Cancer genetics-guided discovery of serum biomarker signatures for diagnosis and prognosis of human cancer

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Take Home Message

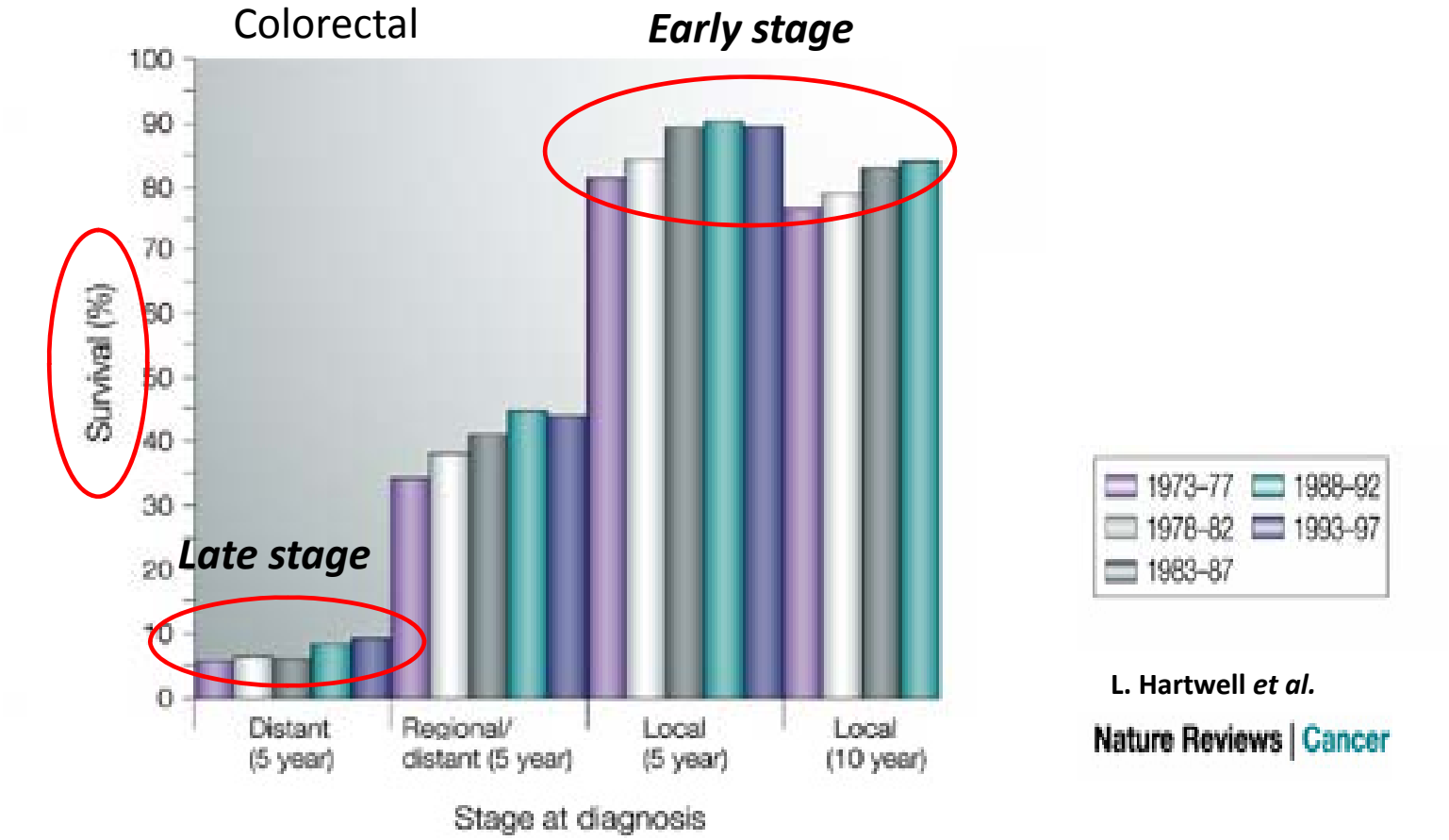
- Thousands of complete (cancer) genomes are being determined

- The genomic information, modulated by external factors, determines the phenotype (disease).

- Genomic analyses need to be complements by measurements of the expressed information (proteins) that indicate the ACUTE STATE of a person

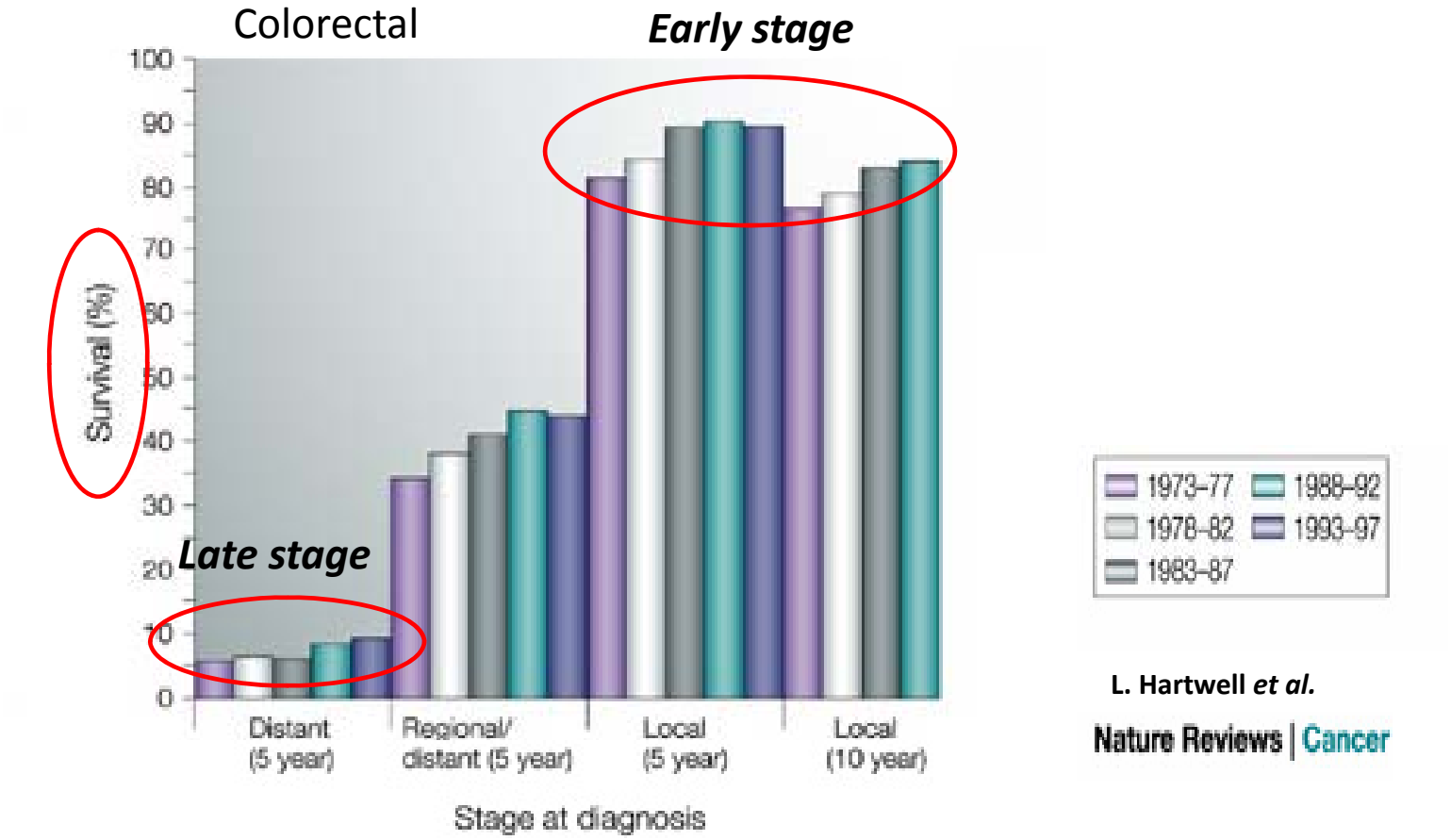
- Measurements to “remotely” measure the state of a tissue are becoming a reality

Cancer: Diagnosis and Survival



Same observations for other cancers: lung, breast, and prostate cancers

Cancer: Diagnosis and Survival



Same observations for other cancers: lung, breast, and prostate cancers

There is an urgent need to detect disease early and to optimize treatment for a patient

JOINT STATEMENT BY PRESIDENT CLINTON AND PRIME MINISTER TONY BLAIR

PRESIDENT CLINTON ANNOUNCES THE COMPLETION OF THE FIRST SURVEY OF THE ENTIRE HUMAN GENOME Hails Public and Private Efforts Leading to This Historic Achievement June 26, 2000 Today….

Press release US. Govt.

....announced that the international Human Genome Project and Celera Genomics Corporation have both completed an initial sequencing of the human genome -- the genetic blueprint for human beings.He congratulated the scientists working in both the public and private sectors on this landmark achievement, which promises to lead to a new era of molecular medicine, an era that will bring new ways to prevent,diagnose, treat and cure disease.

Now, scientists will be able to use the working draft of the human genome to:

* Alert patients that they are at risk for certain diseases.

* Reliably predict the course of disease.

