

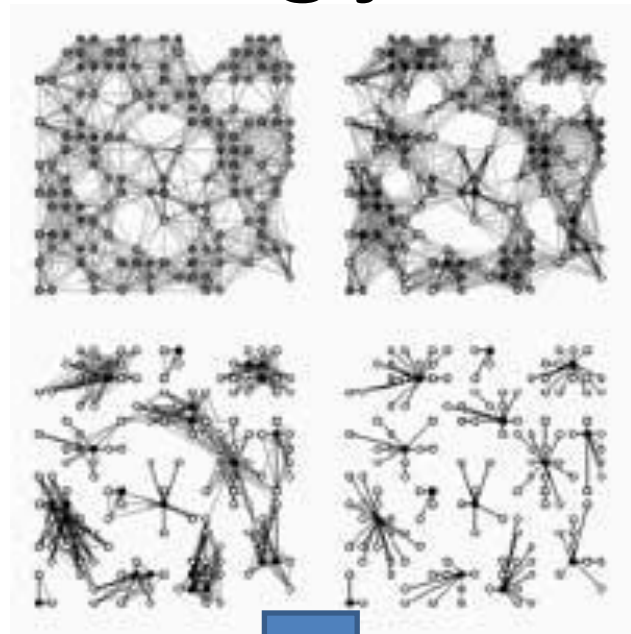
Sistemska biologija pri zdravljenju raka

Doc. dr. Kristina Gruden

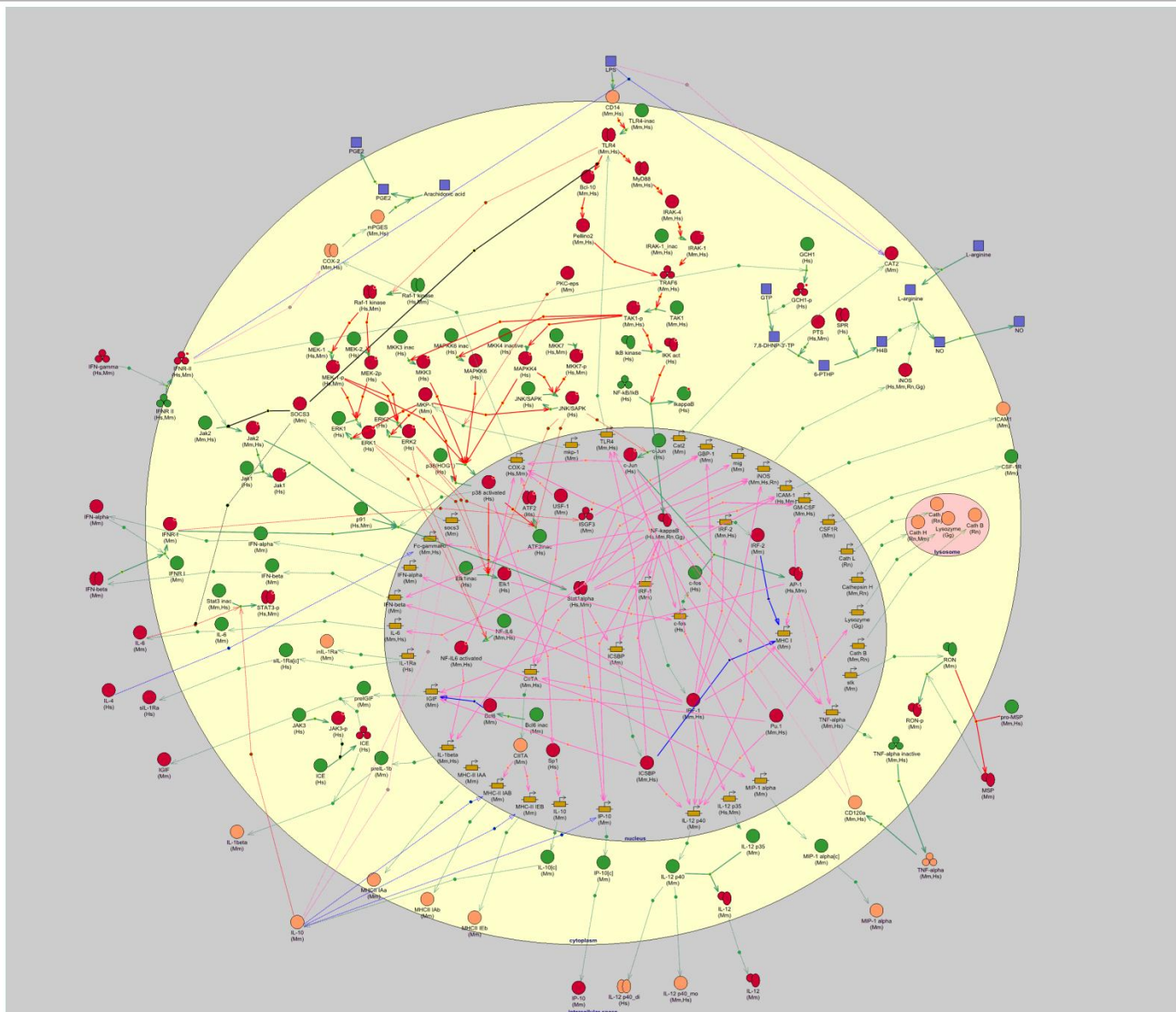
Oddelek za biotehnologijo in sistemske biologije
Nacionalni inštitut za biologijo

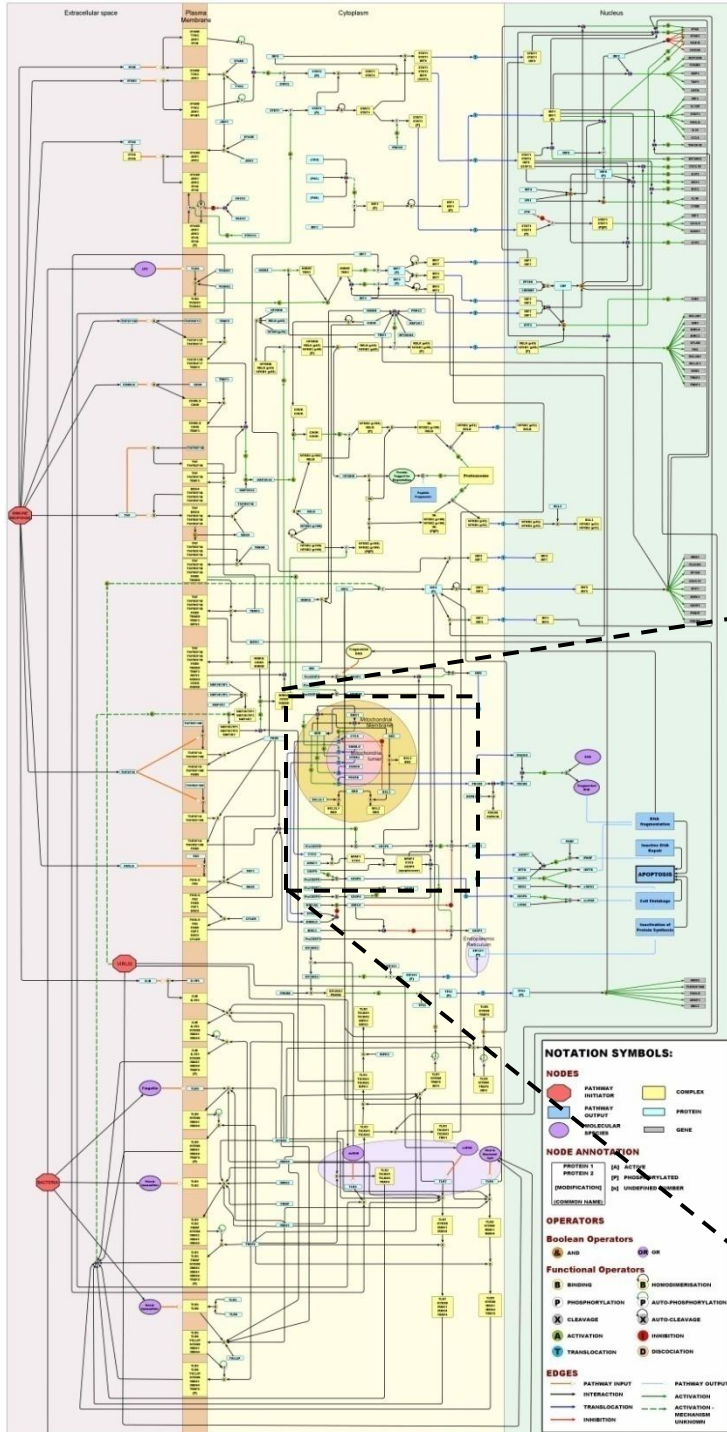
Kaj je sistemska biologija?

- razumevanje dinamike procesov v bioloških sistemih
- razumevanje delovanja organizmov
- Interdisciplinarnost



Kompleksni procesi – aktivacija makrofagov

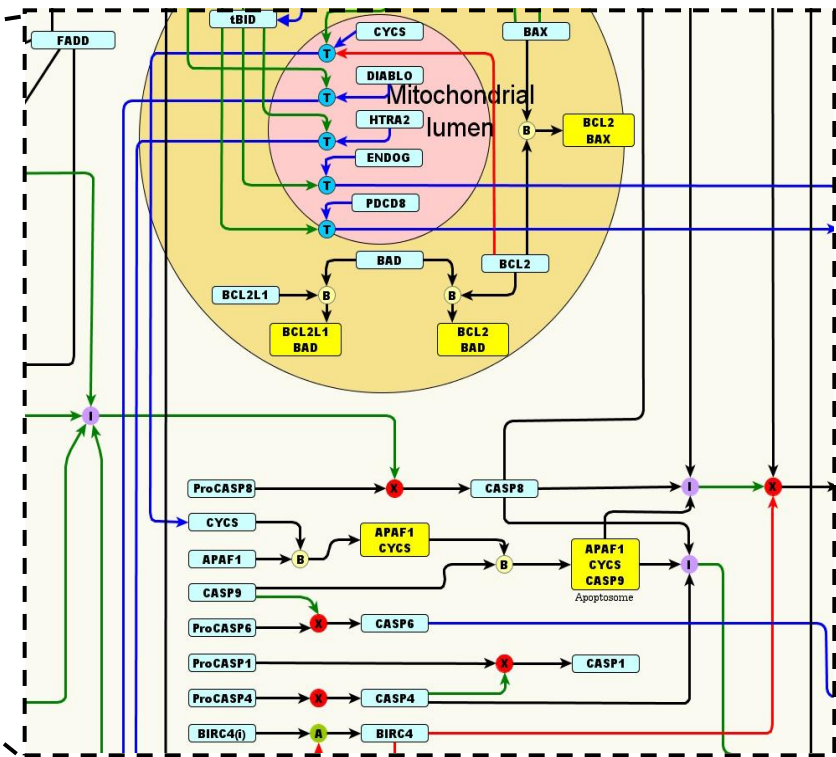




Framework Map of Macrophage Signalling Pathways

140 proteins, 99 complexes, 44 genes
285 interactions

Raza *et al.*, 2008, BMC Systems Biology. 2:36-51, 2008



Pristop sistemske biologije

- Opustimo tarčni pristop = iščemo in analiziramo le določene elemente sistema (gene, proteine, metabolite...)
- Sistematična analiza celotnega sistema

