NMR, GC-MS, UPLC-MS



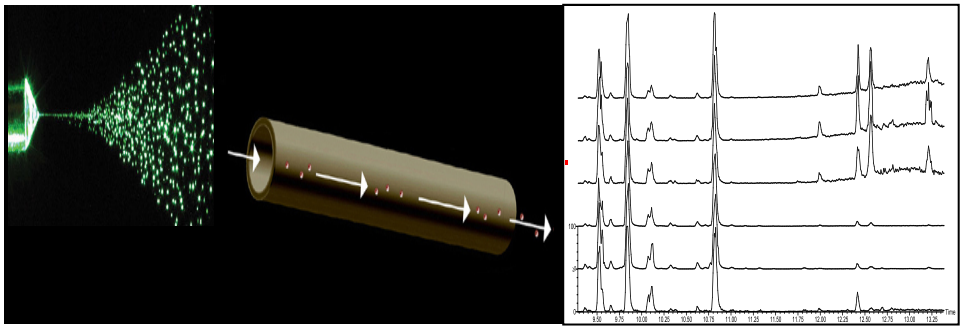
^{1}H NMR spectrum – a metabolic snapshot



Liquid chromatography -MS Analysis



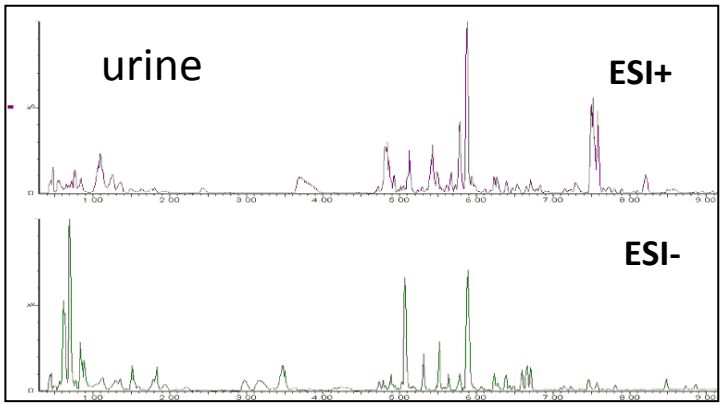
Sample preparation

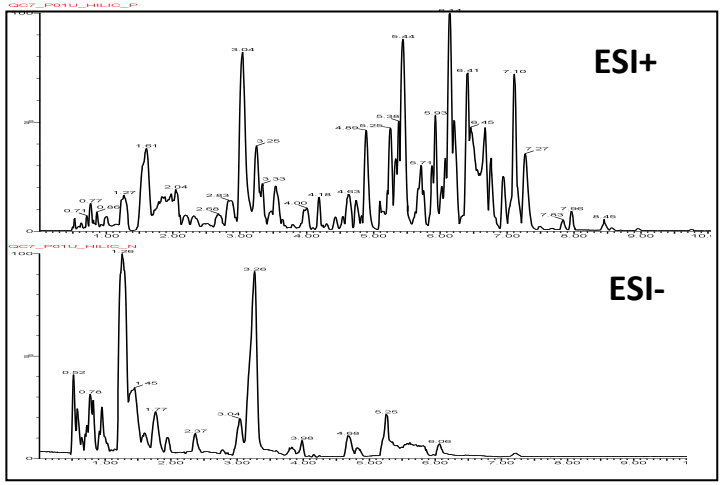


UPLC-MS Analysis

Untargeted Profiling using TOF or Q-TOF

Reverse phase chromatography

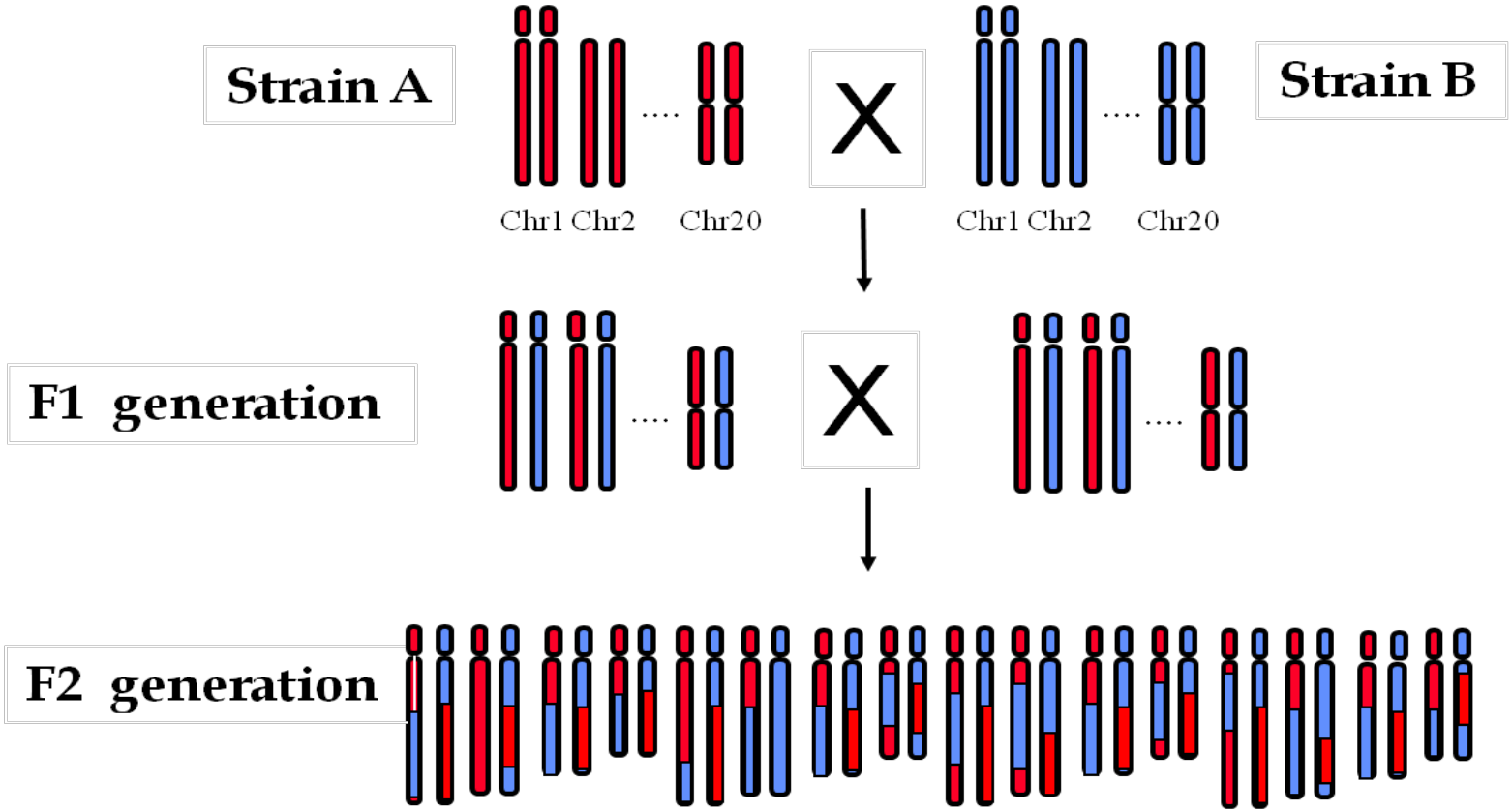