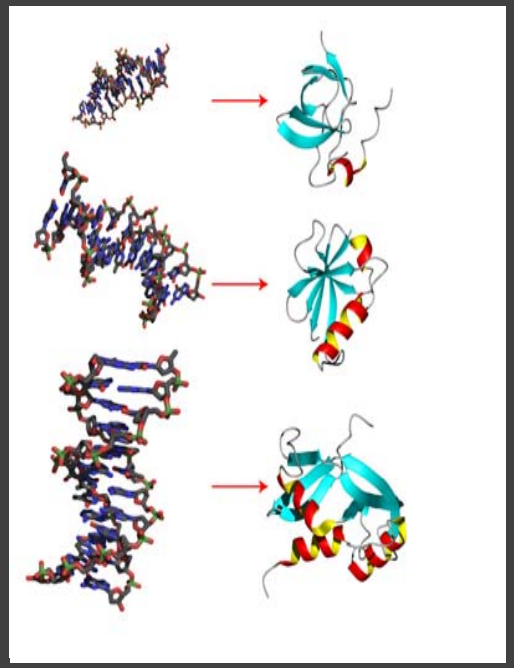
•Precisely diagnose disease and ensure the most effective treatment is used.

•Developing new treatments at the molecular level.

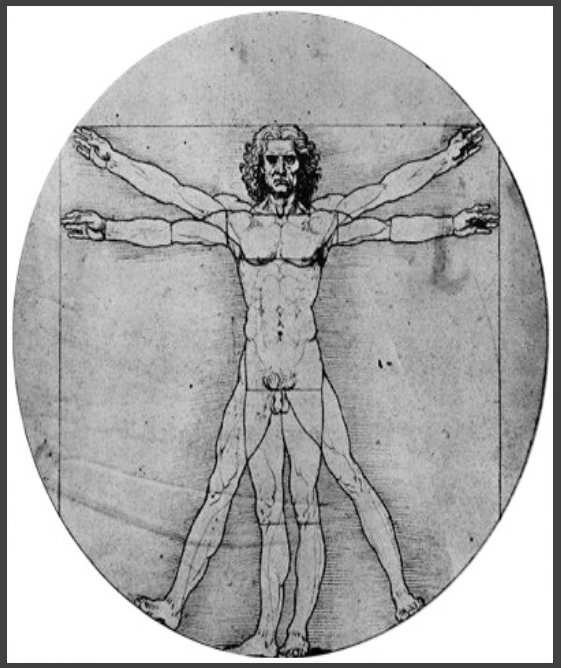
Press release US. Govt.