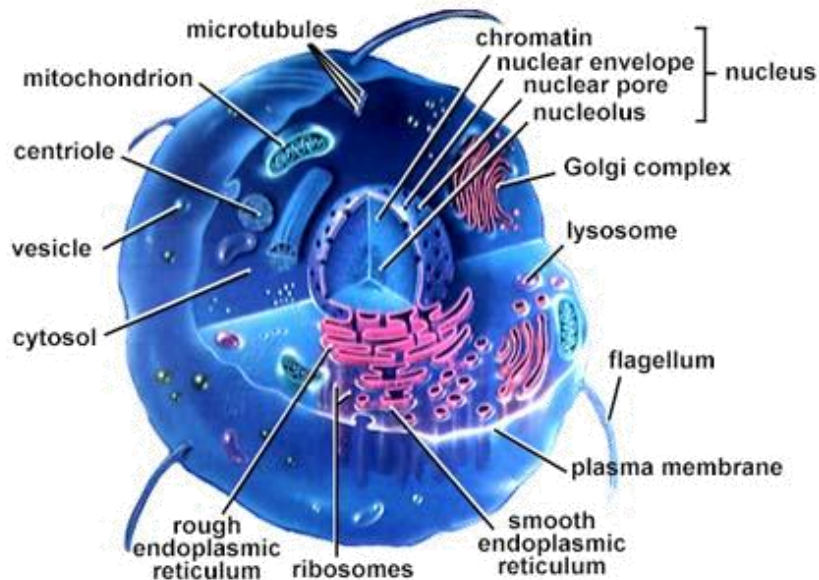
Pristop sistemske biologije

- Sistematičen opis elementov sistema
- Odkrivanje funkcije
- Modeliranje sistema

Interdisciplinarnost:

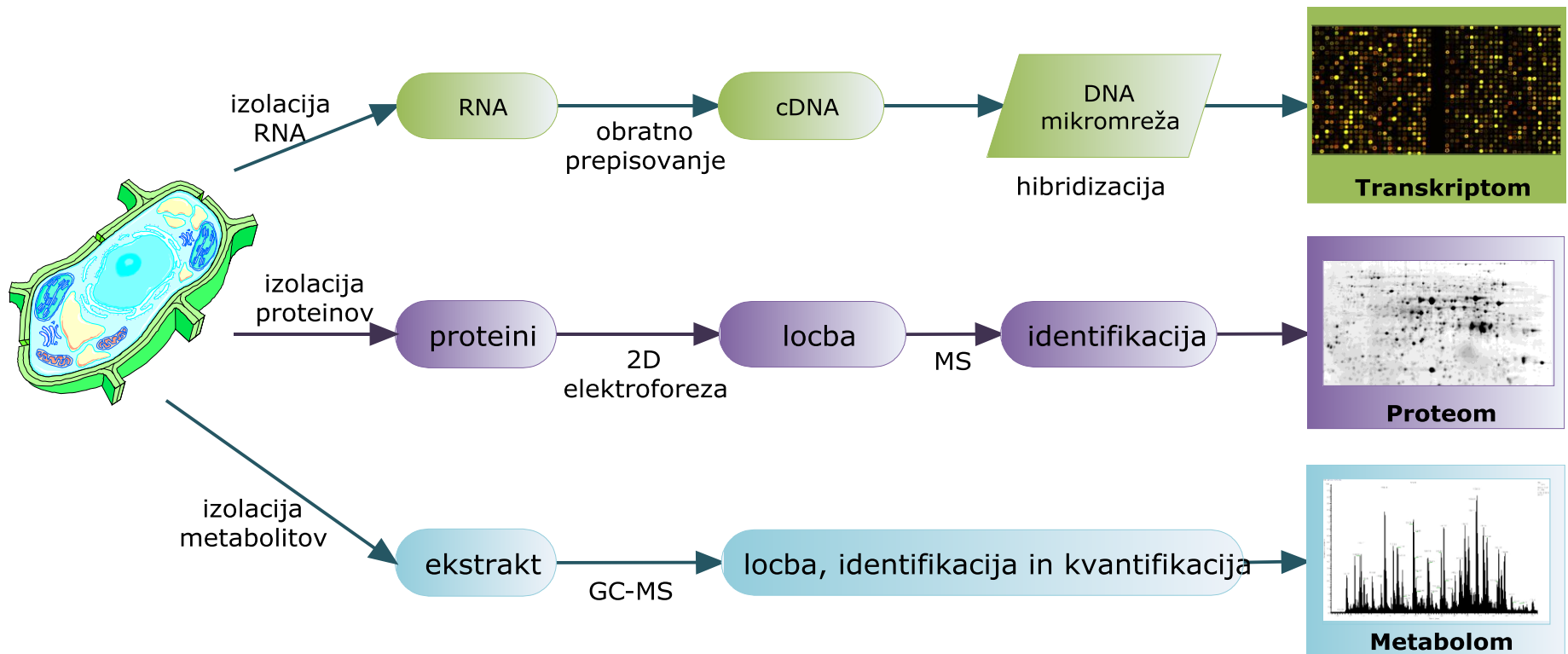
Biološke in medicinske znanosti +
matematika in računalništvo

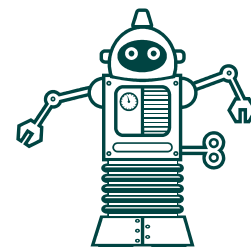
Omike



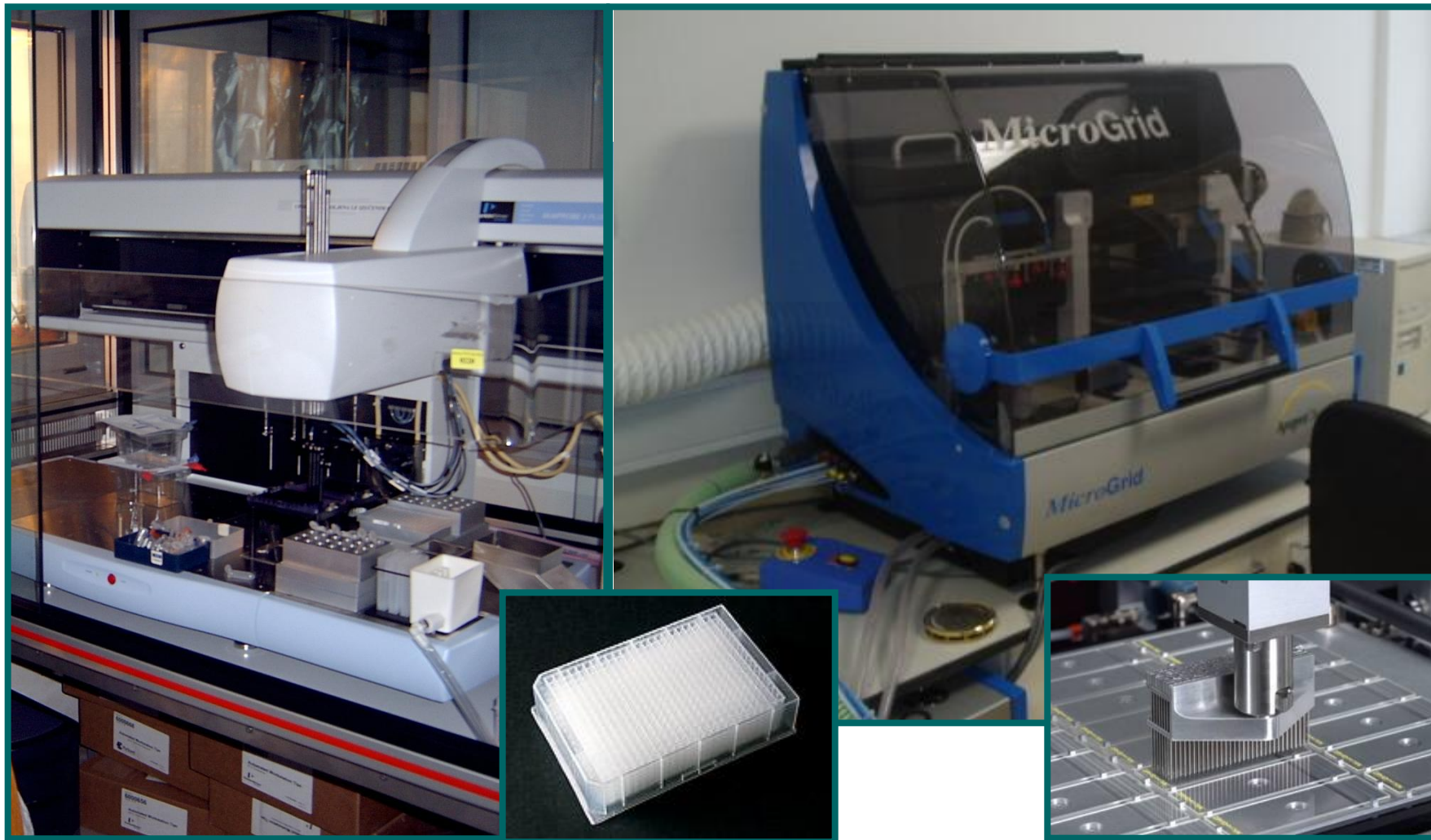
Term	Description
Genome	The full complement of genetic information both coding and non coding in the organism
Proteome	The protein-coding regions of the genome
Transcriptome	The population of mRNA transcripts in the cell, weighted by their expression levels
Physiome	Quantitative description of the physiological dynamics or functions of the whole organism
Metabolome	The quantitative complement of all the small molecules present in a cell in a specific physiological state
Phenome	Qualitative identification of the form and function derived from genes, but lacking a quantitative, integrative definition
Morphome	The quantitative description of anatomical structure, biochemical and chemical composition of an intact organism, including its genome, proteome, cell, tissue and organ structures
Interactome	List of interactions between all macromolecules in a cell
Glycome	The population of carbohydrate molecules in the cell
Secretome	The population of gene products that are secreted from the cell
Ribonome	The population of RNA-coding regions of the genome