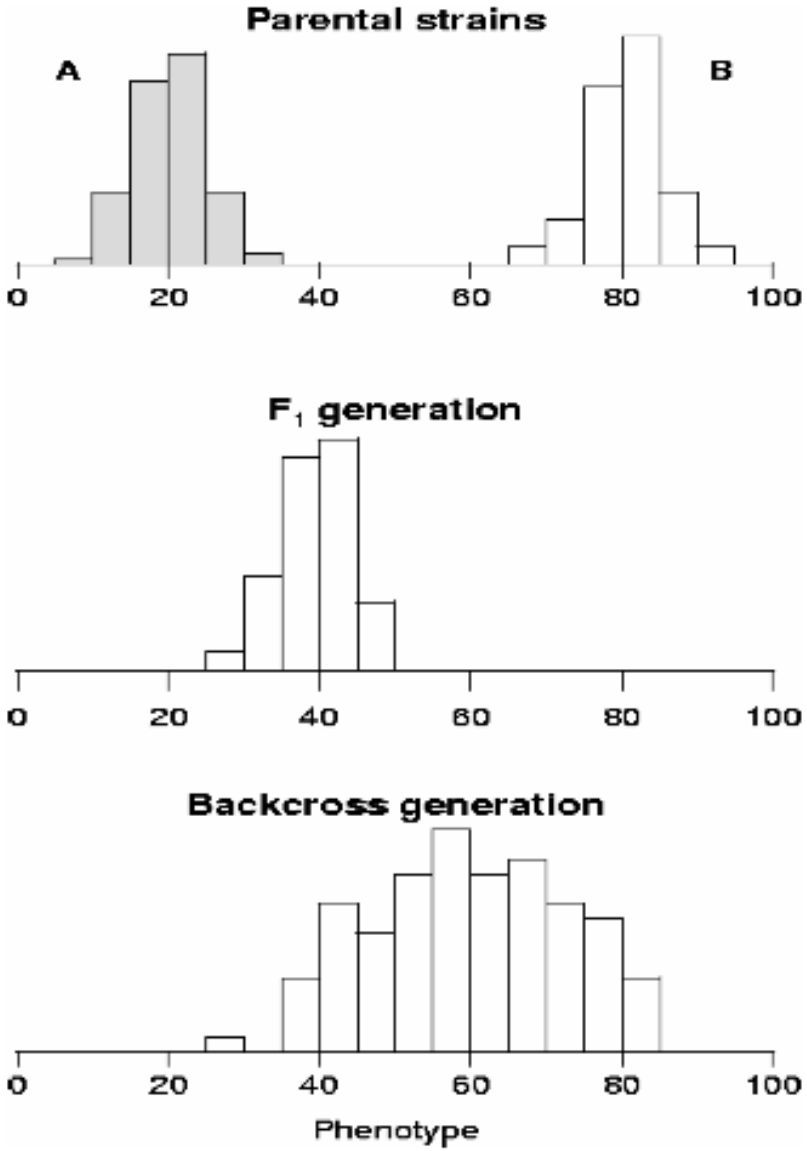
Hydrophilic interaction (HILIC)

Genotype/phenotype association

Experimental cross - genetic variation



Phenotype variation



Genome mapping of transcriptomic data

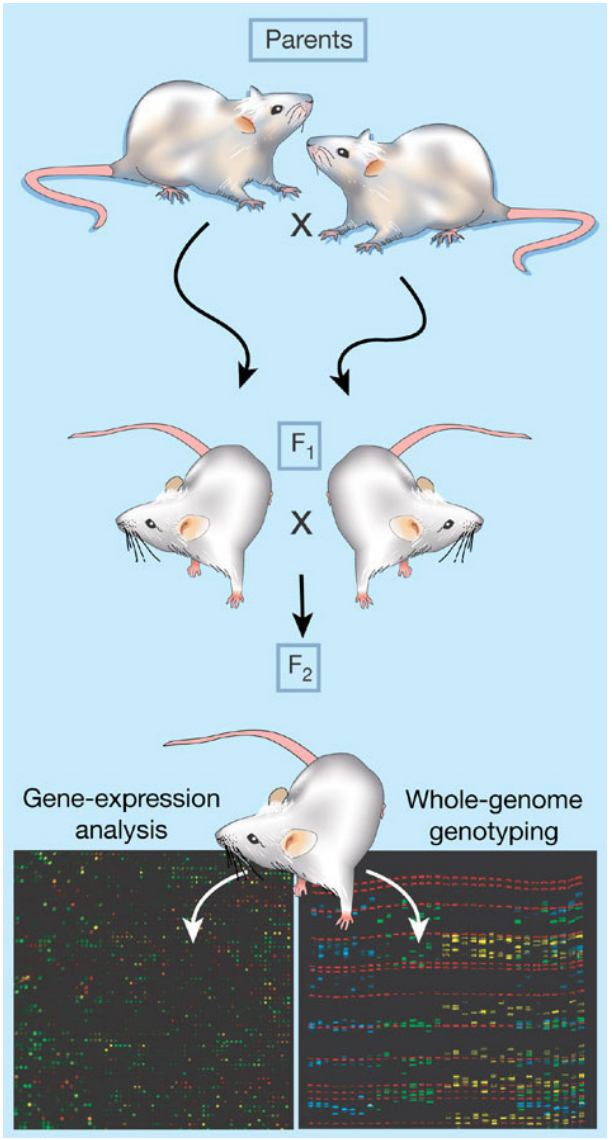
Genetical genomics, eQTL
based (amongst others) on the paper from
Schadt et al., Nature 2003, 422, 297-302.