Genotype --- Phenotype

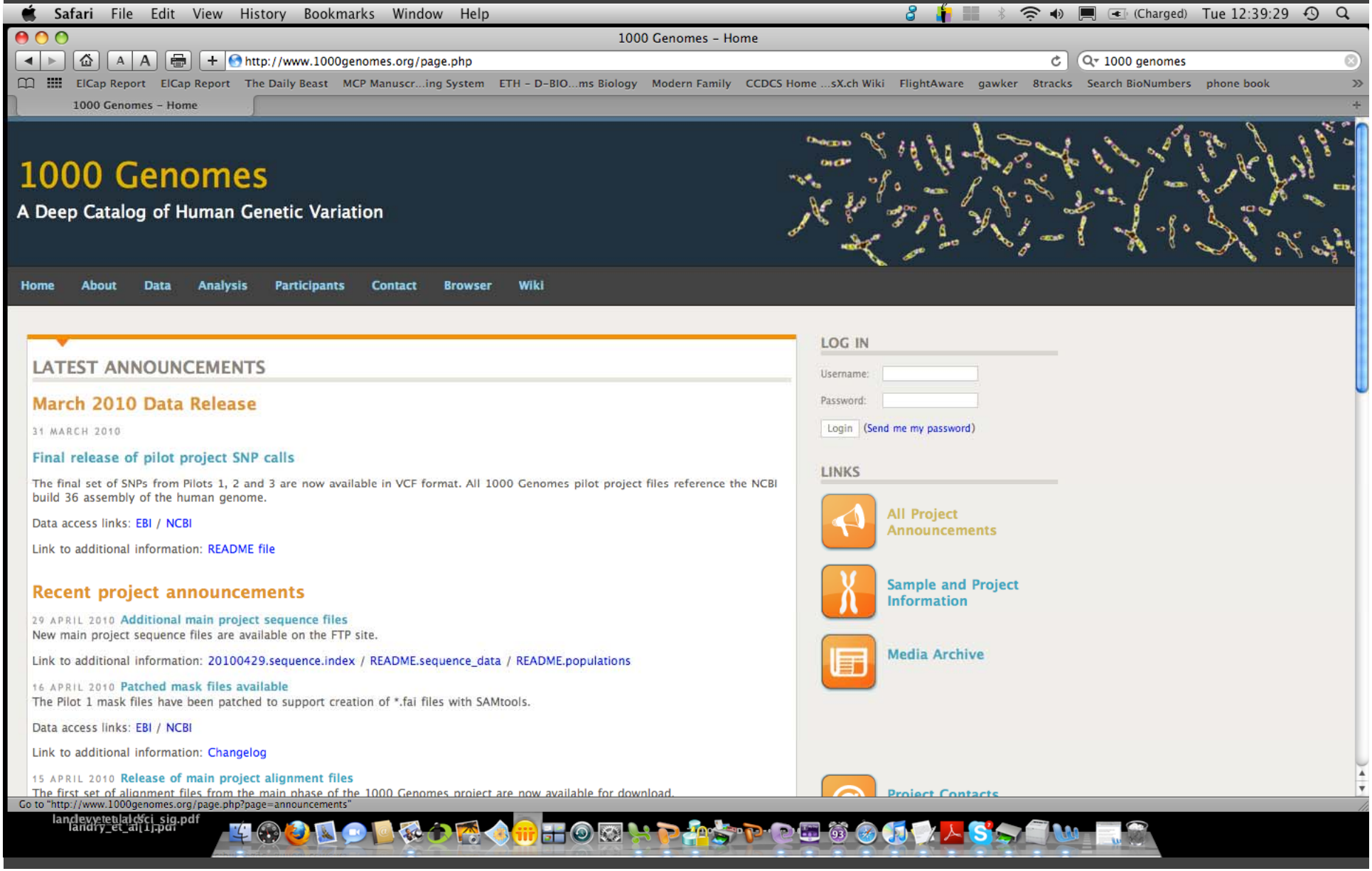
Genomics



Phenotype/Disease

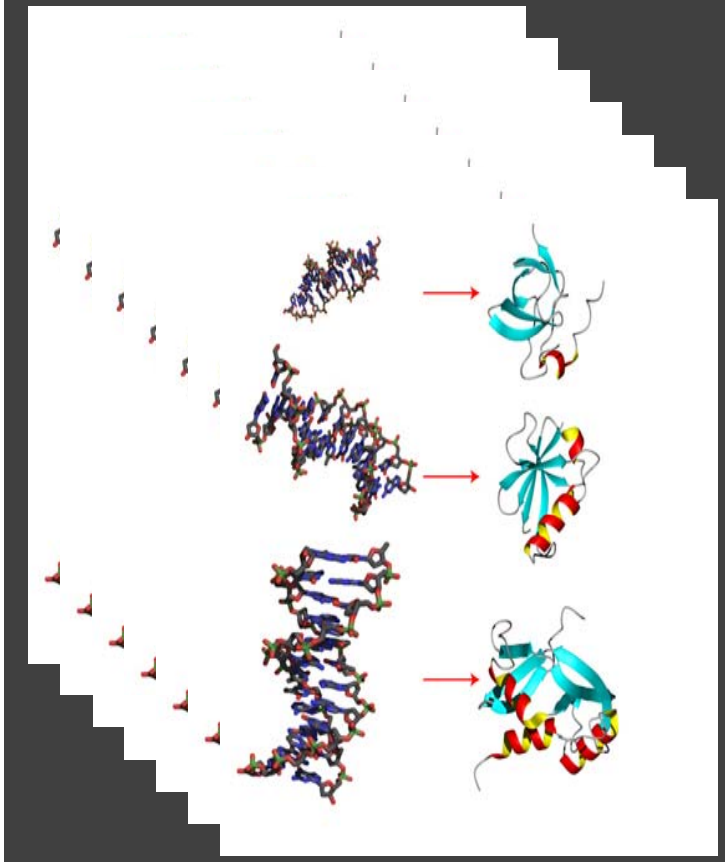


Multiple Genomes/Cancer Genomes

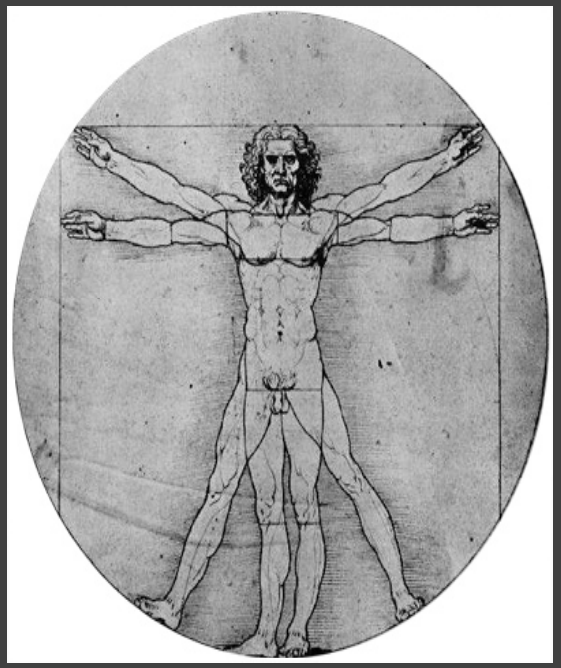


Genotype --- Phenotype

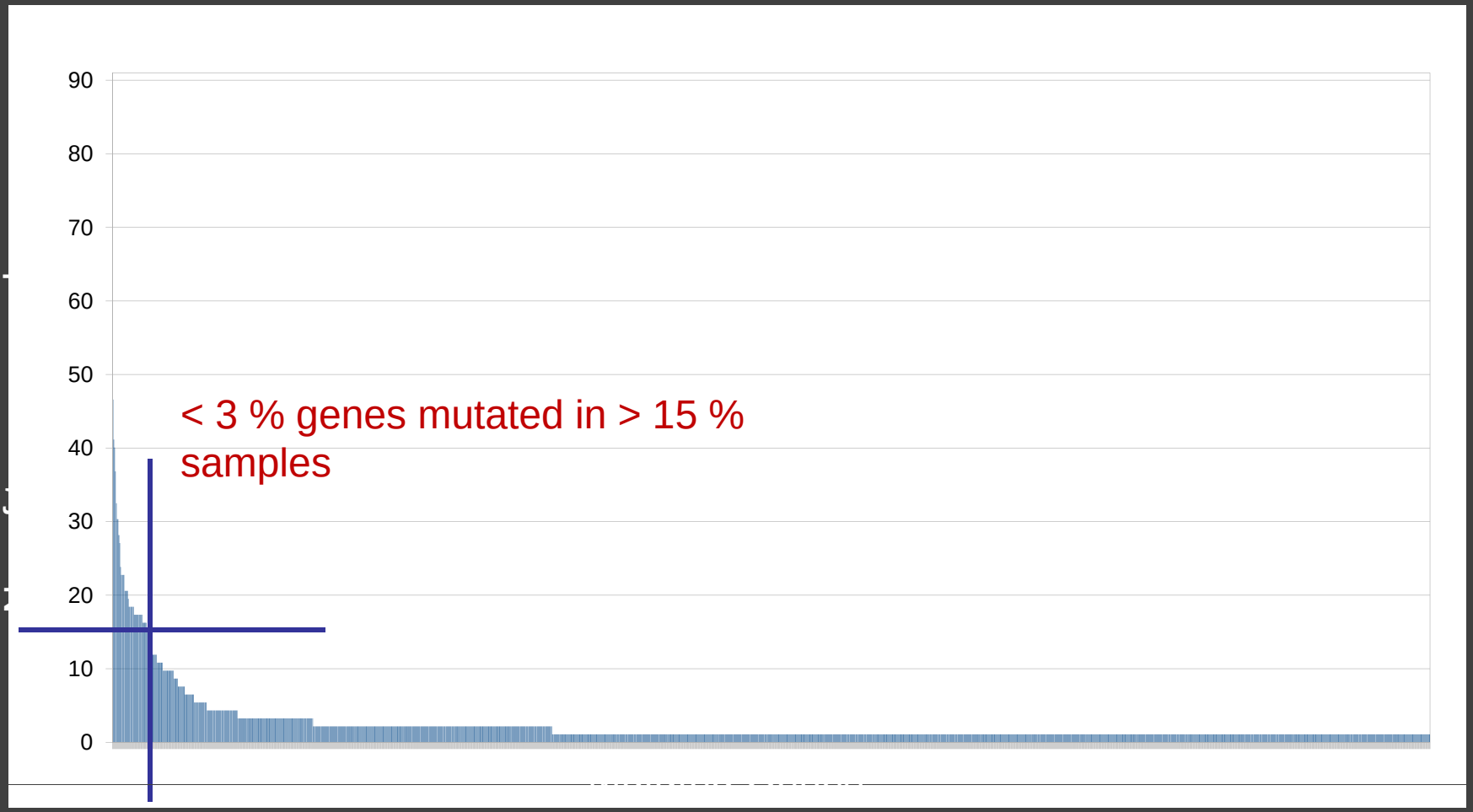
Genomics



Phenotype/Disease



Glioblastoma Multiforme (TCGA dataset)

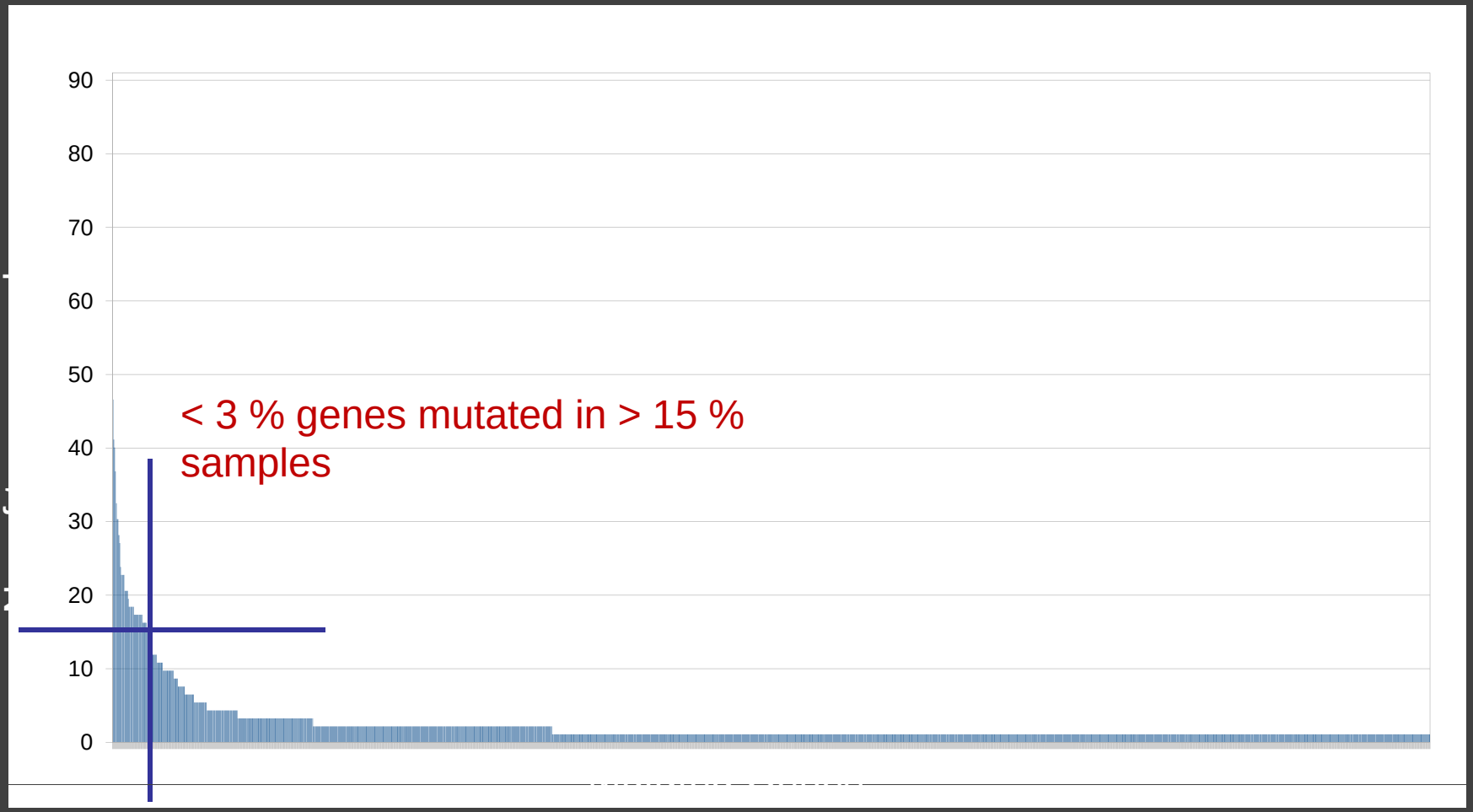


Comprehensive genomic characterization defines human glioblastoma genes and core pathways, The Cancer Genome Atlas Research Network, *Nature 455*, 1061-1068