Metode sistemske biologije





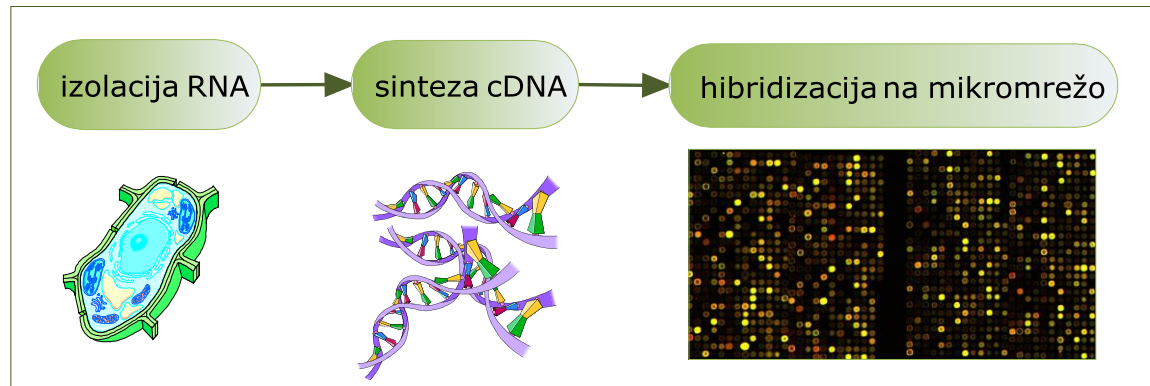
Metode - avtomatizacija



Transkriptomika

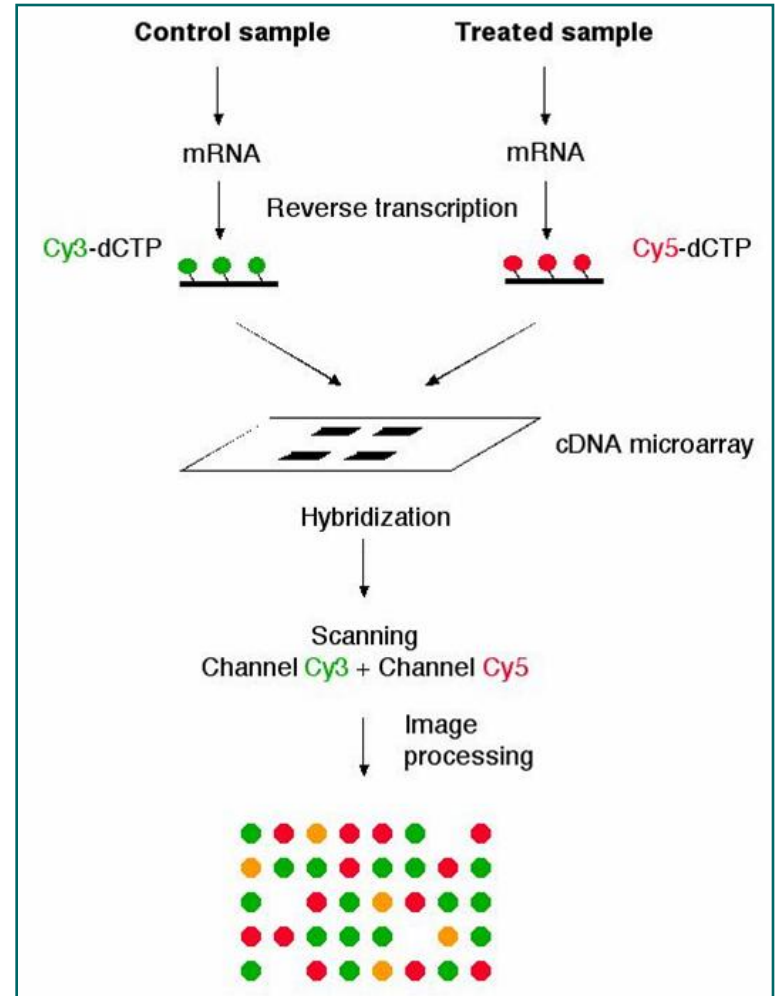
- Izražanje genov na nivoju celotnega organizma
- Relacija mRNA – aktivnost proteinov
- Metode

- DNA-mikromreže (microarrays)



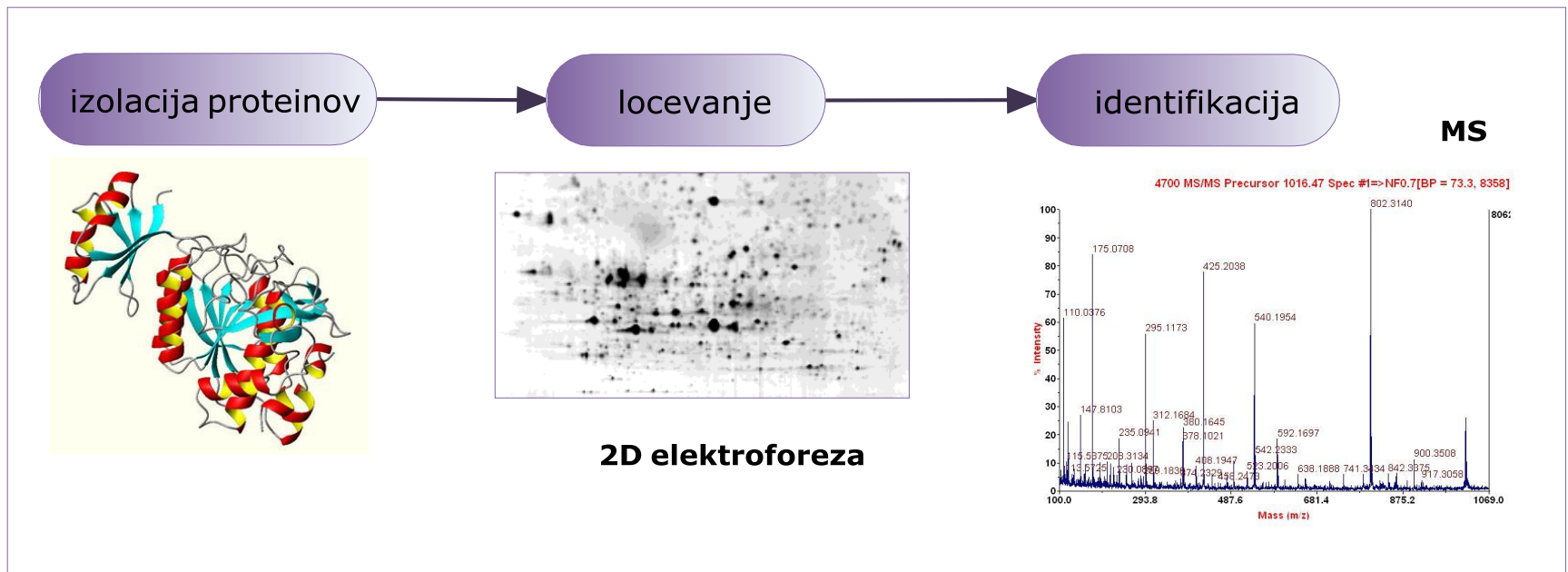
- PCR v realnem času (real-time PCR, qPCR)
- Zaporedna analiza izražanja genov (serial analysis of gene expression - SAGE)
- Masovno vzporedno sekveniranje podpisa (massive parallel signature sequencing – MPSS)

DNA-mikromreže



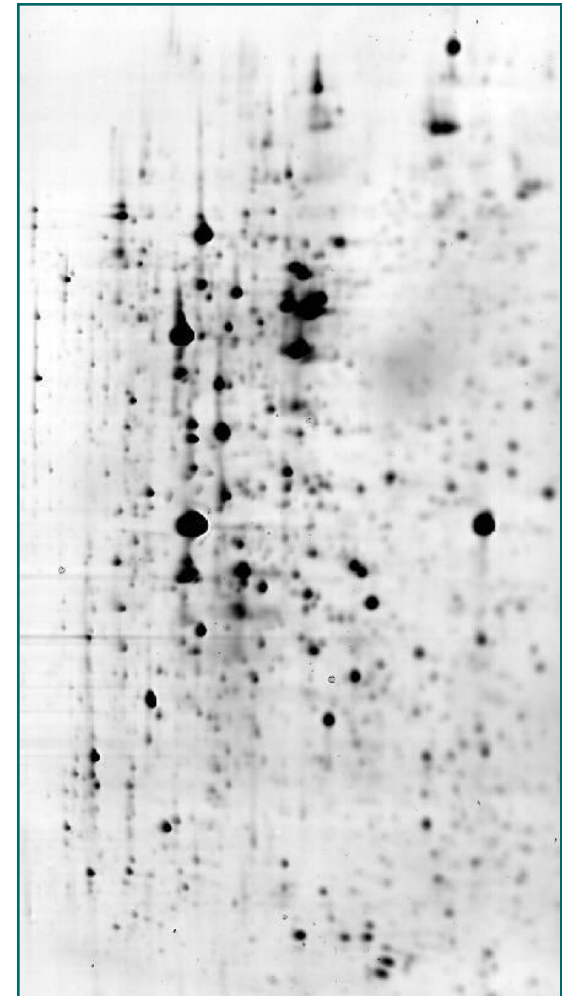
Proteomika

- Proteom = vsi proteini organizma
- geni < proteini
- Bolj realna slika dogajanja v celici kot transkriptom, vendar težja karakterizacija

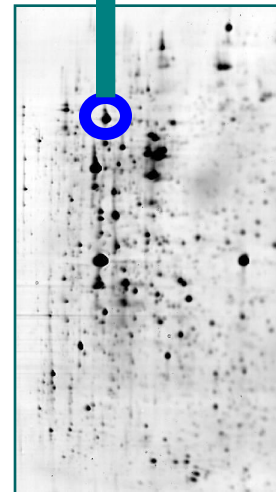
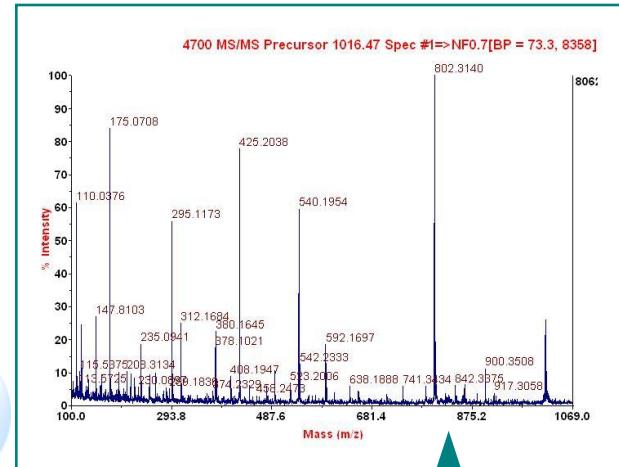
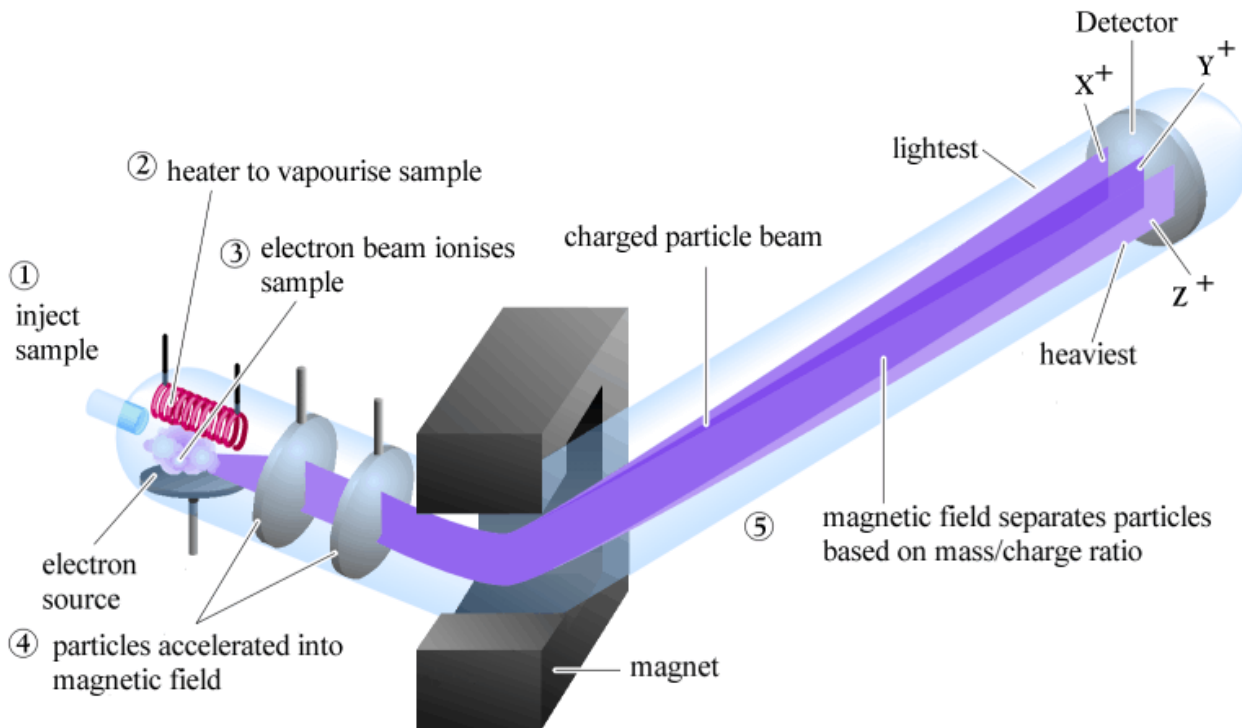