- **F2 cross:**
start seeing segregation of alleles on chromosomes

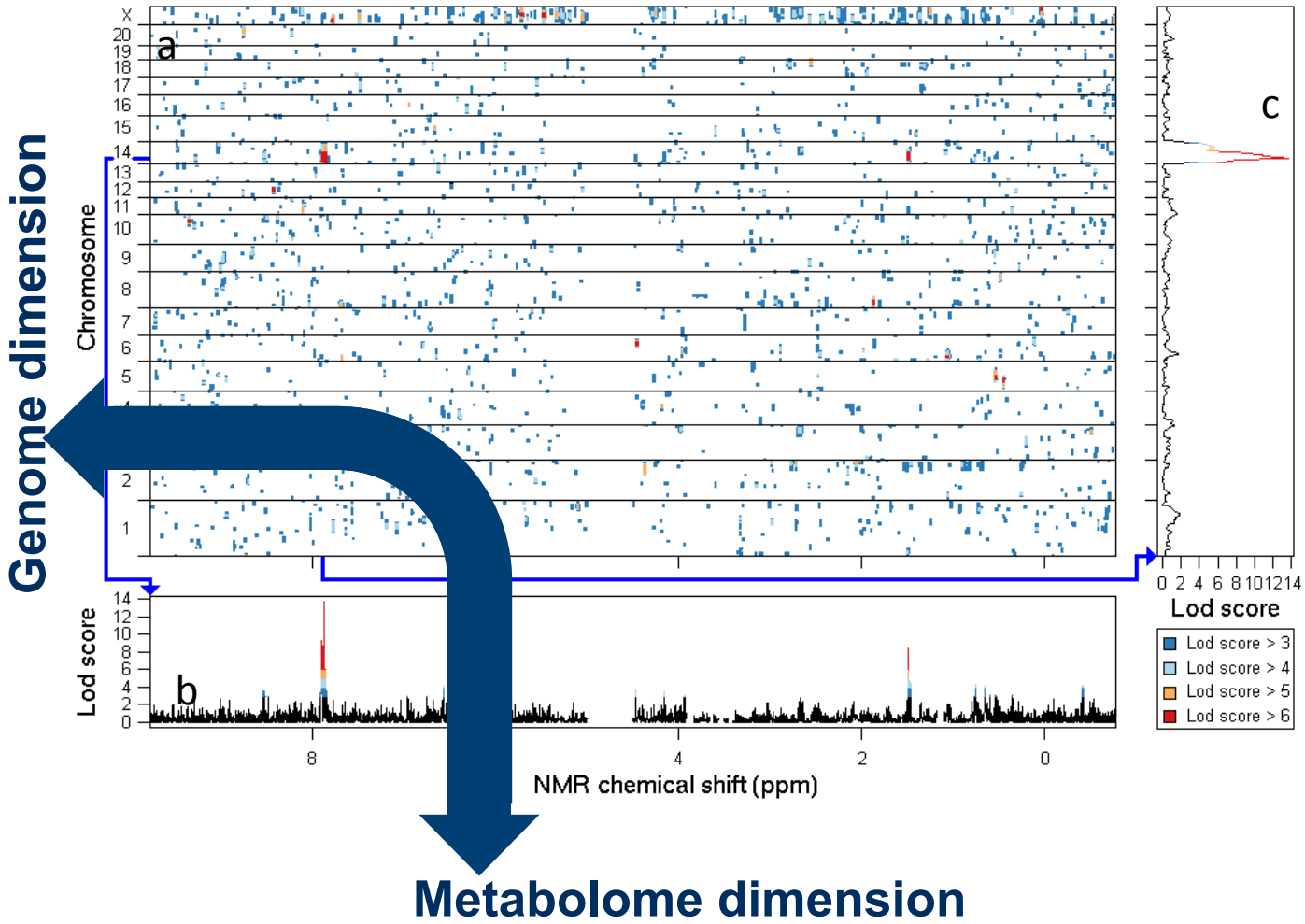
- **Genotyping:** SNP

- **Phenotyping:** transcriptomics

- **eQTL:** treating **expression** data like phenotypes to map **Quantitative Trait Loci**



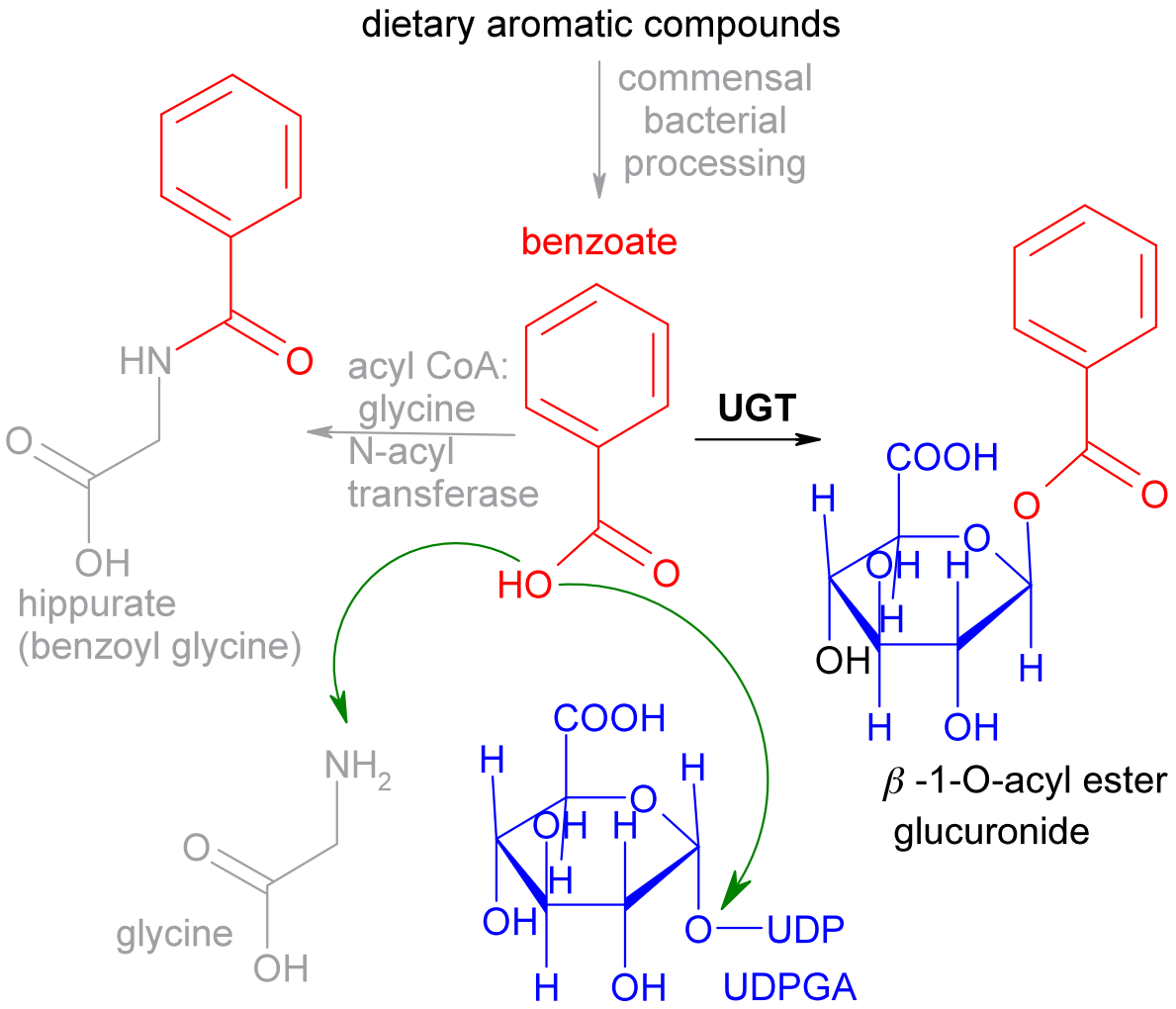
Metabotype Quantitative Trait Locus (mQTL)



- **GK x BN F2 cross:**
  BN = **normoglycemic**
  GK = **diabetic**
- **Genotyping:**
  microsatellites / SNP
- **Phenotyping:**
  plasma NMR
- **qtl mapping:**
  LOD score > 15

Dumas, Wilder et al., *Nature Genetics* 2007, 39, 666-672

Typical mQTL - metabolite association



Dumas, Wilder et al., Nature Genetics 2007, 39, 666-672

Pharmaco-metabonomics

Pharmacometabonomics

- **PHARMACOGENOMICS:** “Use of genome-based techniques in drug development”