Summary

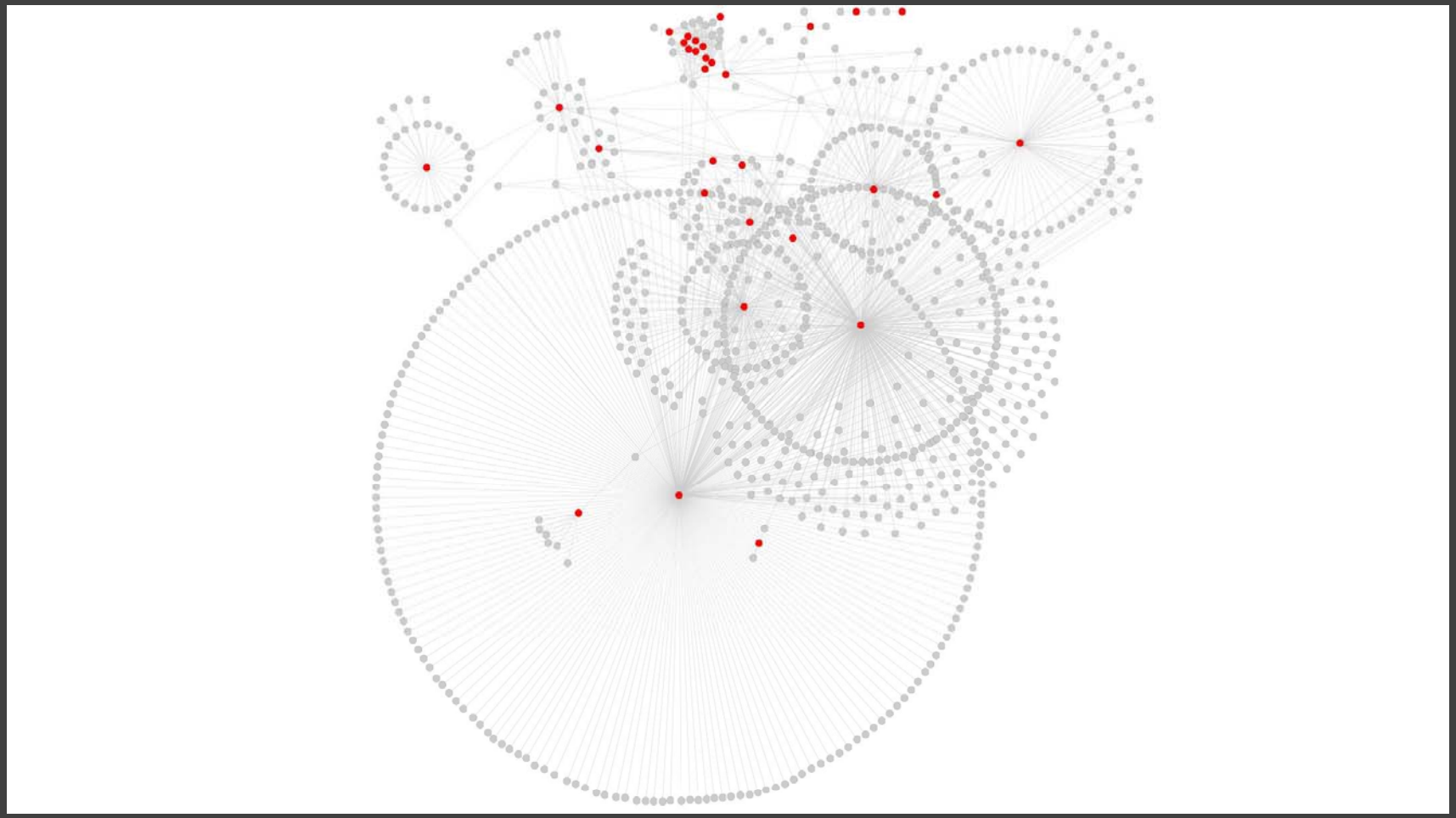
- Individual genome sequences differ by 1000's of positions
- Different genome sequences have indistinguishable phenotypes
- Most phenotypes are caused by the interaction of many molecules, mutations
- It has been difficult to relate (cancer) genotype to phenotype
- Genotype indicates THE POTENTIAL for a disease, as opposed to its ACUTE STATE

Glioblastoma Multiforme (TCGA dataset)



Comprehensive genomic characterization defines human glioblastoma genes and core pathways, The Cancer Genome Atlas Research Network, *Nature 455*, 1061-1068

Frequently Mutated Genes are Functionally Linked



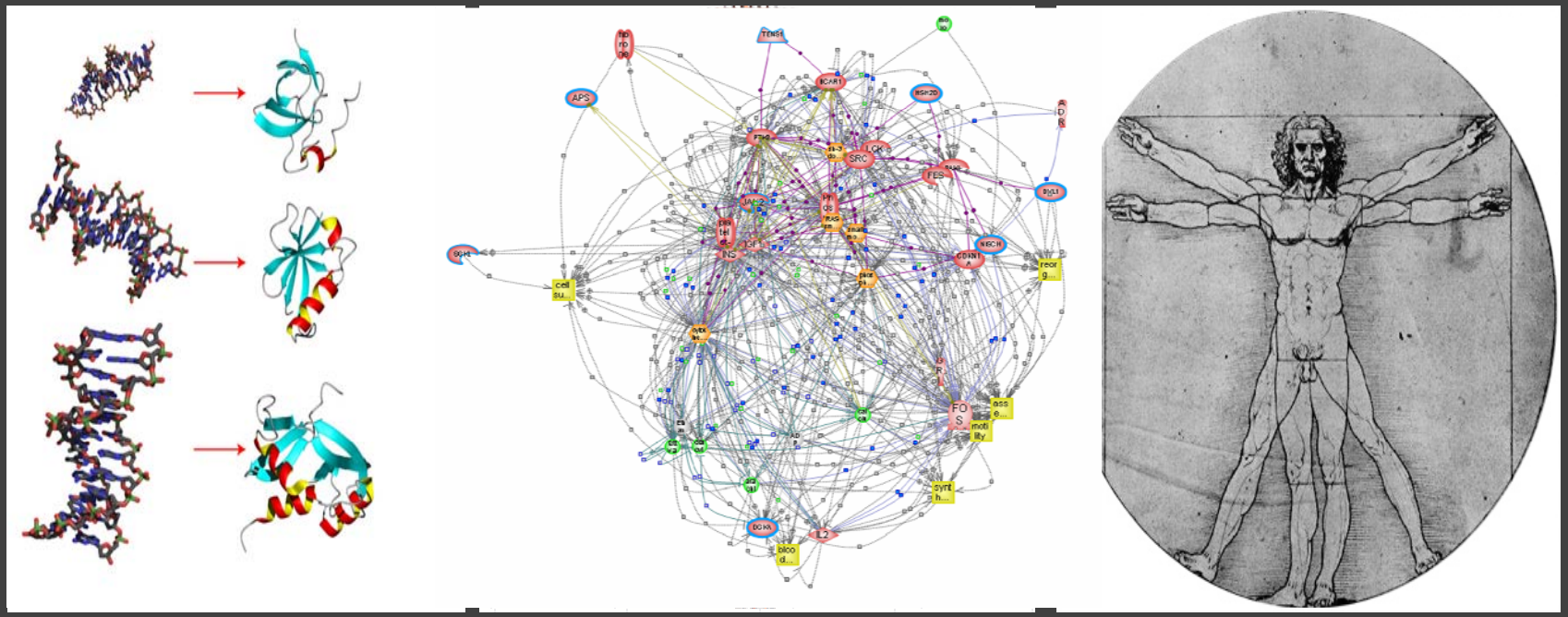
A human functional protein interaction network and its application to cancer data analysis, *Genome Biology* 2010, **11**:R53