2D elektroforeza

- 1. dimenzija:
 - Gel s pH gradientom
 - Ločevanje glede na naboj
- 2. dimenzija
 - SDS PAGE elektroforeza
 - Ločevanje glede na molekulsko maso
 - Barvanje proteinov (srebro, fluorescentna barvila, protitelesa) → kvantifikacija

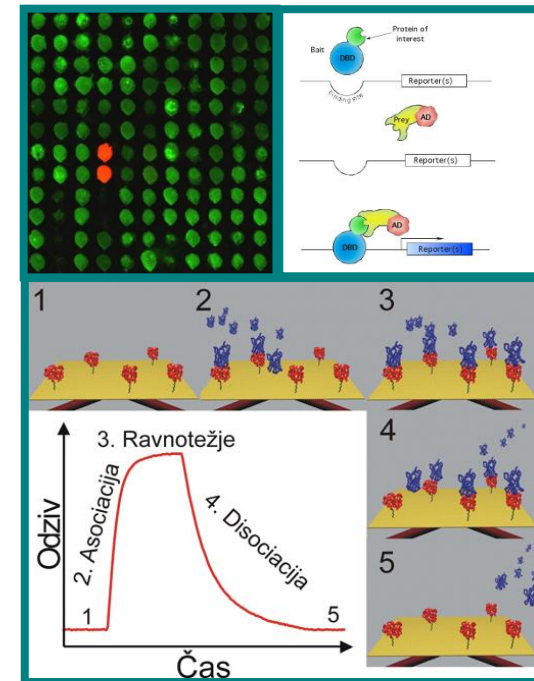


Masna spektrometrija



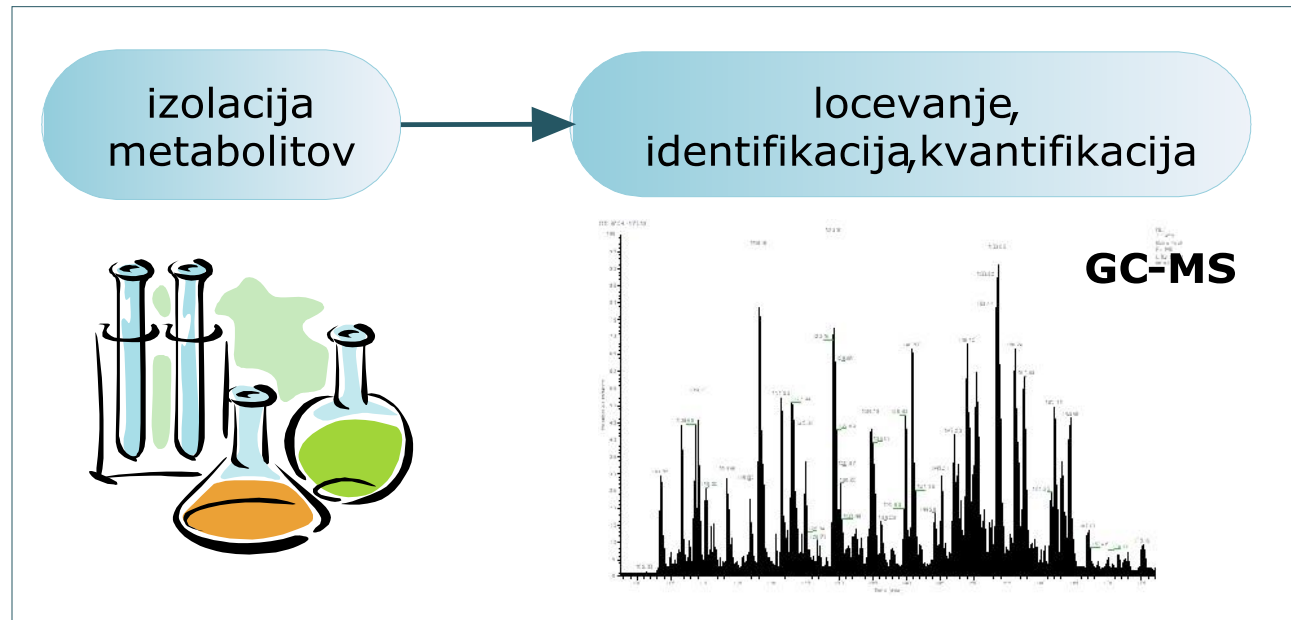
Proteomika

- Druge metode
 - Proteinske mikromreže
 - Dvohibridni sistem kvasovke (yeast two-hybrid system)
 - Površinska plazmonska resonanca
→ Interakcije
- Uporaba
 - Biomarkerji za bolezni

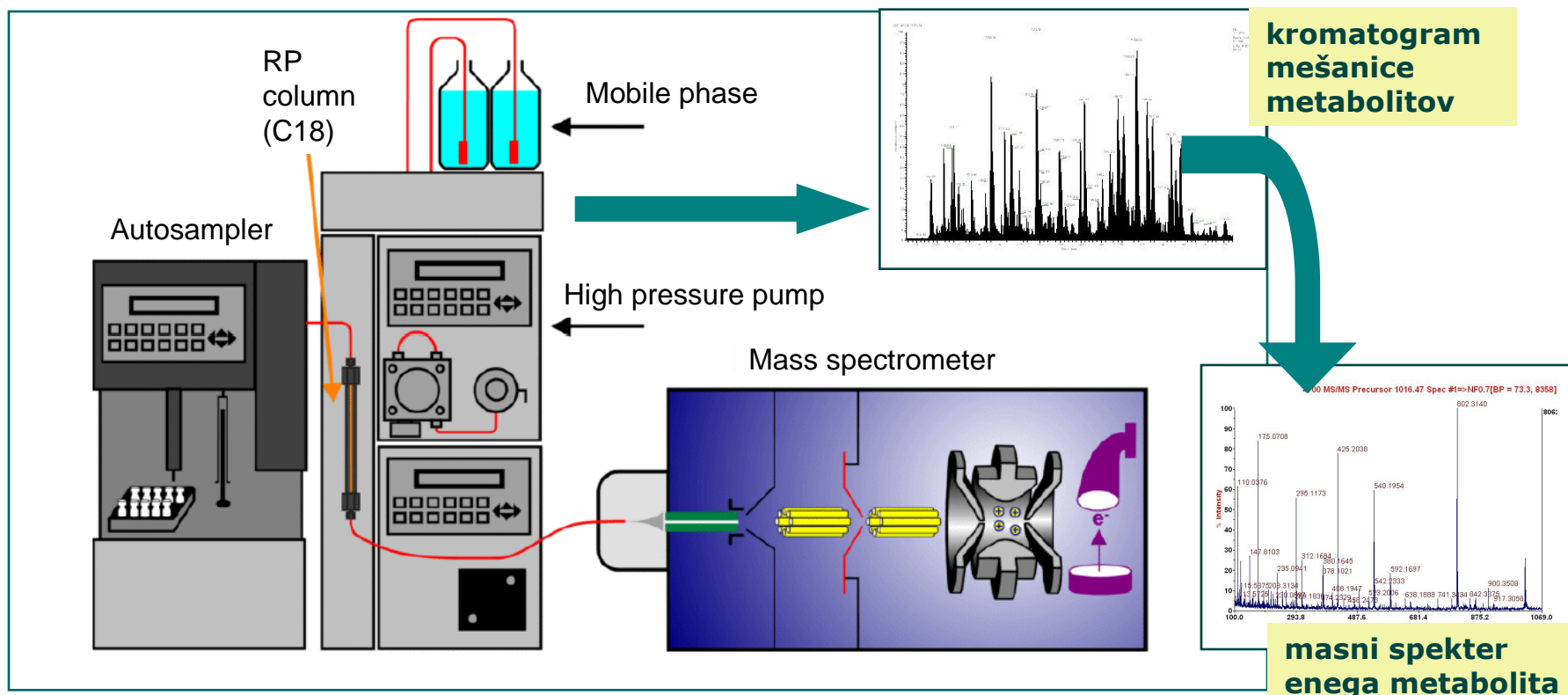


Metabolomika

- Metabolom = majhne (ne-proteinske) molekule različne kemijske sestave
- geni < proteini < metaboliti
- veliko število, različnost → problem identifikacije
- Uporaba: markerji bolezni, varnost hrane