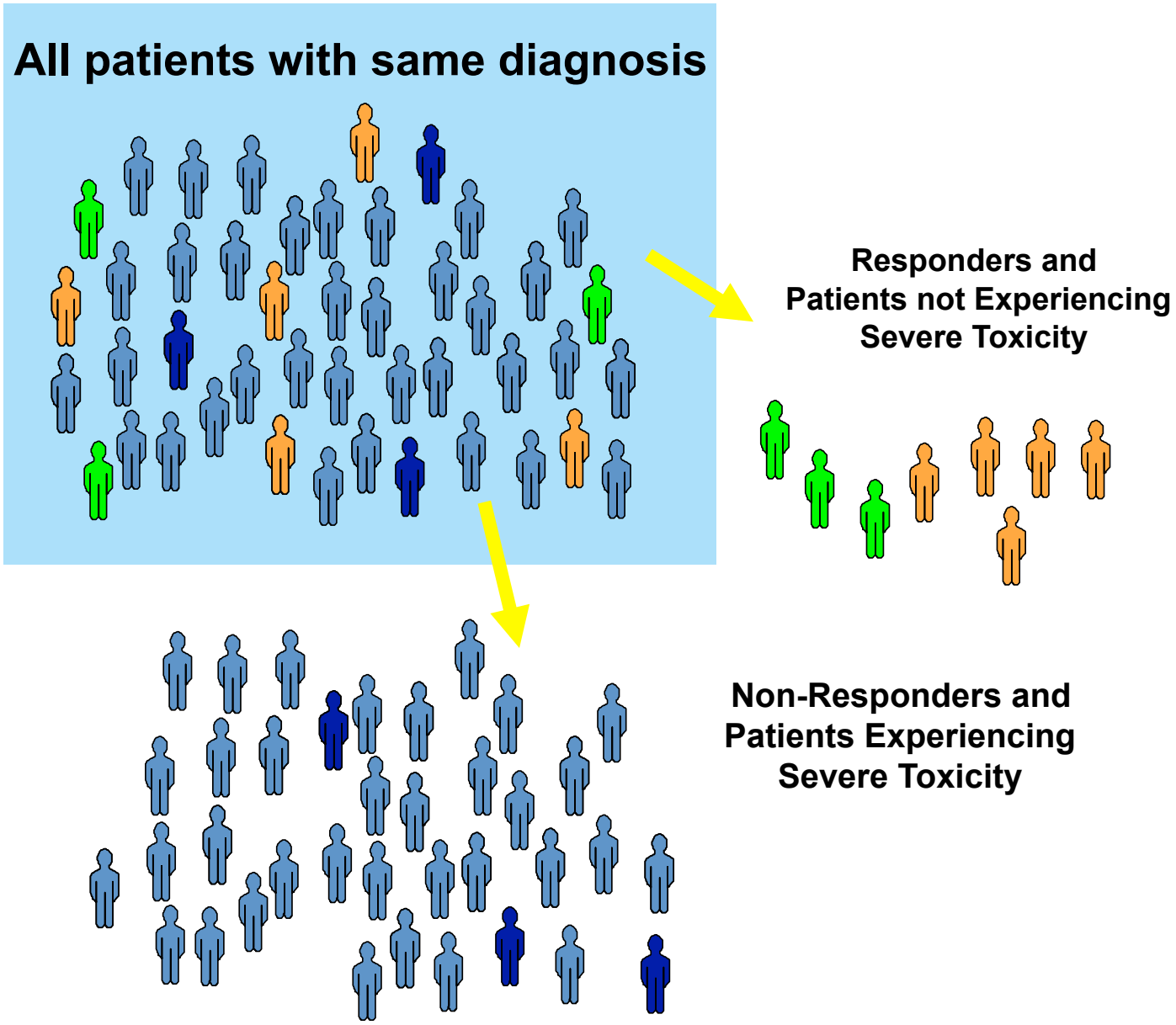
- **PHARMACOMETABONOMICS:** “Use of metabolome-based techniques in drug development”