Genotype --- Phenotype

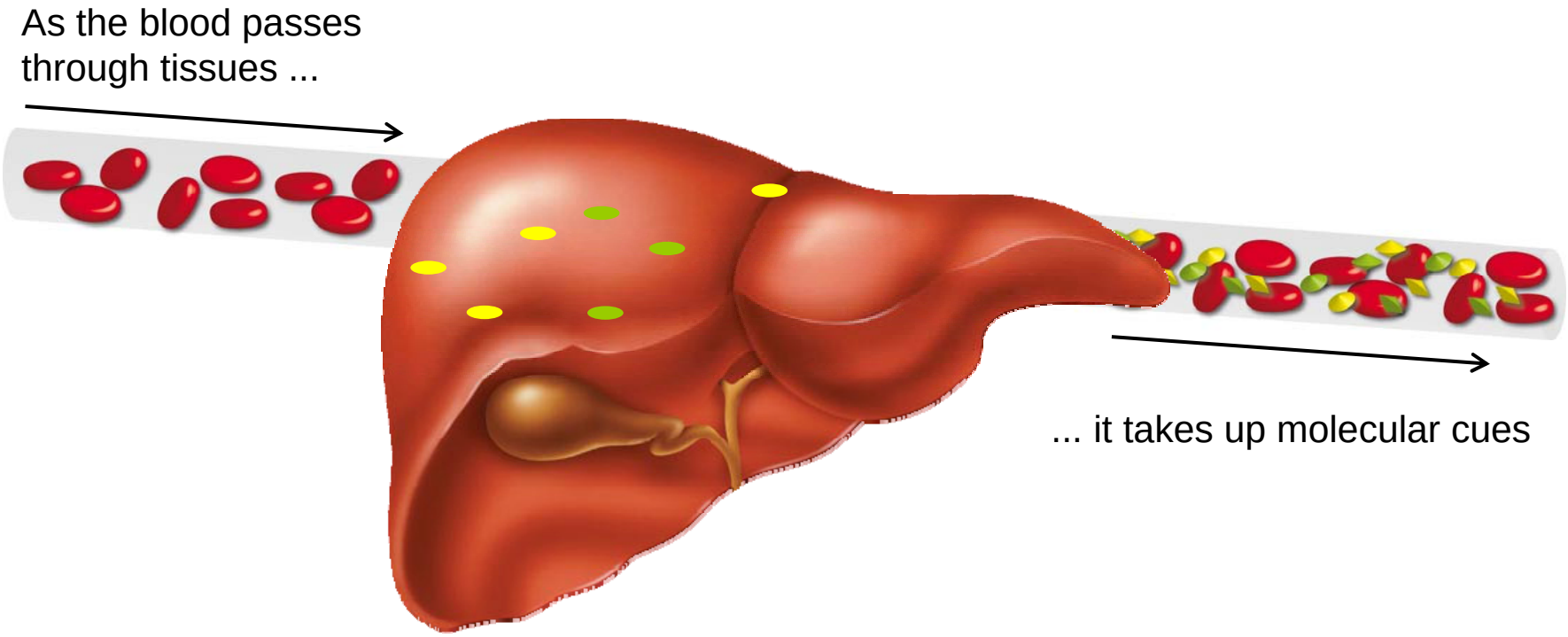
Genomics

Molecular Phenotype?network

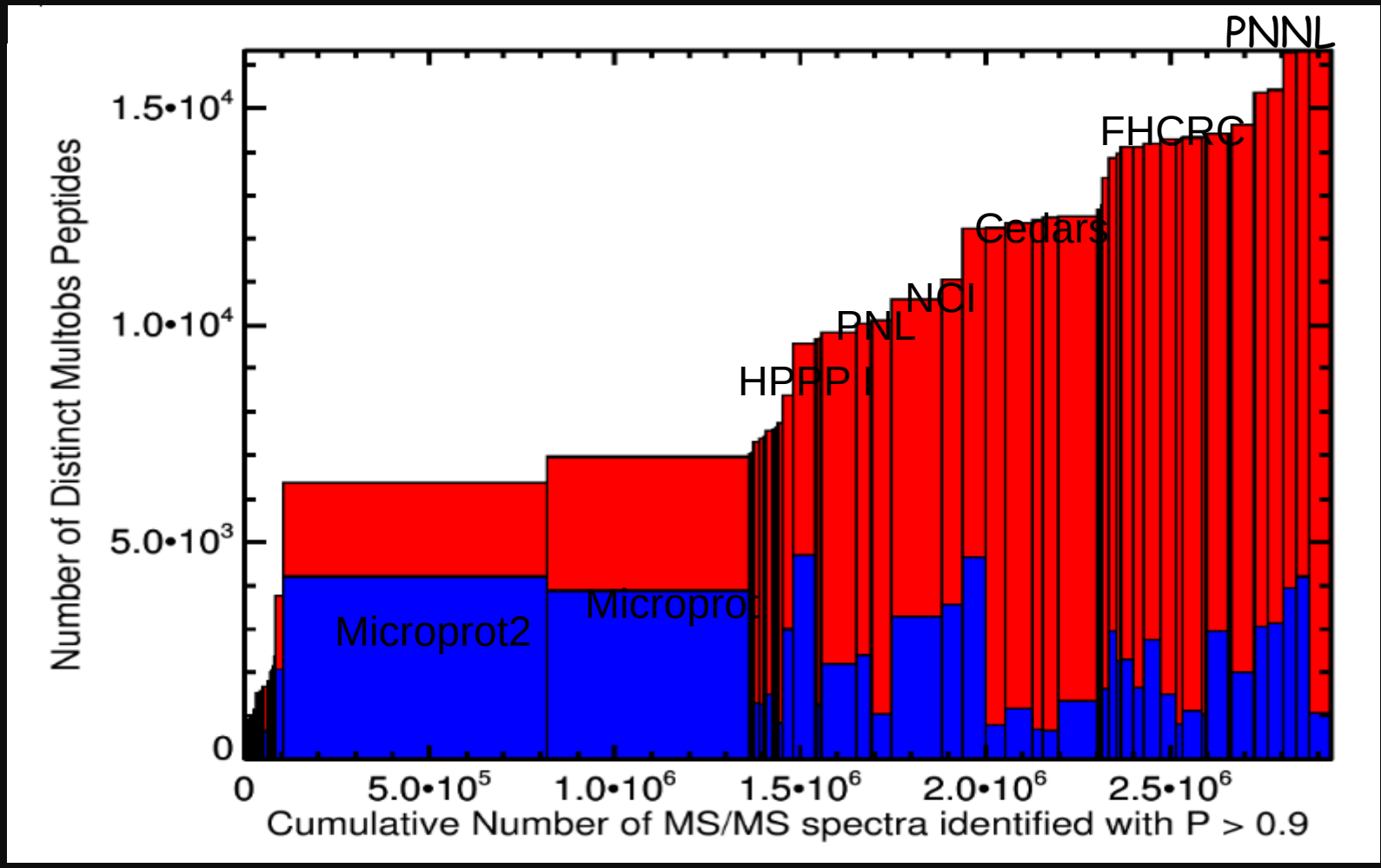
Phenotype/Disease



Remote sensing of cancer perturbed networks



Current State of Plasma Proteome Discovery



Cumulative data from leading groups:

>100 experiments

- 20 million measurements
- Millions of $$
- Years of time

2355 proteins, decoy-estimated FDR 1.0%

Deutsch E, Farrah , T, in press

Navigating with digital maps

Search map

complete digital map
e.g. google street view

Digital record of object
e.g. digital photo on iphone

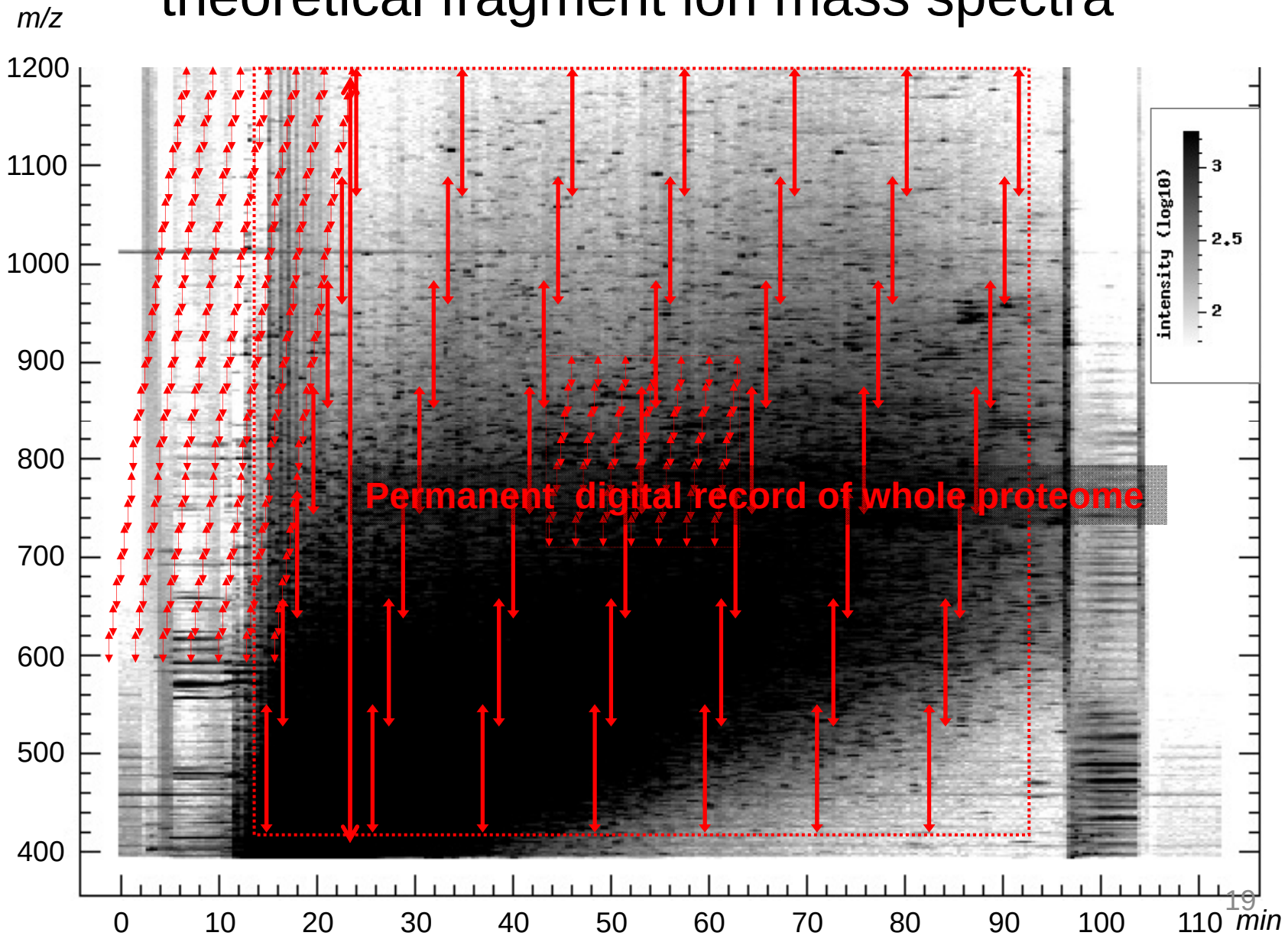
Principle: Identify and localize known feature in digital map

SWATH-MS: Sequential windowed acquisition of all theoretical fragment ion mass spectra