Ločevanje in identifikacija metabolitov



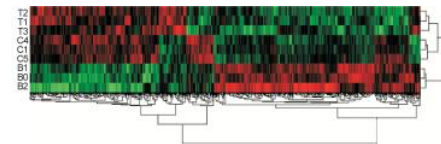
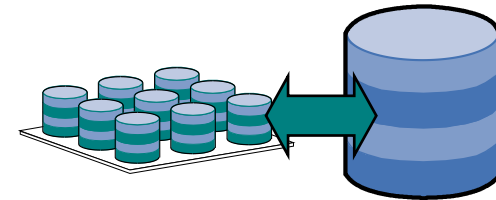
avtomatski vzorčevalnik

tekočinska kromatografija

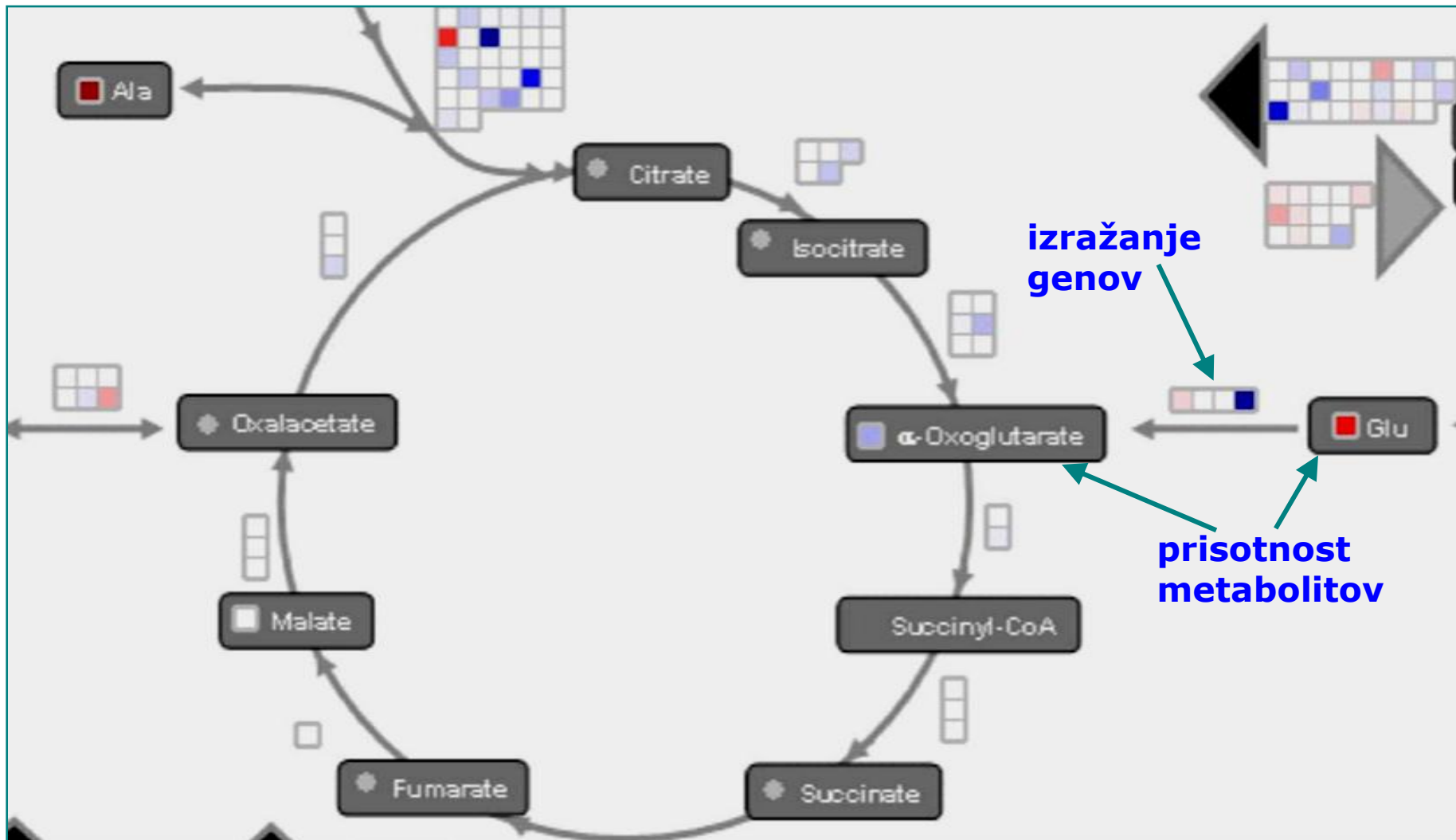
masni spektrometer

Bioinformatika

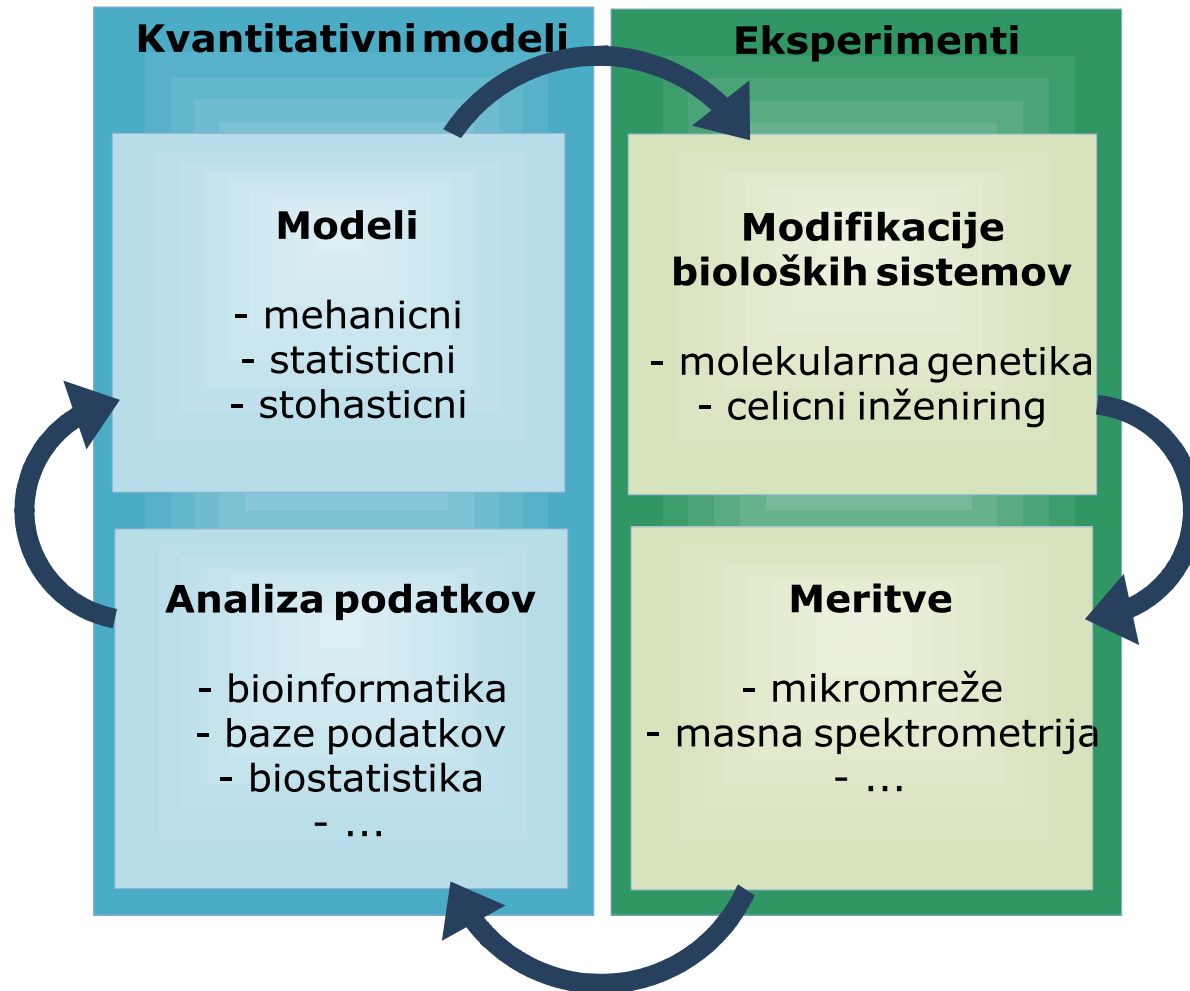
- Velika količina podatkov!
- Naloge bioinformatike:
 - Shranjevanje podatkov: baze, odlagališča
 - Kontrola kakovosti
 - Statistična obdelava
 - Vizualizacija podatkov
 - Modeliranje procesov
 - Podatkovno rudarjenje



Orodja za vizualizaciju

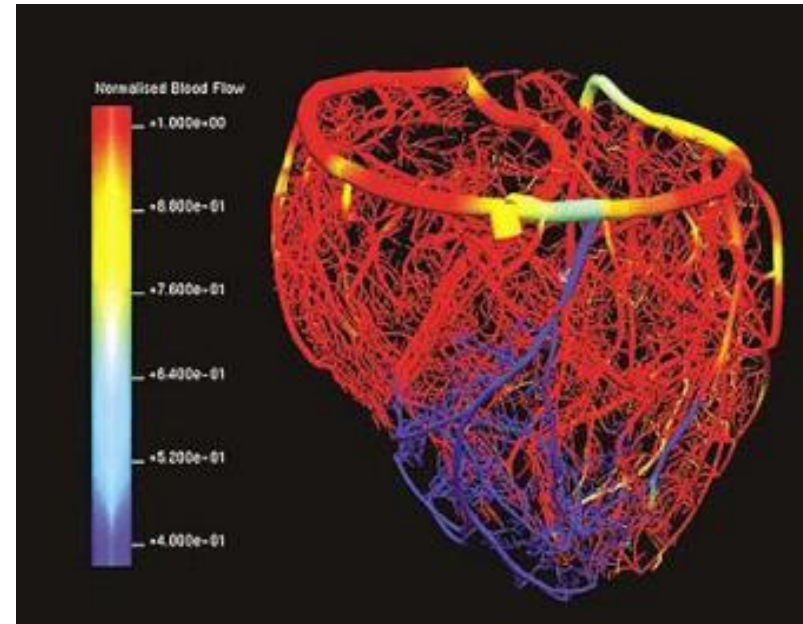


Modeliranje procesov



Modeliranje procesov

- Premalo eksperimentalnih podatkov in orodij za integracijo
- Prokariontski modeli
 - Enocelični, majhen genom, omejene metabolne kapacitete
- Model človeškega srca
 - Nobel, 1960

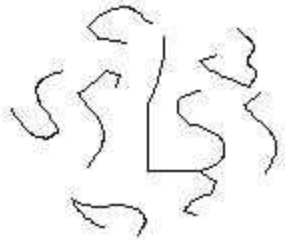


Spremljanje izražanja genov

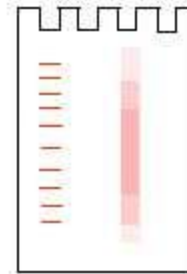
- Transkriptom:
 - Tehnologija DNA mikromrež
 - Potrjena s strani FDA