⇒ Access to genetic and environmental variation in metabolism

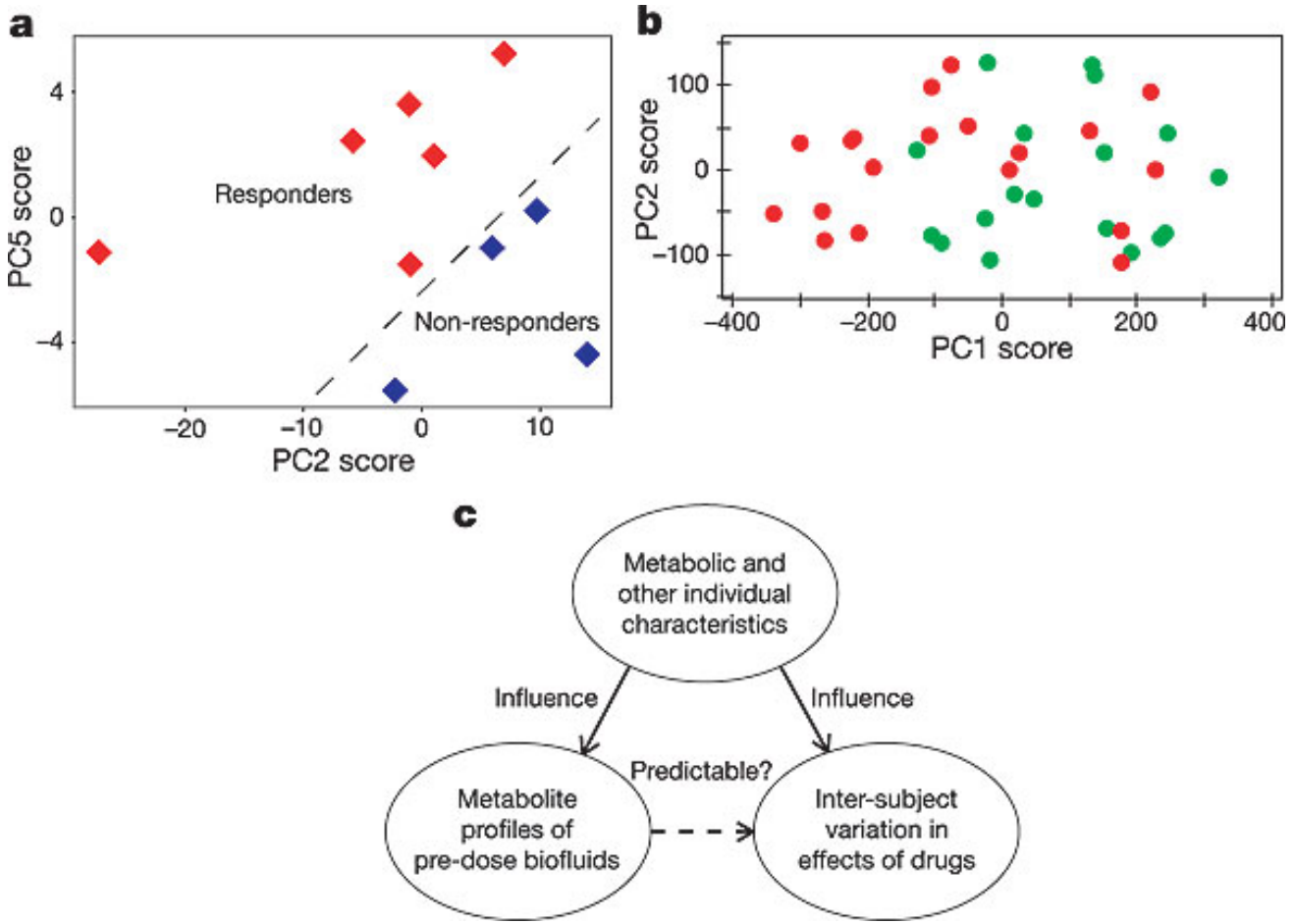
⇒Complementary to genetic studies

⇒Focus on identification of responders and non-responders and early response

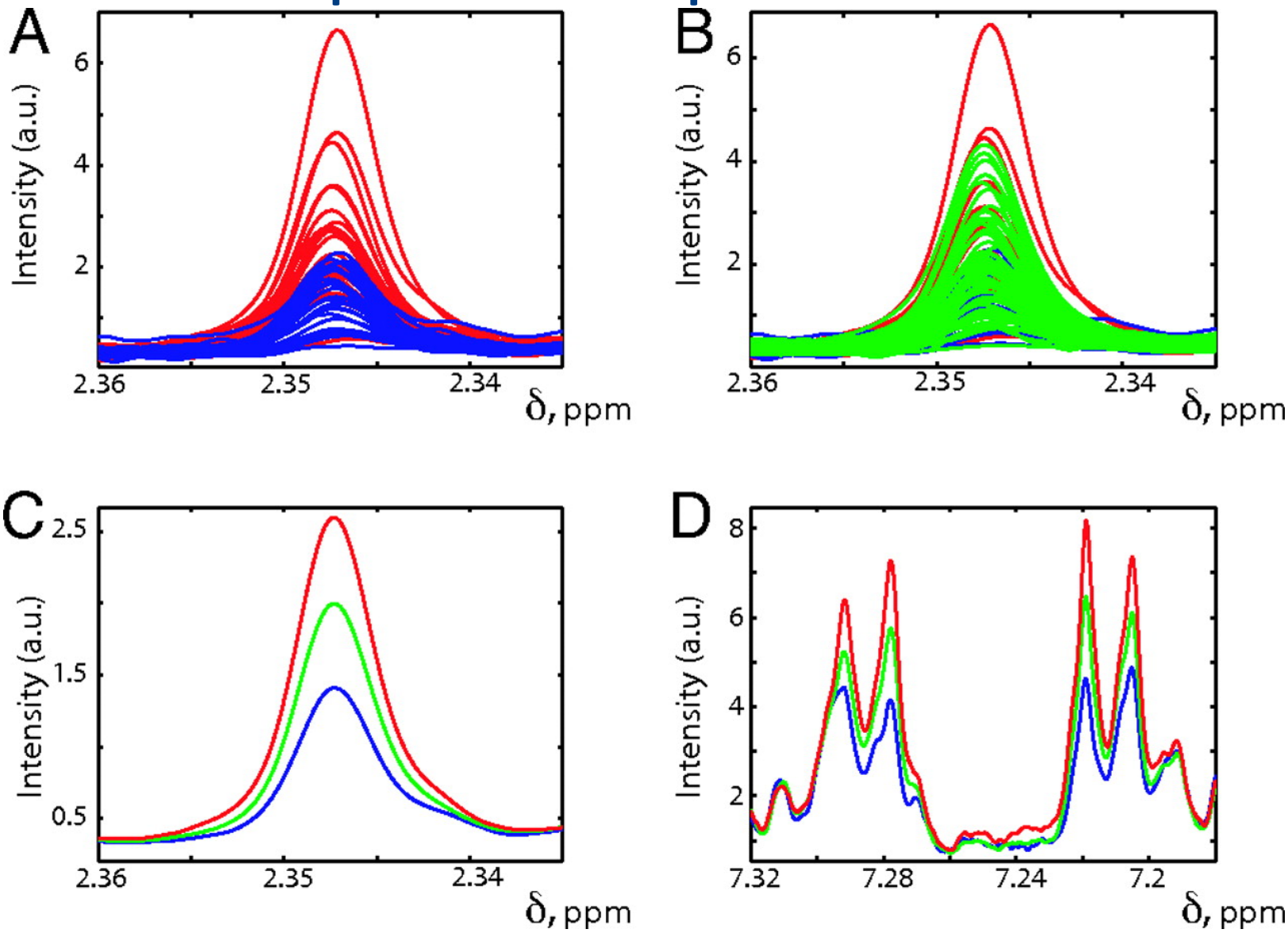
Pharmacometabonomics



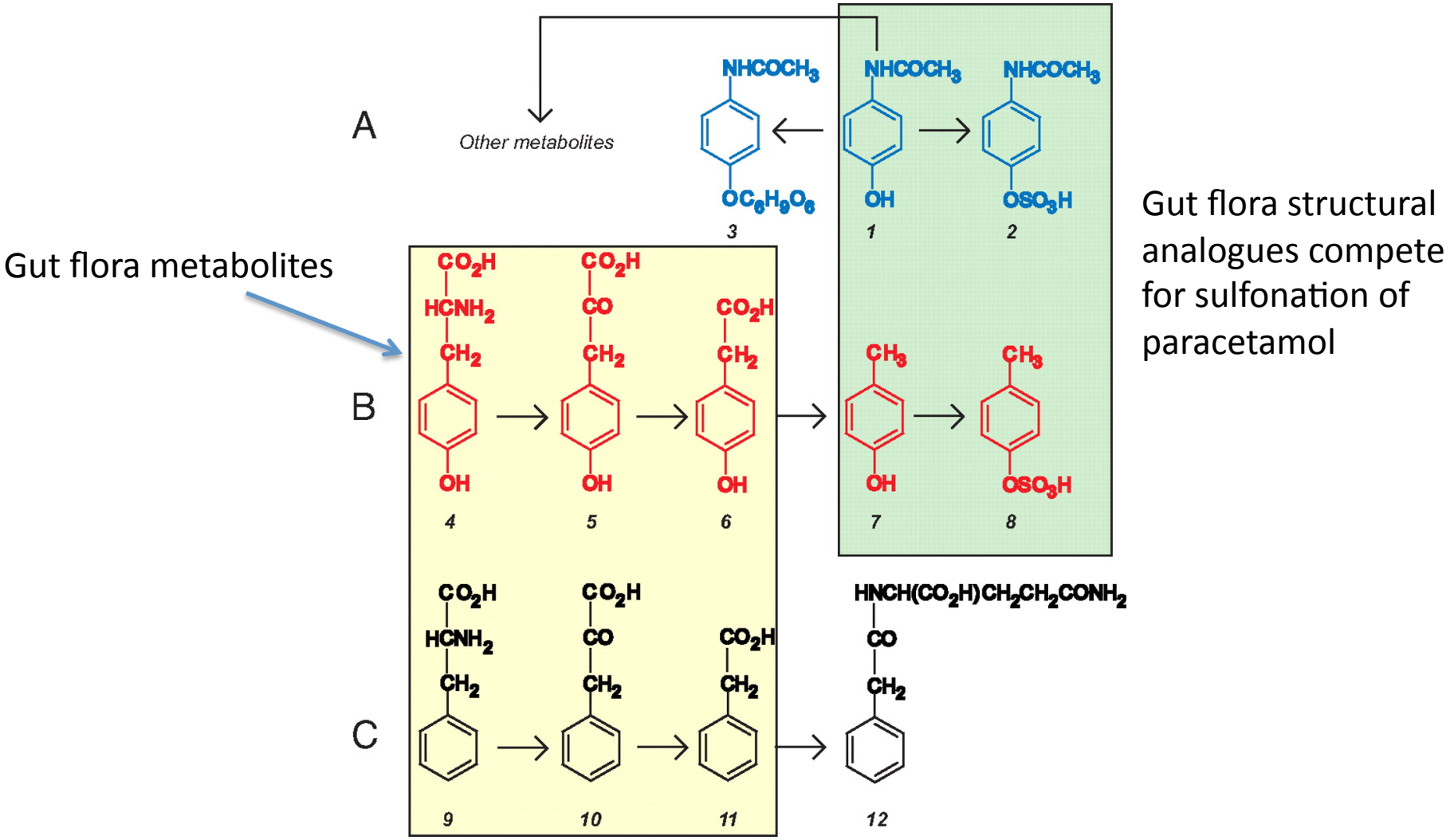