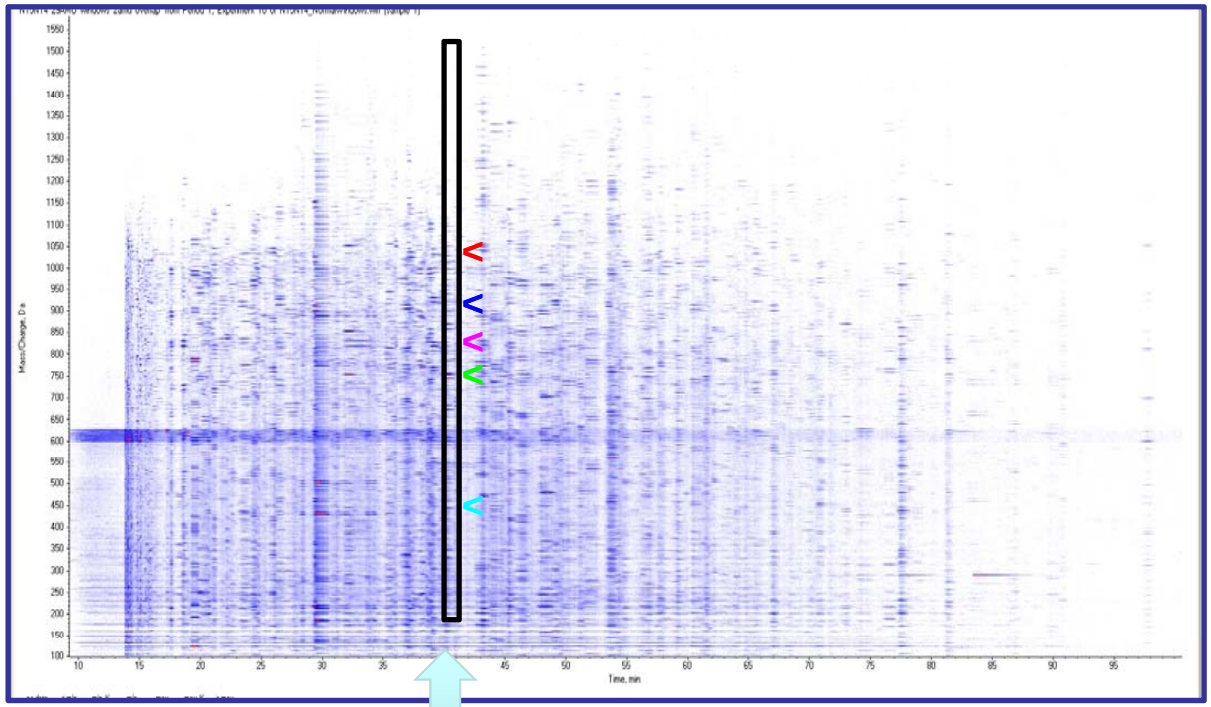
Satellite swath acquisition

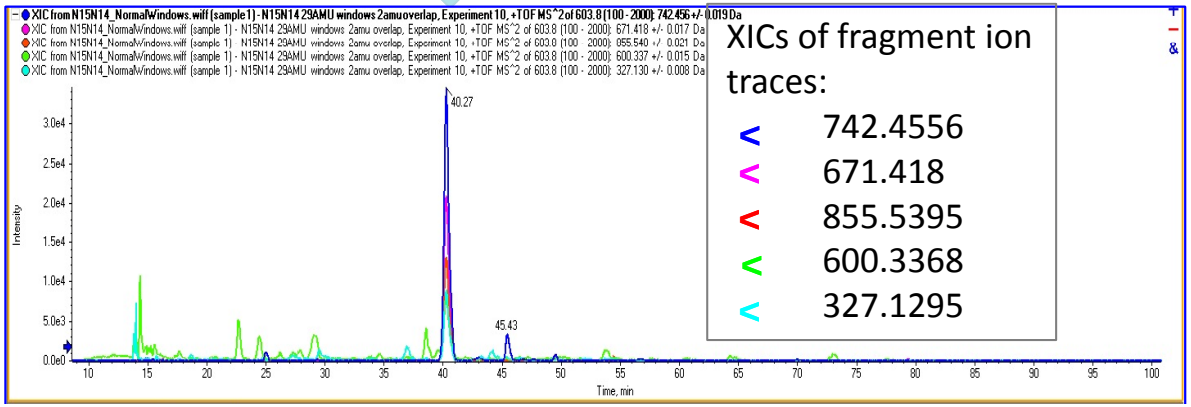
Permanent digital record
of all scanned objects

SWATH-MS: Sequential windowed acquisition of all theoretical fragment ion mass spectra



Targeted Analysis of SWATH-MS Data





XICs of fragment ion traces:

- 742.4556
- 671.418
- 855.5395
- 600.3368
- 327.1295

Digital record of specific protein (iPhone analogy

Spectral Library

Peptide #1 - ACDEFGK

Q1	Q3	RT
453	560	34min
453	678	34min
453	784	34min

Summary

• Genes mutated in cancer are linked in functional networks

•To determine the ACUTE STATE of a person we need to sense these networks remotely

•Measurement of target proteins in plasma is remote sensing

•SWATH mass spectrometry generates a searchable digital record for all plasma proteins

Hypothesis #1

Cancer Associated Proteins (CAP’s) are related to cancer driver mutations (CAN’s)

Study design

- Curate all cancer associated proteins from the literature (>1200 proteins)
- Generate definitive assays for their reproducible measurement (analog to iPhone pictures) in plasma protoemic datasets
- Relate the plasma detectable proteins to cancer genomic data

CANCER-ASSOCIATED PROTEINS (CAPS)

Differentially expressed in cancer

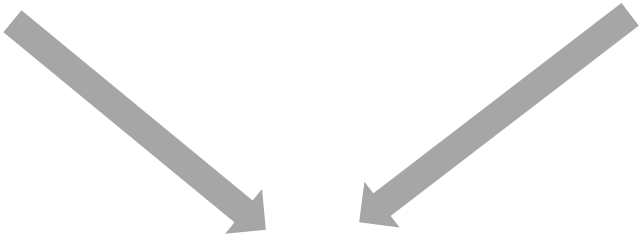
A List of Candidate Cancer Biomarkers for Targeted Proteomics

Malu Polanski and N. Leigh Anderson

FDA-approved assay

The Clinical Plasma Proteome: A Survey of Clinical Assays for Proteins in Plasma and Serum

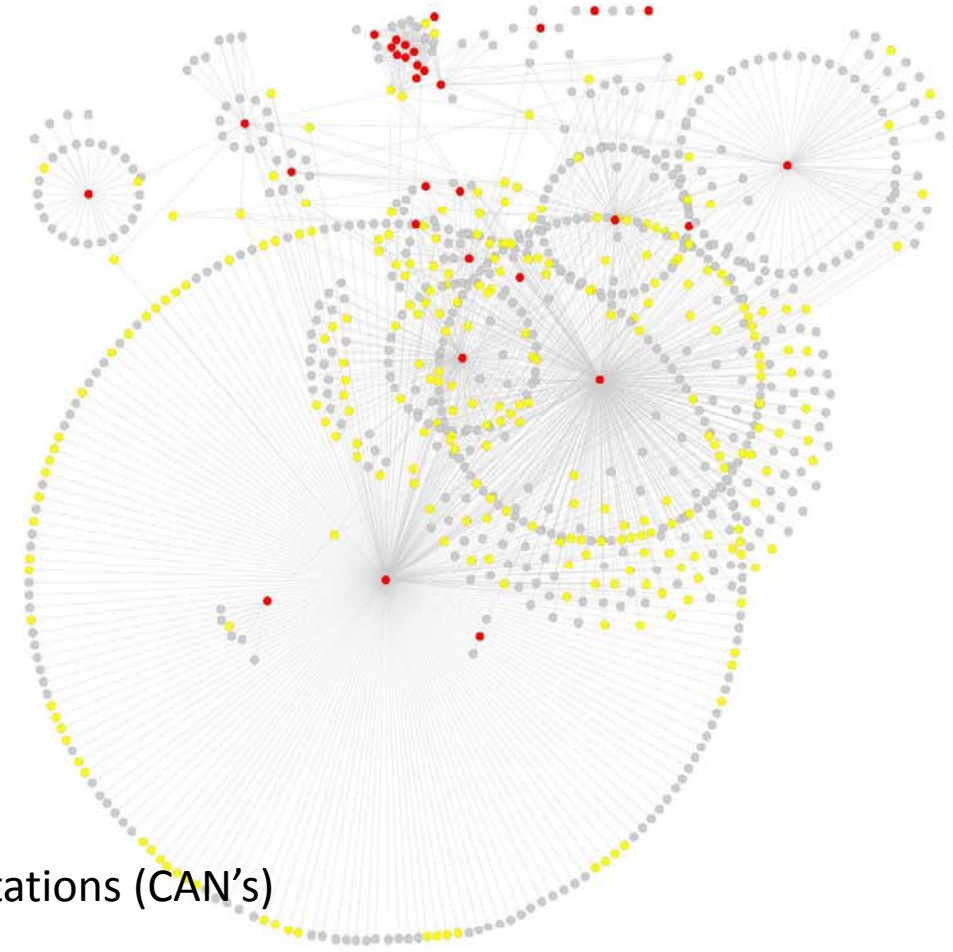
N. Leigh Anderson[1]



1172 target proteins

Polanski M *et al*. List of Candidate Cancer Biomarkers for Targeted Proteomics. *Biomarker Insights* (2006)
Anderson NL. The clinical plasma proteome: a survey of clinical assays for proteins in plasma and serum. *Clin Chem* (2010)

Cancer associated proteins and Cancer driver mutations are functionally linked



Red: Cancer driver mutations (CAN's)
Yellow: CAP's
Grey: CAN functional interactors