'Northern' hibridizacija

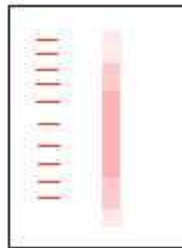
1. Izolacija RNA



2. Elektroforeza

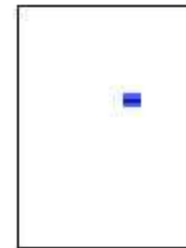


3. Prenos na membrano

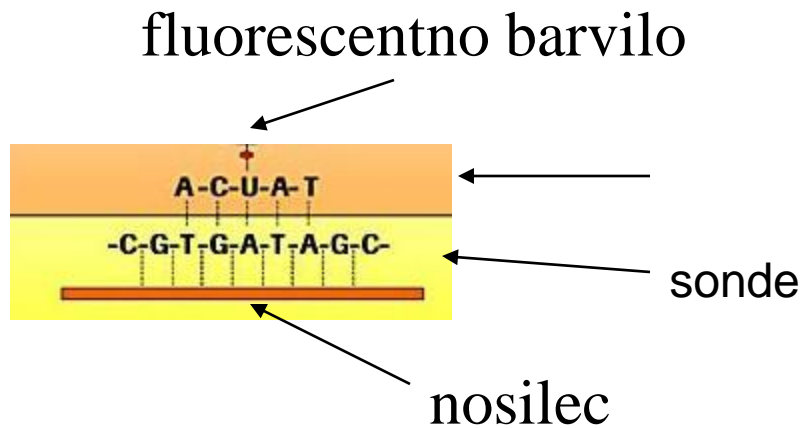


4. Hibridizacija s sondo

= en gen



Princip hibridizacije



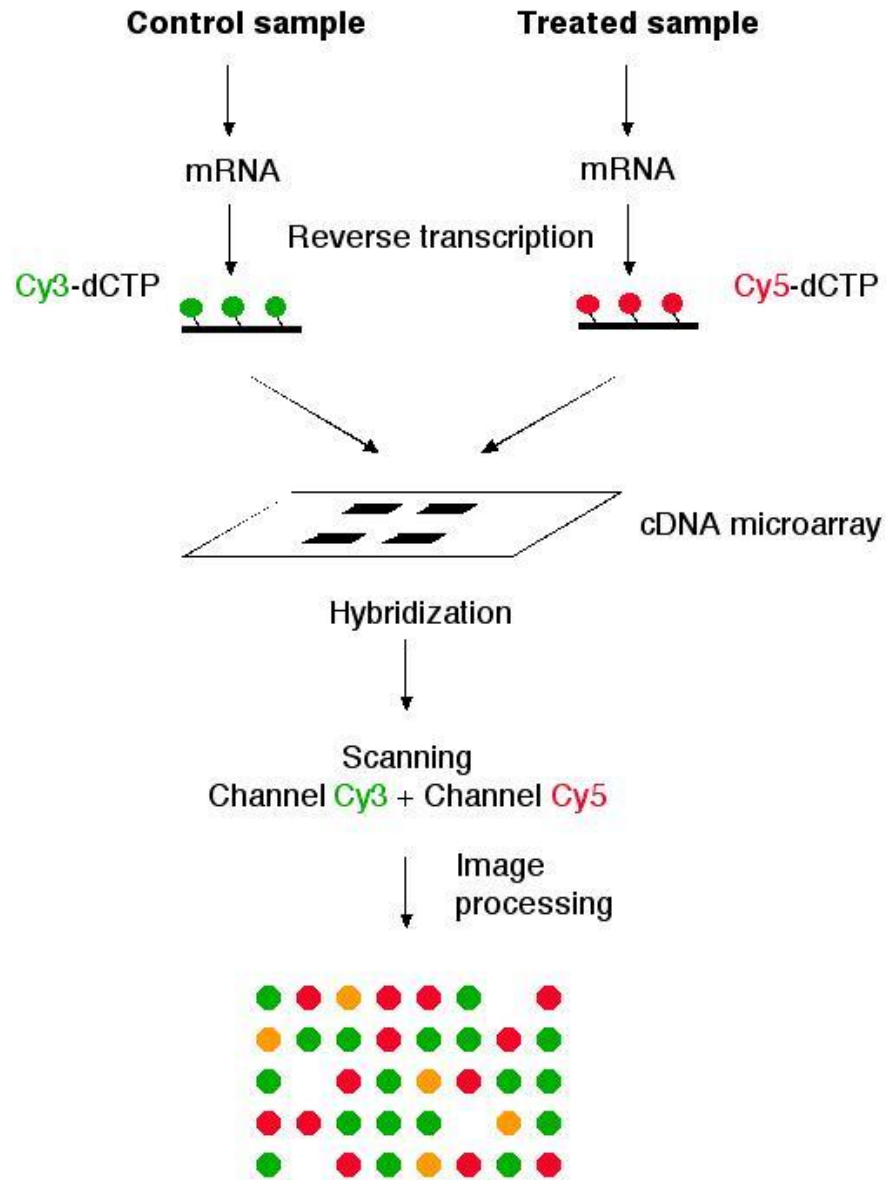
Cy3, Cy5

Mešanica mRNA

Izbrani geni

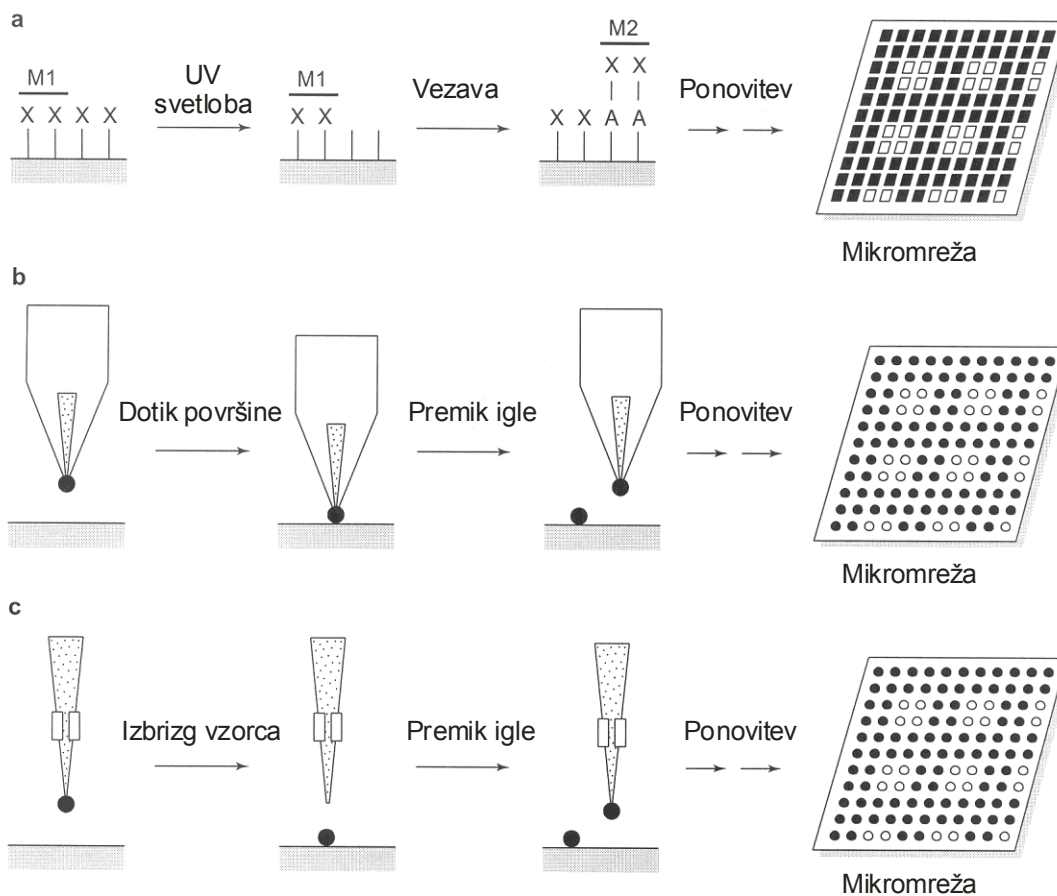
Objektnik

Hybridization



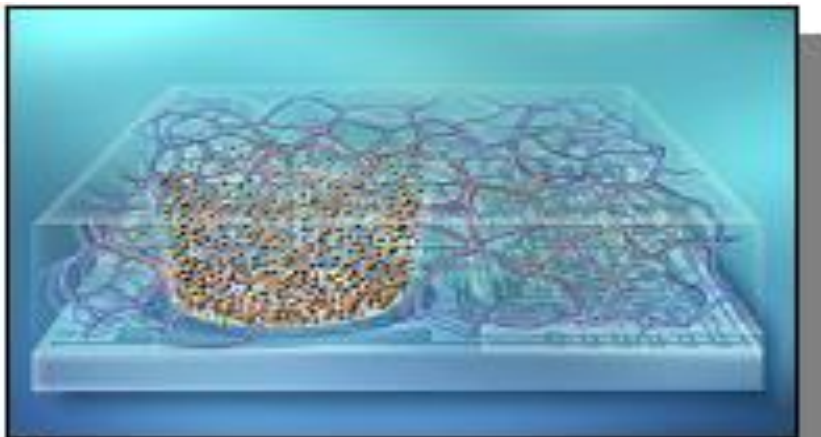
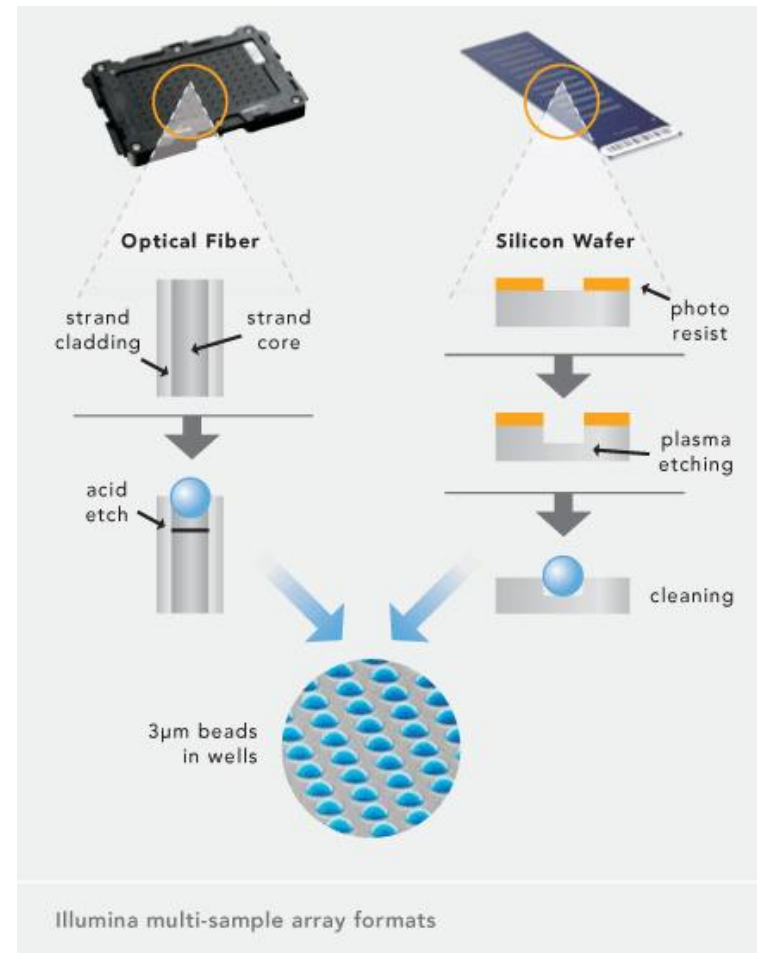
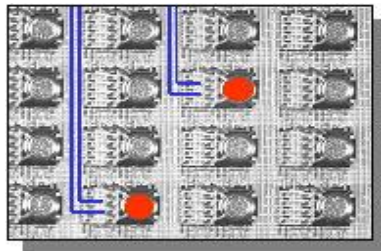
Princip tehnologije genskih čipov