Pharmacometabonomics



Post dose behaviour according to pre-dose spectra



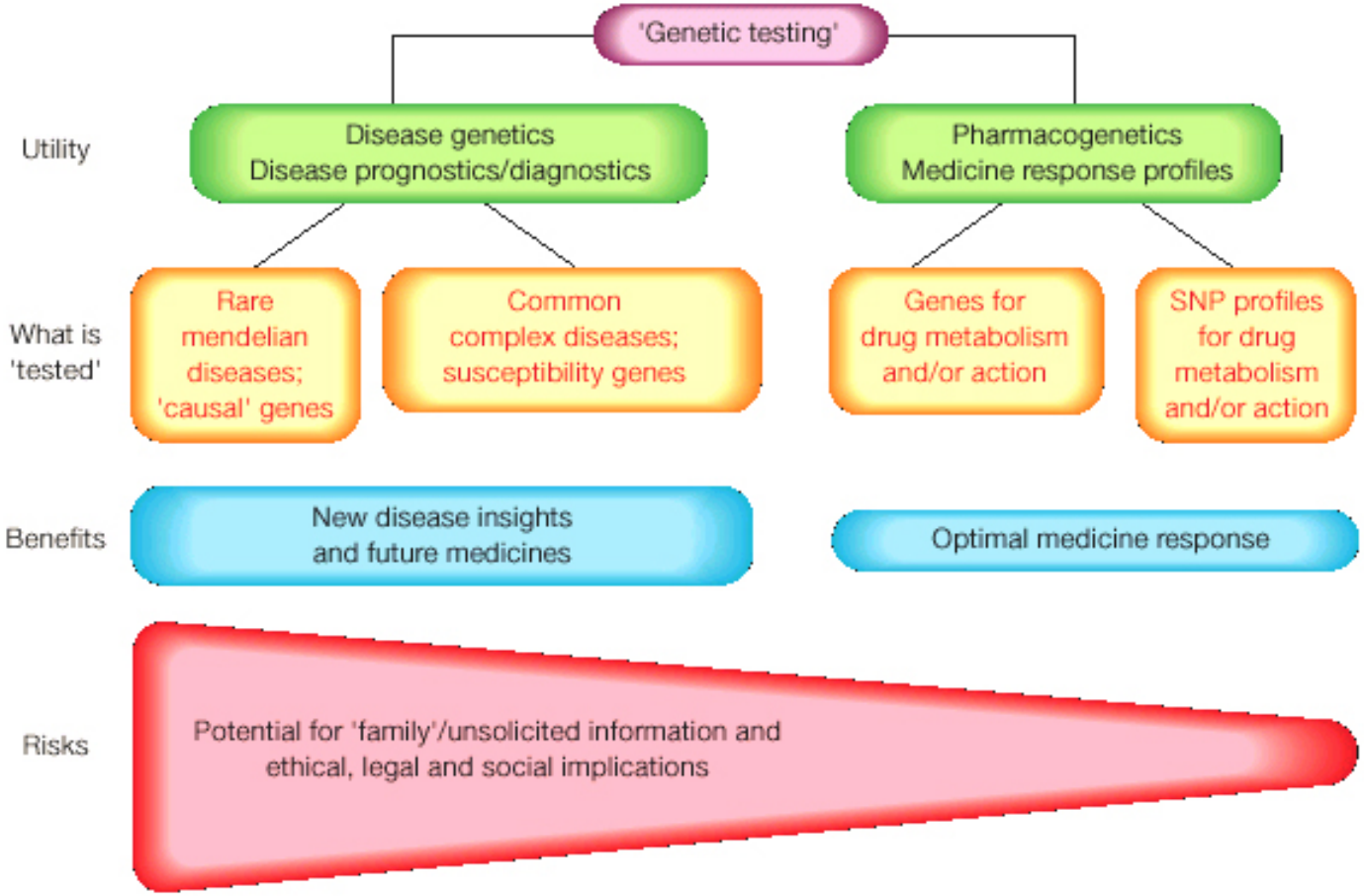
Indirect role of gut flora in paracetamol metabolism



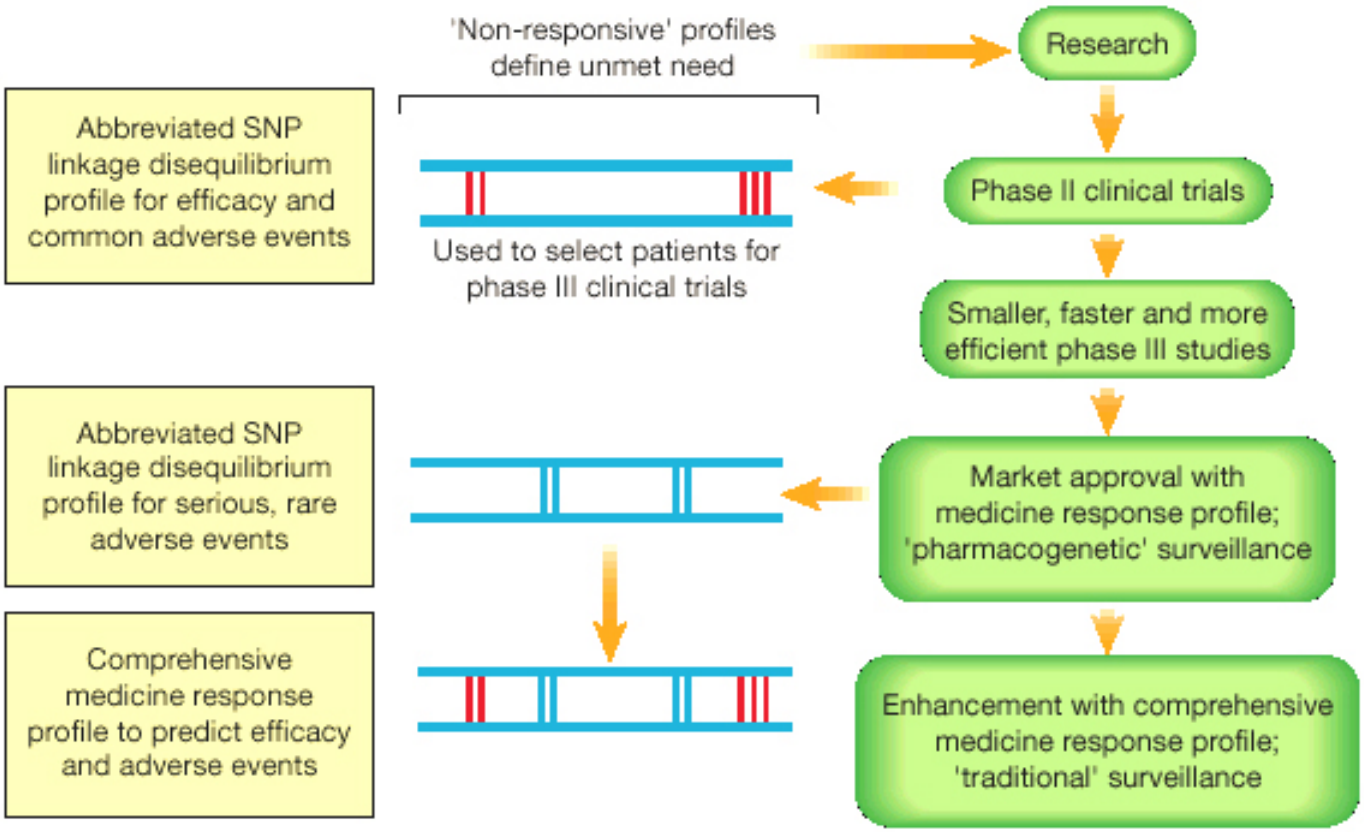
Gut flora structural analogues compete for sulfonation of paracetamol