NETWORK ANALYSIS

Functional relationship of CAPs to candidate cancer driver mutations (CANs) discovered by whole exome sequencing:

- Creating a subnetwork of 379 CANs for 7 cancer types and their interaction partners in the Reactome functional interaction network
- Examining the subnetwork for the presence of CAPs

Cancer-associated proteins (CAP)		Overlap of CANs with CAPs	# of CAPs in the sub-network of CANs	p-value (Enrichment of CAPs in subnetwork of CANs)	# of CANs that can be monitored by CAPs
All	1172	43	612	$< 1e^{-17}$	136
Detectable	473	18	242	$4e^{-10}$	125

$\Rightarrow$ Subnetwork of CANs is significantly enriched for cancer-associated proteins

Wu *et al*. A human functional protein interaction network and its application to cancer data analysis. *Genome Biol* (2010)

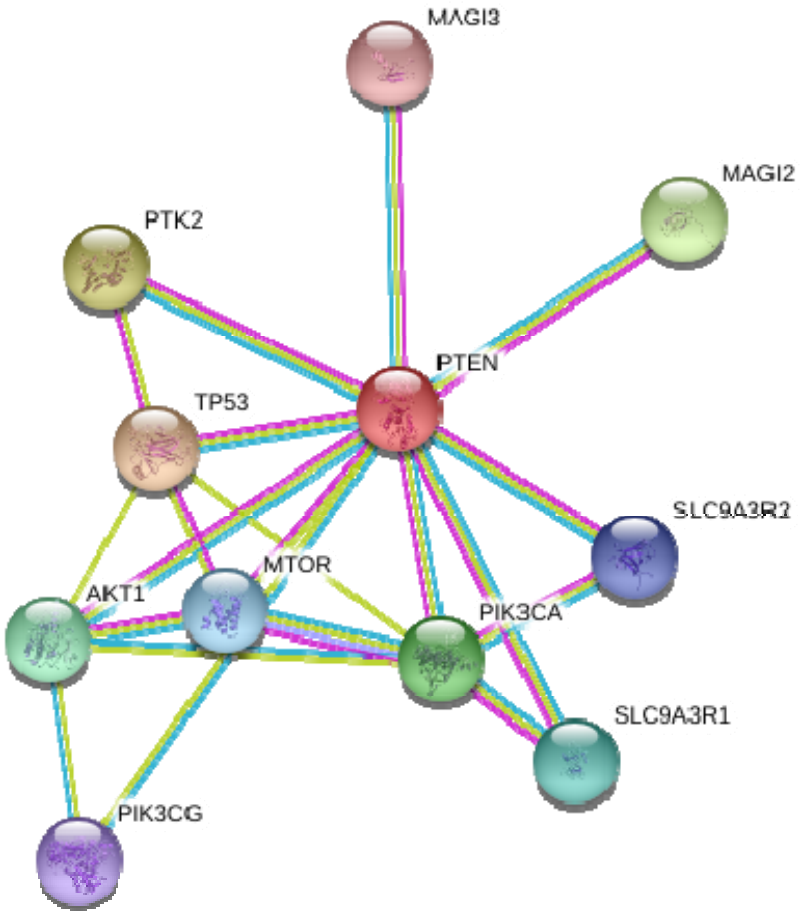
Hypothesis #2

Cancer associated cellular networks perturbed in cancer tissue are detectable in plasma

Study design

- Perturb a cancer associated network by genetic engineering in animal model
- Measure perturbed network in affected tissue
- Measure network associated proteins in plasma

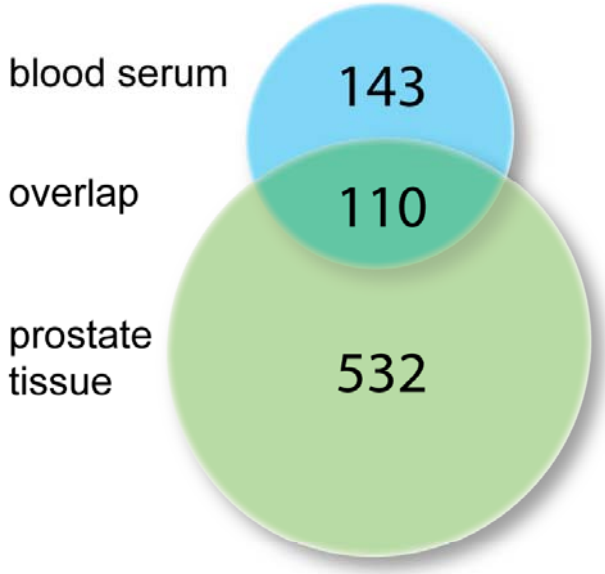
PTEN/TOR signaling network

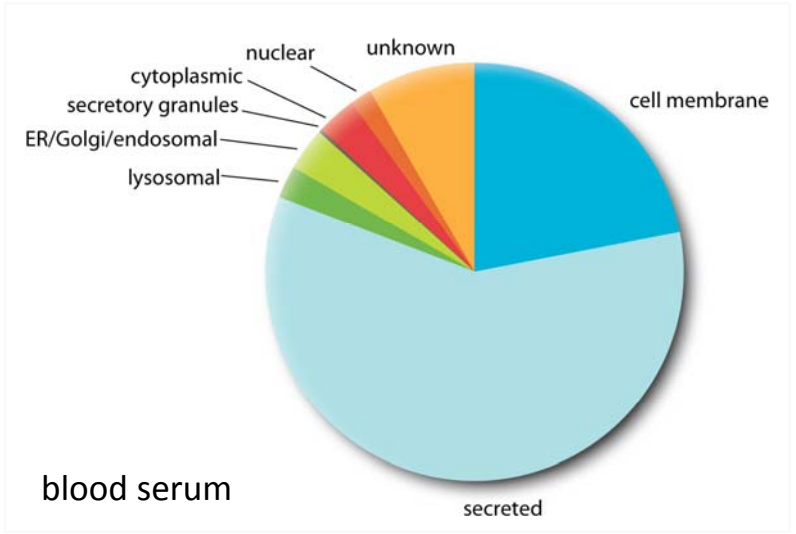


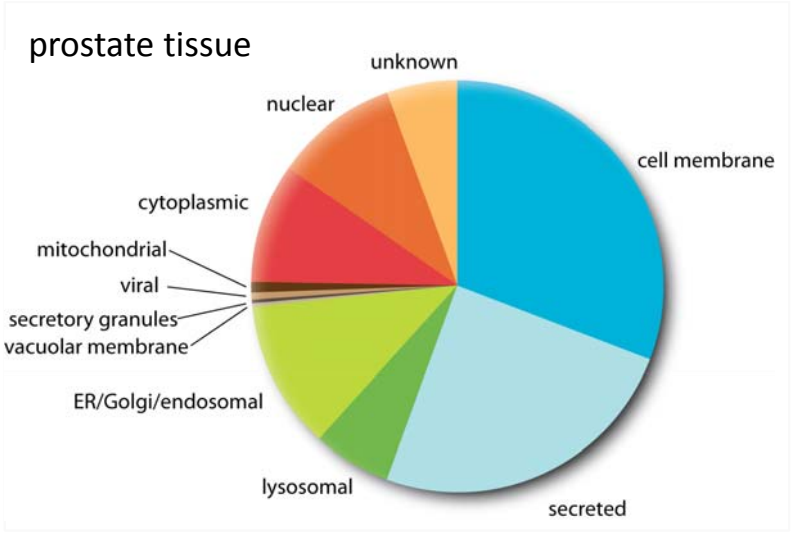
- PTEN: Tumor suppressor protein
- TOR: target of rapamycin
- PTEN mutated in many cancers