- Priprava čipov



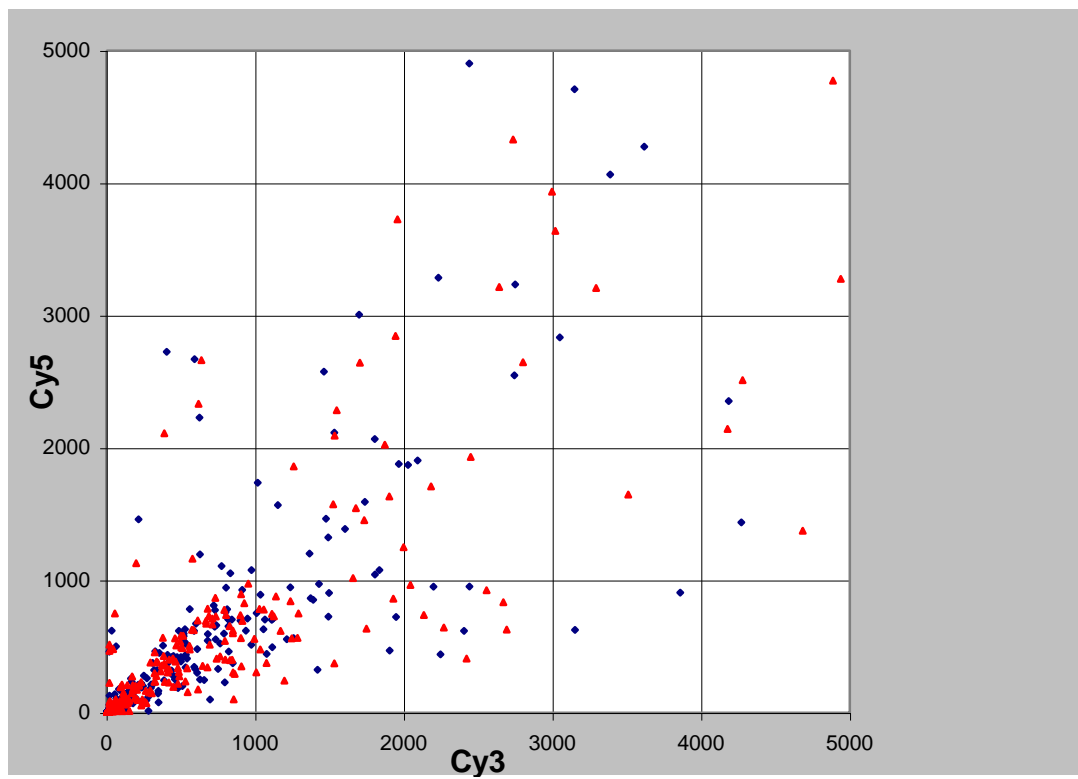
Princip tehnologije genskih čipov

- Priprava čipov



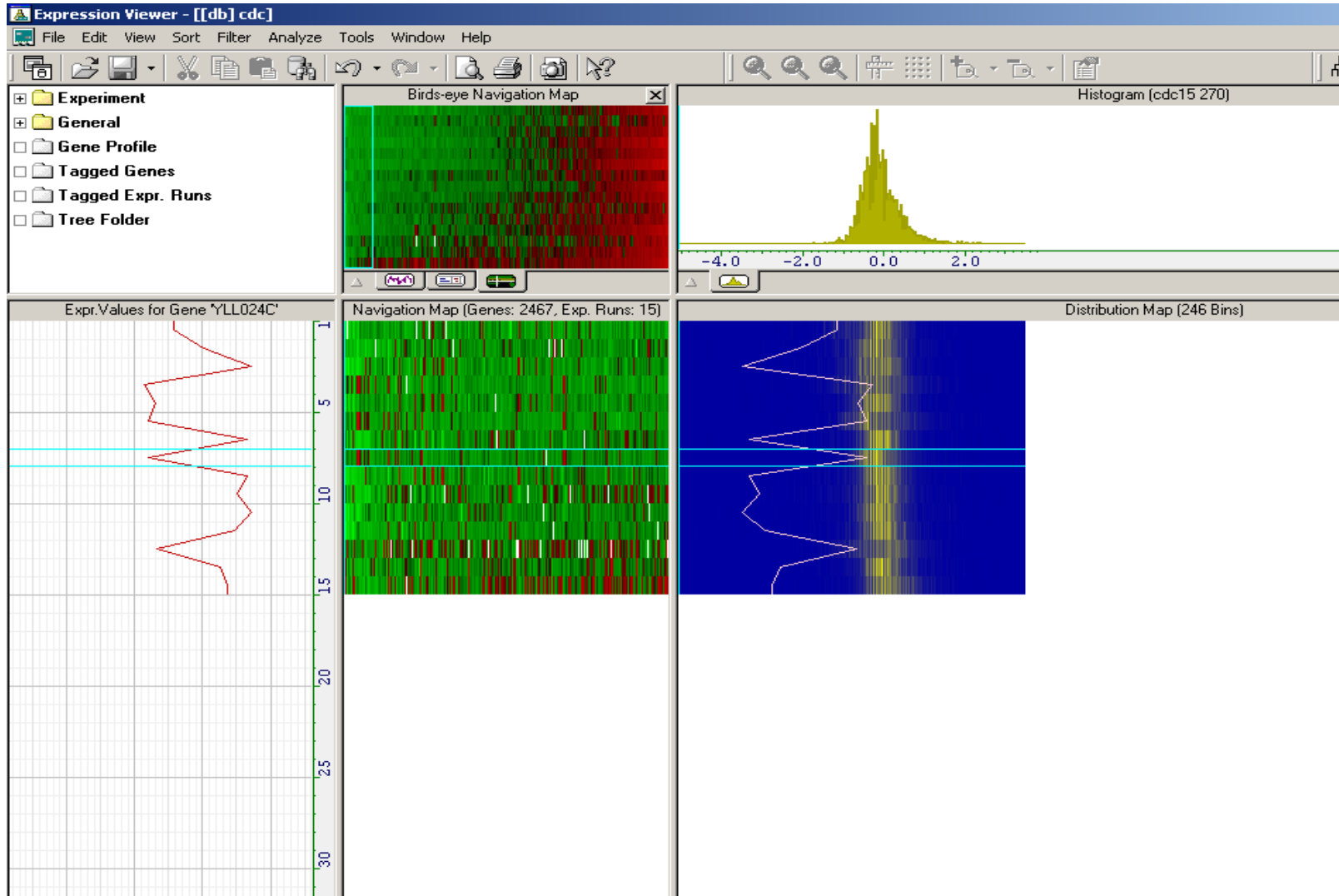
Genski čipi - analiza podatkov

Tretirano
Tkivo
Ali
Bolezensko
stanje



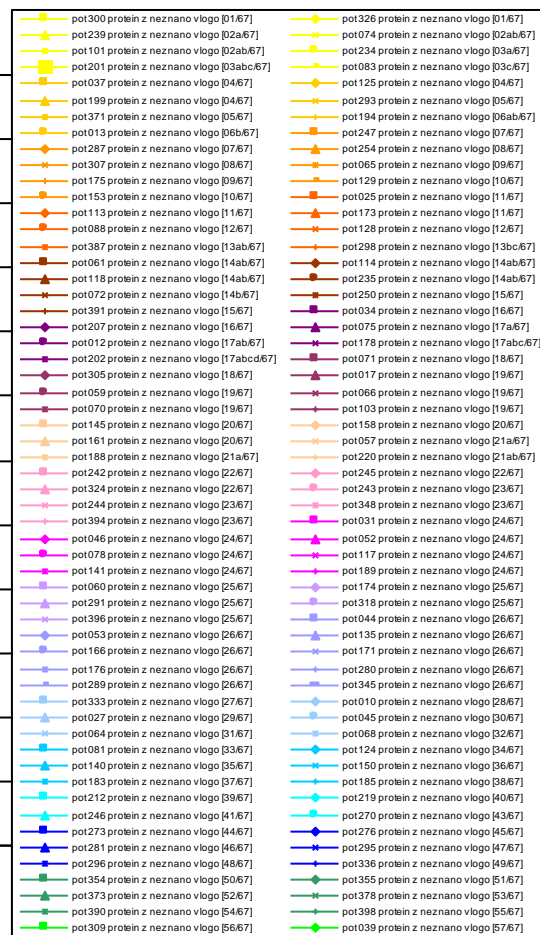
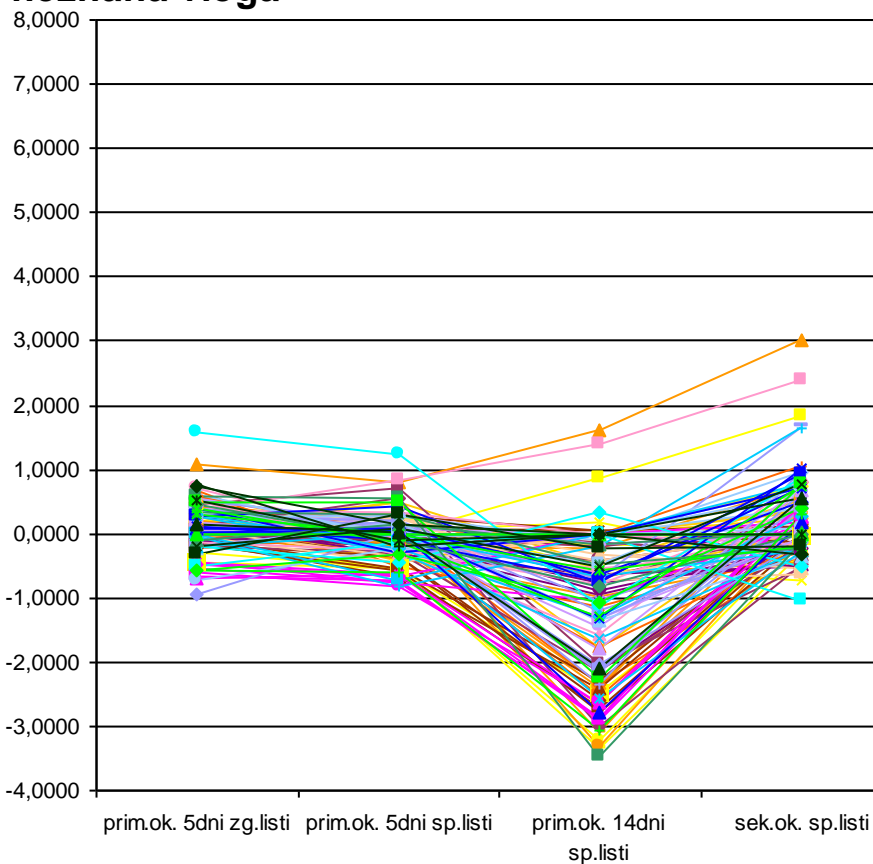
Kontrolno tkivo