Personalized healthcare

Risk assessment in pharmacogenomics

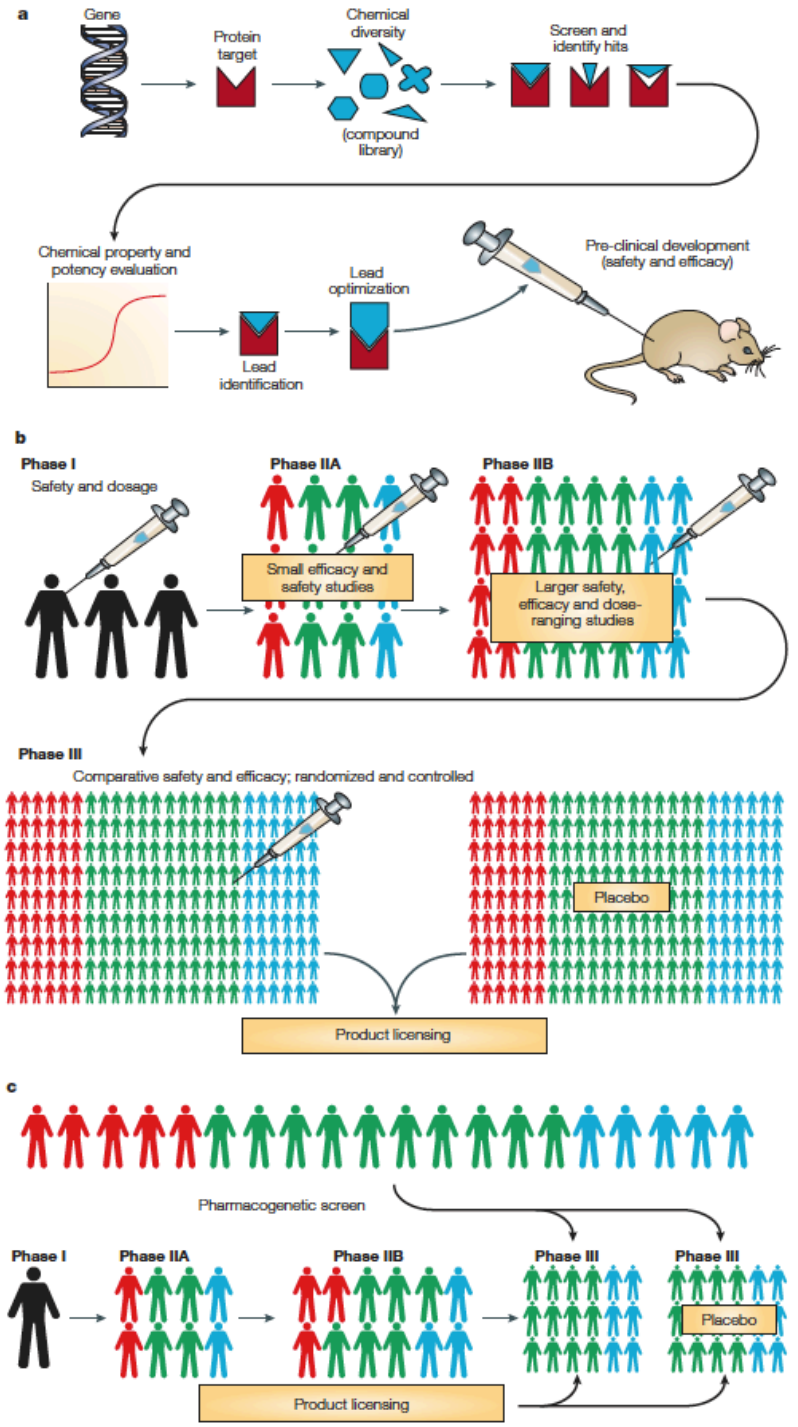


Pharmacogenomics in clinical trials



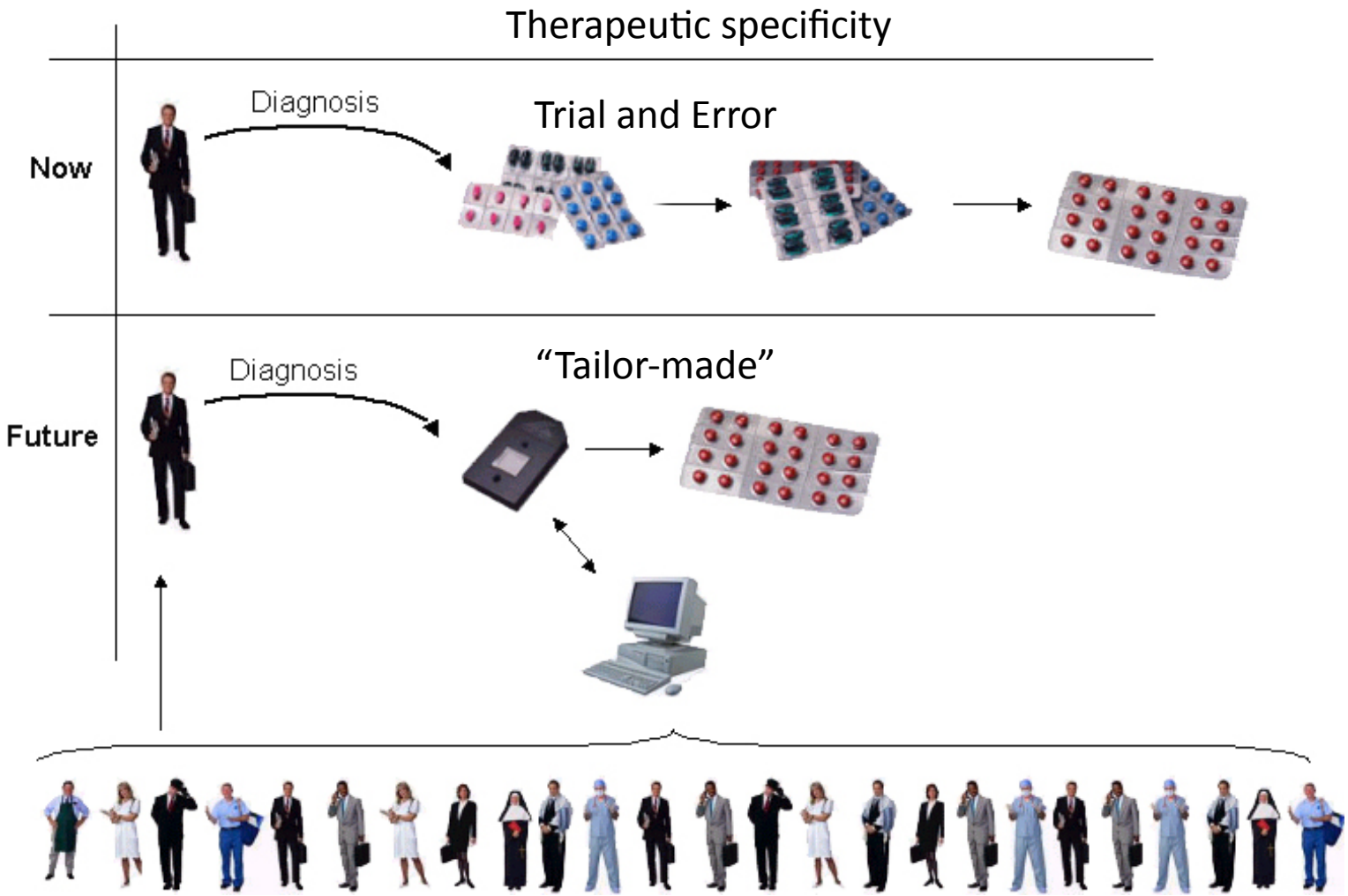
Pharmacogenomics in pharmaceutical pipeline