Identification of Glycoproteins from Mice

- 785 glycoproteins identified from mouse tissue and serum

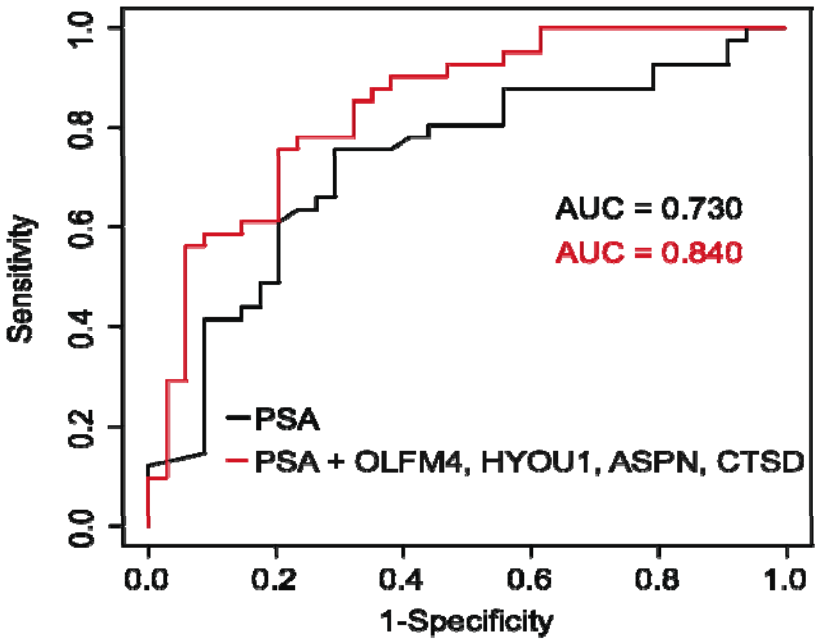




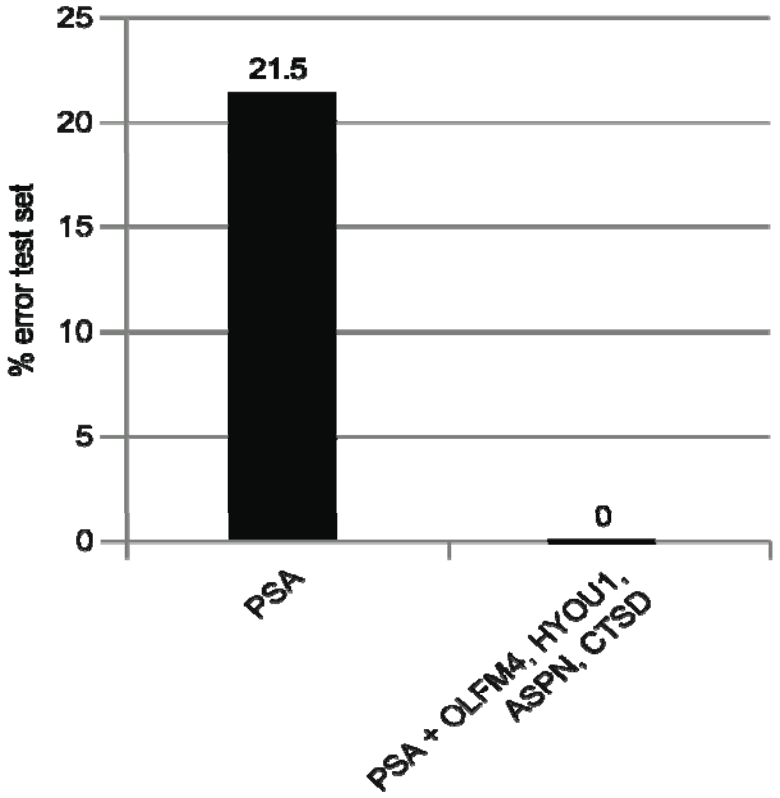


Plasma panel 3: More Specific Diagnosis of Prostate Cancer

(BPH, n=35; locPCa, n=41)



(BPH, n=6; locPCa, n=8)



PSA: sens. 89%, **spec. 32%**

PSA + Signature: sens. 90%, **spec. 83%**

Cima , Schiess, et al, PNAS, 2010

Hypothesis #3

The measurement of cancer associated cellular networks in plasma can stratify patients, e.g. responders vs. non responders to drugs

One Size does not fit all

DIABETES DRUGS	43%	
ARTHRITIS DRUGS	50%	
ALZHEIMER DRUGS	70%	
CANCER DRUGS	**75%**	

do not respond

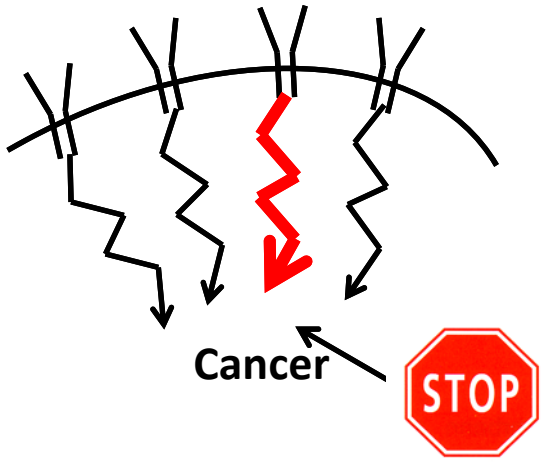
→ 50% of cancer drugs terminated in Phase II

→ Lack of efficacy

Patient Selection

to Maximize the Likelihood of Response to Drugs

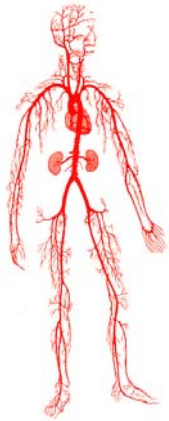
Many possibilities for cancer



Who will respond?

Everolimus
(Novartis)

Test patients for response



12 responders

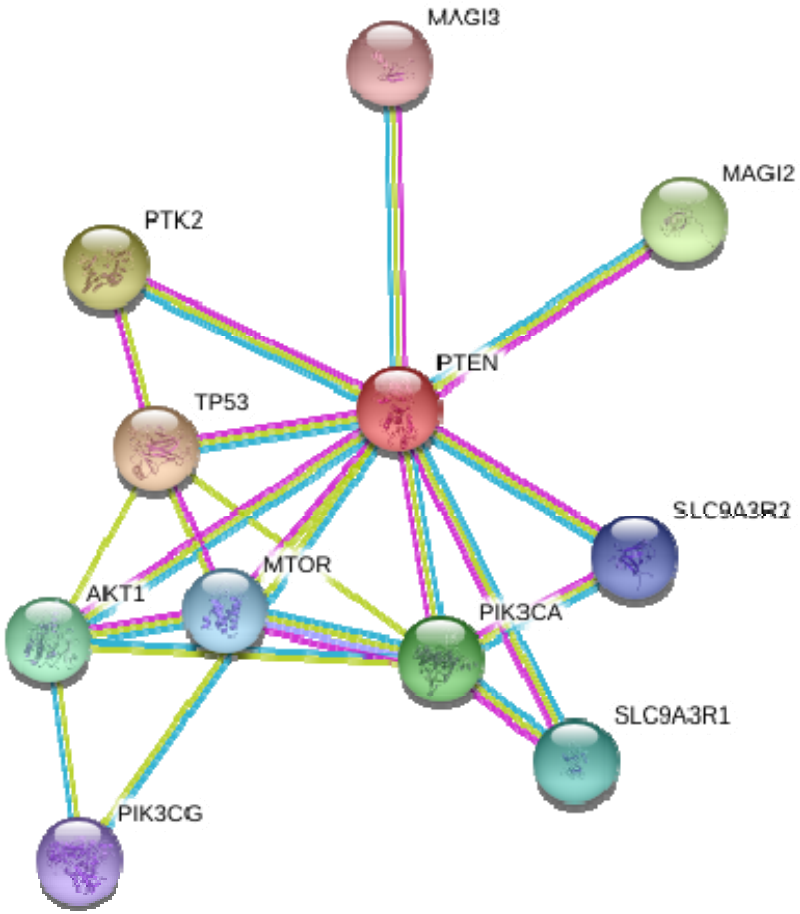
IP protected

Acc. 85%
NPV 90%

16 non responders

Templeton A., et al. Presentation at ASCO 2011, J Clin Oncol, 2011. 29.

PTEN/TOR signaling network



- PTEN: Tumor suppressor protein
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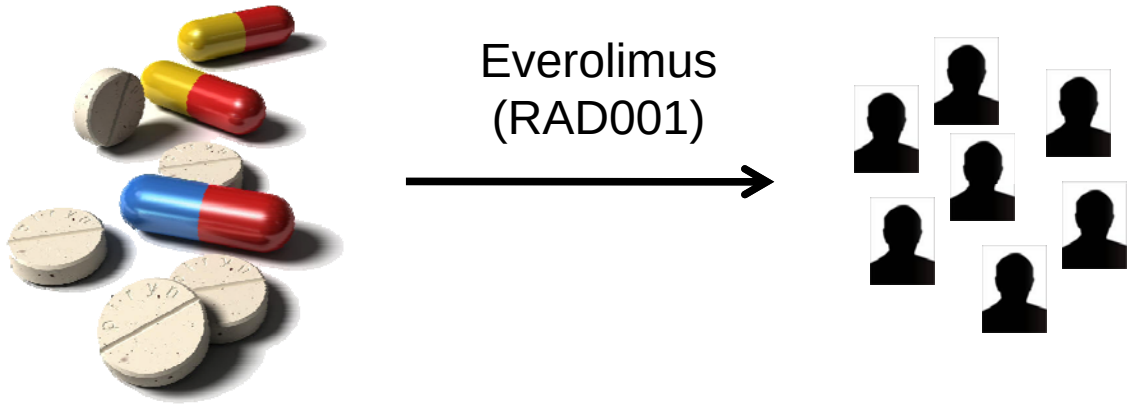
Optimal Protein Panel

Logistic regression:

TP = 15	FP = 3
FN = 1	TN = 8
NPV	89%
PPV	83%
sens	94%
spec	73%
acc	85%

Matching patients with the right therapy



Swissmedic approved multicenter phase II clinical trial using Everolimus as a first-line therapy in non-rapidly progressive castration resistant prostate cancer

Take Home Message

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- The genomic information, modulated by external factors, determines the phenotype (disease).

- Genomic analyses need to be complements by measurements of the expressed information (proteins) that indicate the ACUTE STATE of a person

- Measurements to “remotely” measure the state of a tissue are becoming a reality

Outlook

• We will have available soon 1000's of genome sequences

•We have available the ability to identify every human protein by mass spectrometry

• We envisage the following process:
    • Genome sequencing
    •Inference of perturbed networks
    •Inference of potential plasma markers indicating perturbed network
    •Quantification of markers in plasma samples to obtain acute picture

•We envisage that every person will have their genome sequenced for risk assessment

•We envisage that every person will have their plasma protein map recorded periodically to test the acute state

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