Genski čipi - analiza podatkov



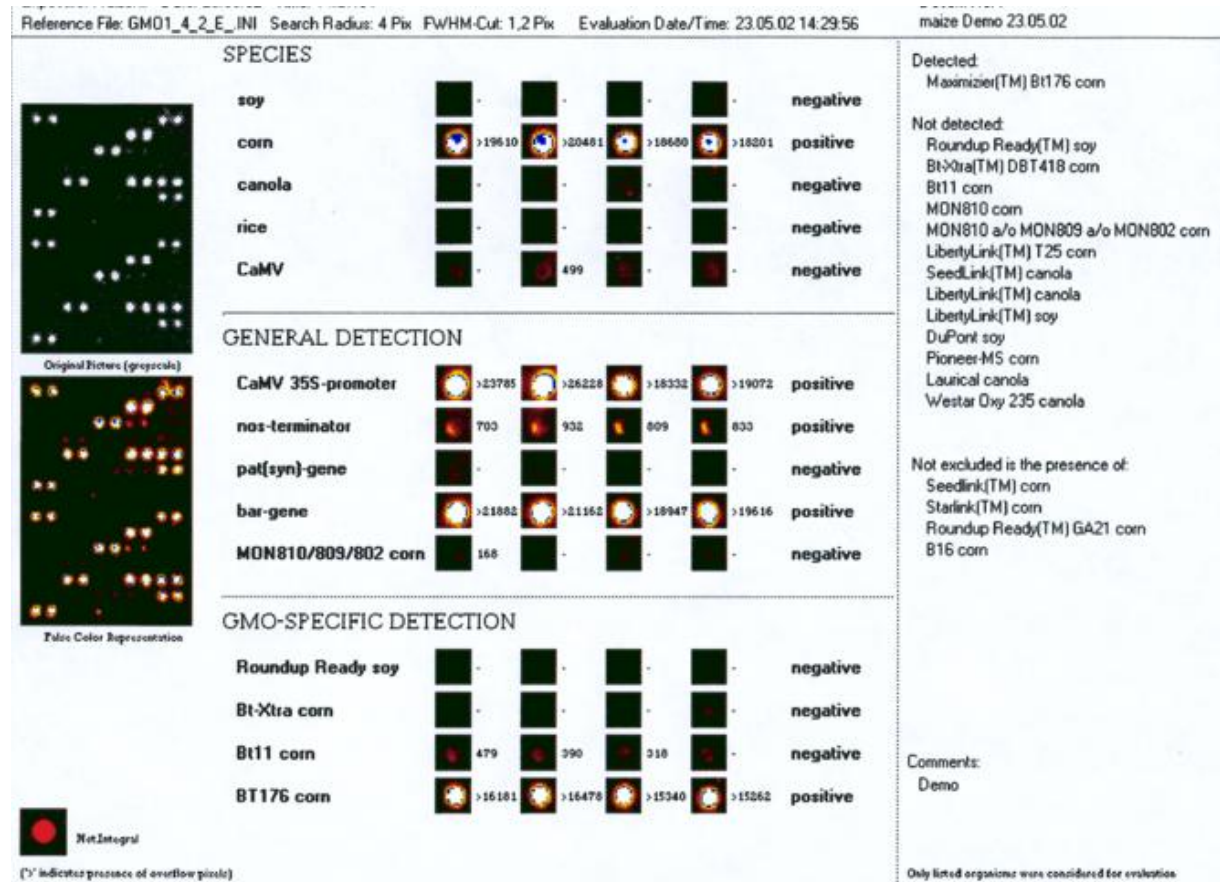
Genski čipi - analiza podatkov

neznana vloga



Primeri uporabe

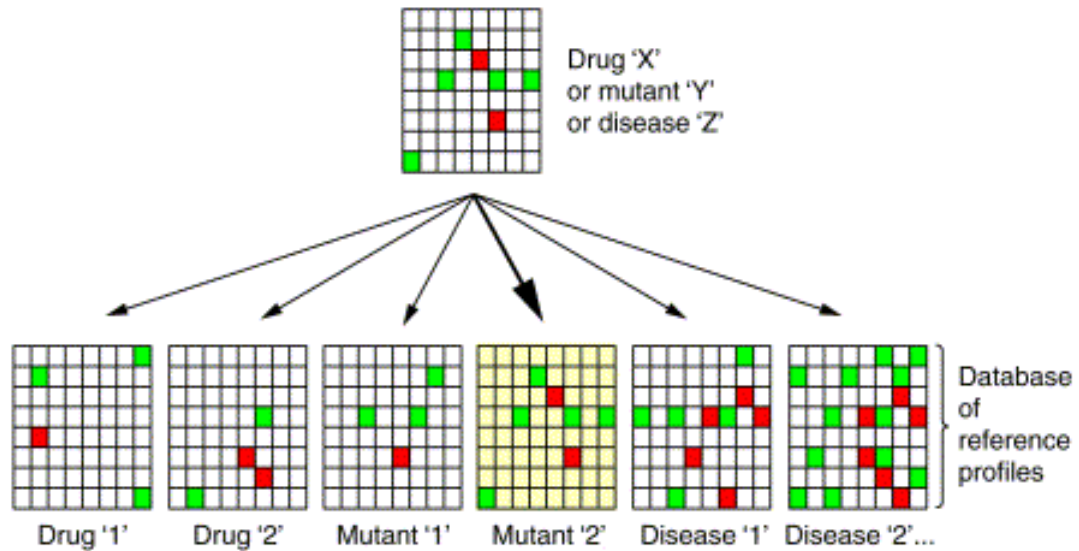
- Detekcija patogenov



Primeri uporabe

- za diagnostiko in terapijo
 - rak
 - prognoza bolezni
 - uspešnost terapije

Primeri uporabe



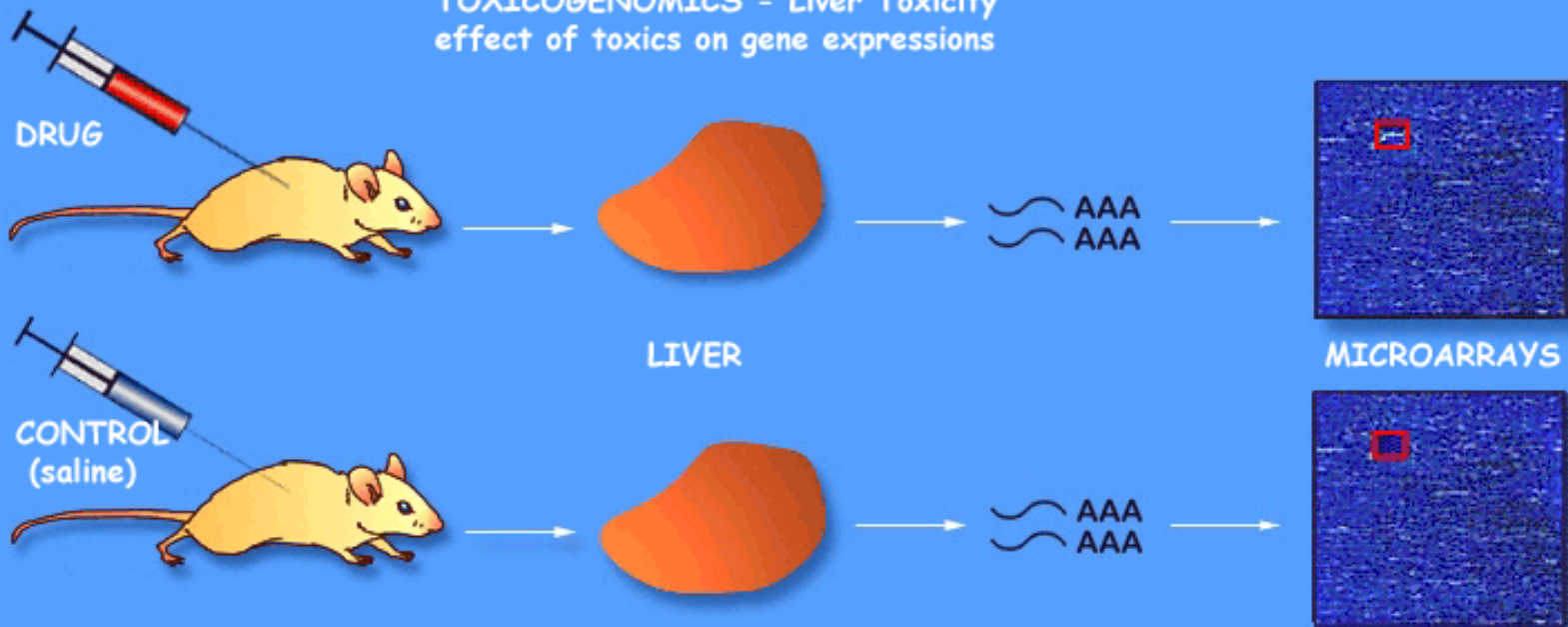
Current Opinion in Chemical Biology

Personalizirano zdravljenje

Uporaba v biotehnologiji

- Iskanje novih zdravil
- Razvoj novih terapij
 - V obeh primerih razumevanje mehanizmov delovanja
 - Predvsem pri multifactorialnih boleznih
- Testiranje stranskih učinkov zdravil
- Izboljšava proizvodnjih procesov

TOXICOGENOMICS - Liver Toxicity
effect of toxics on gene expressions



tells which genes are increased/decreased in presence of toxics in rat model

Systems Biology Brings Hope Of Speeding Up Drug Development

- 28 de Enero de 2009

Almost evermore so often day brings news of an apparent breakthrough *against cancer, infectious diseases, or metabolic conditions coextensive diabetes*, but these rarely translate into effective therapies or drugs, and even if they do clinical development most often takes well concluded a decade. One dialectics is that medical research is conducted in highly fragmented batchs focusing on right on pathways or components leading to drugs that bow out not to exertion properly or to have dangerous side effects after cycles of animal and next clinical corroborating in humans. that process is expensive and wasteful, resulting from the fact that at present researchers exiguity tools to assess in advance how candidate drugs performance opposite the humans whole biological classification. *The discipline of systems biology represents an attempt to unite the medical research community behind a common approach to understanding and modelling the complex interactions of the human, leading to more effective and faster drug development.*

Sklepi

- Pristop sistemske biologije bo omogočil reševanje kompleksnih problemov kot sta npr. rak ali sladkorna bolezen.
- Trenutno je še premalo podatkov za postavitve natančnih modelov delovanja celic, tkiv, organov in organizmov in posledično uporabe v medicini
- Vizija Digital Human leta 2050