- Identification of target proteins
- Identification of ligands
- Assessment of drugability
- Tests in animals / cells
- Clinical trials
- Population studies



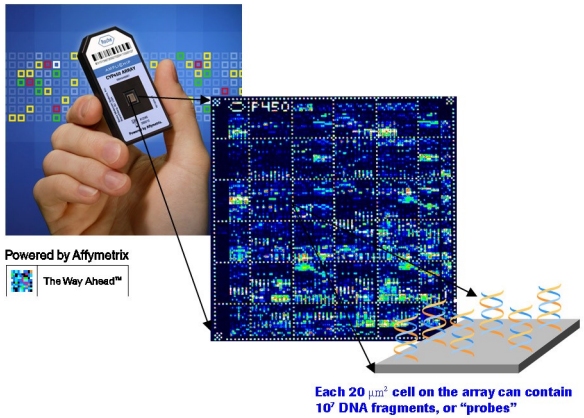
Pharmacogenomics prospect

Diagnosis - therapy associated with preciption of a drug now and in the future.



New Frontiers in Genetics

- First Phase of the 'HapMap'
  (Haplotype Map, which maps common patterns of human genetic variation - 'haplotypes')



- **First diagnostic microarray chip approved by FDA in 2005**

The Amplichip, made by Roche, analyzes variation in 2 genes involved in metabolism of 25% of prescription drugs (CYP450 genes)

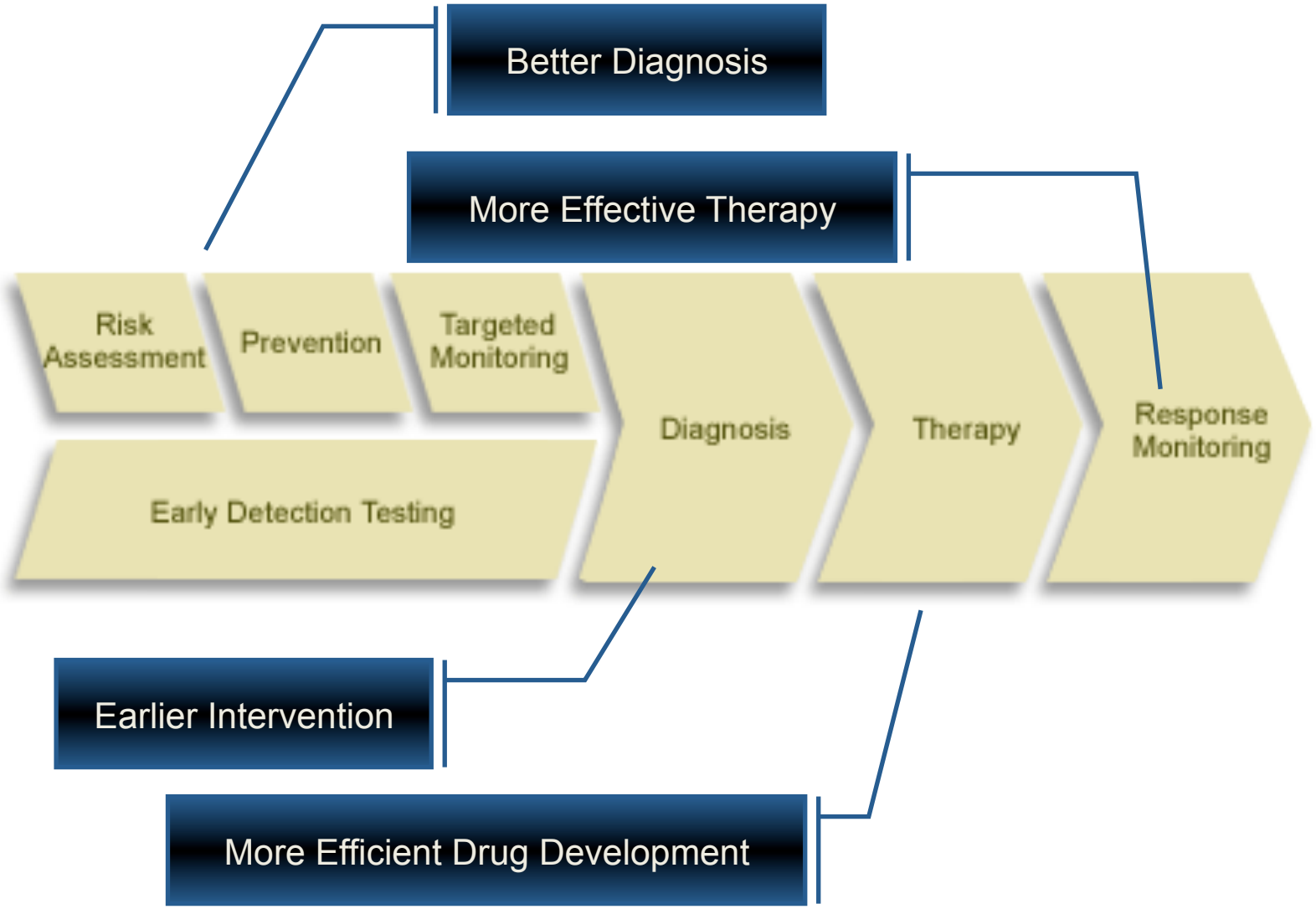
Pharmacogenomics: Industry

- First clinically available, FDA-approved genotype test in 2005 (Roche - Amplichip)
- Chip for SNPs in all CYP450 genes, 2005 (GE)
- Chip for p53 mutations expected mid-2006 (Roche)
- Affymetrix/ParAllele DMET Chip - 1,300 SNPs covering 185 genes (enzymes and transporters) involved in drug metabolism Spring, 2006

Pharmacogenomics - Industry

- Biomarker/Genetic targets:
  Abbott
  Johnson & Johnson
  Roche (Swiss)
- Pharmacogenomics for individualized Rx:
  Pfizer
  Bristol-Myers-Squibb
  Genentech
- Molecular diagnostic kits/devices:
  Roche expected sales for 2005: $6.5 billion
  expected sales by 2010: $12 billion

The Paradigm of Personalized Medicine



Conclusion

- Pharmacogenomics integrates genomic information in drug discovery
- Powerful tool for genetic information but wealth of data
- Environmental variation in Pharmacometabonomics
- Useful industrial applications
- All approaches participate in personalized healthcare