Towards a Metabolomics Platform for Cardiovascular Disease

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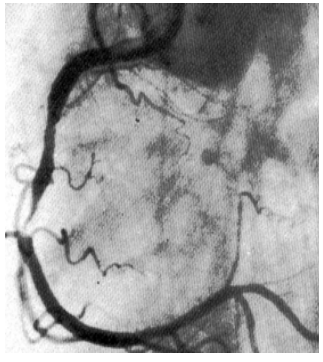
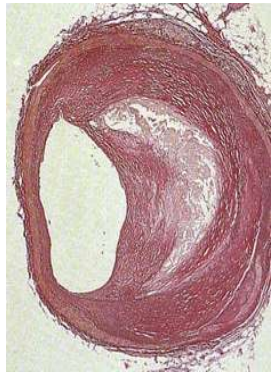
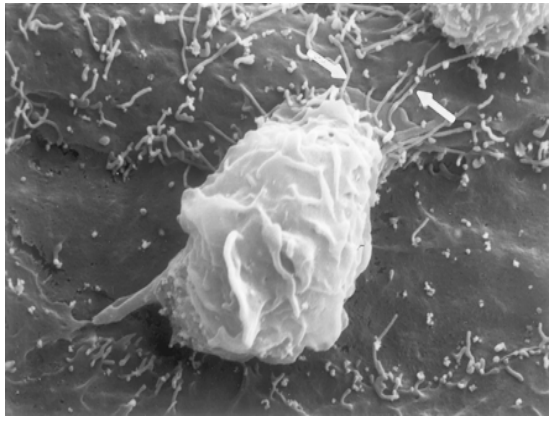
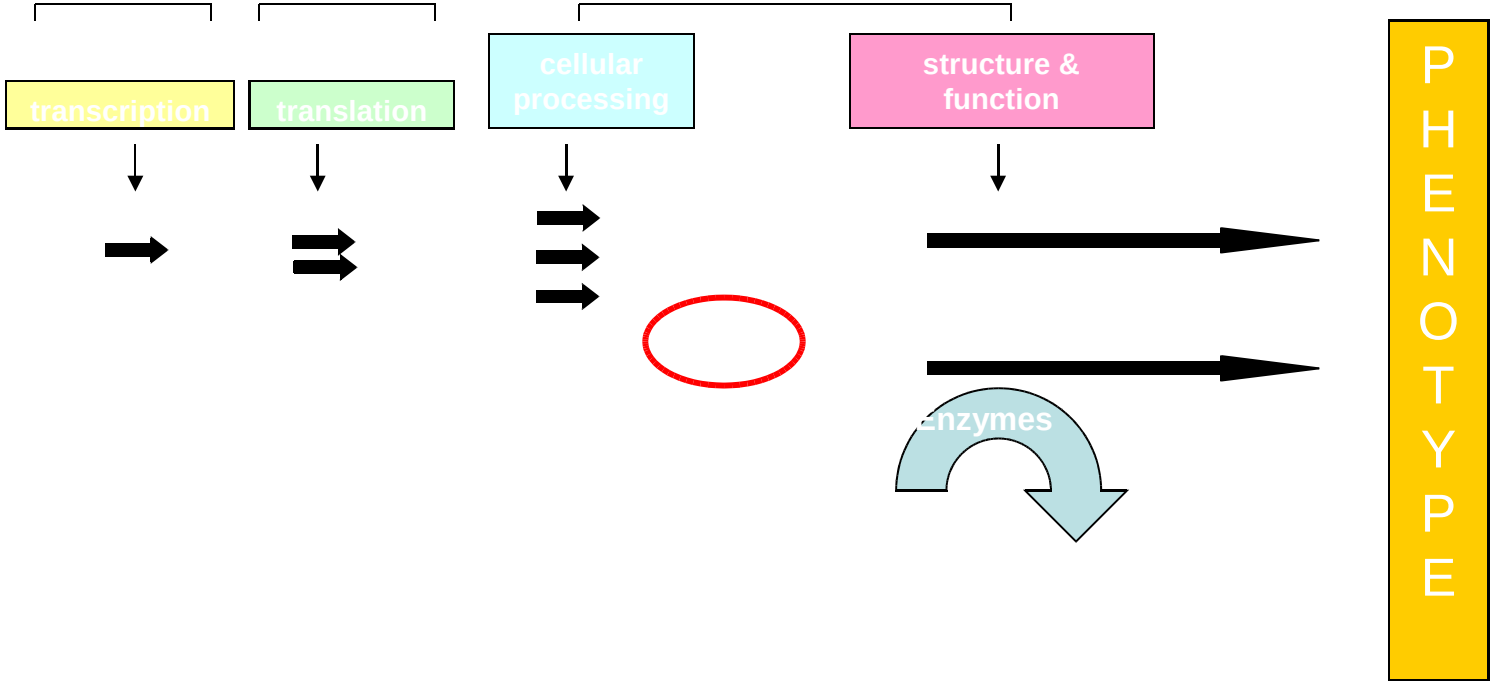
Inflammation & Wound Healing in Cardiovascular Disease

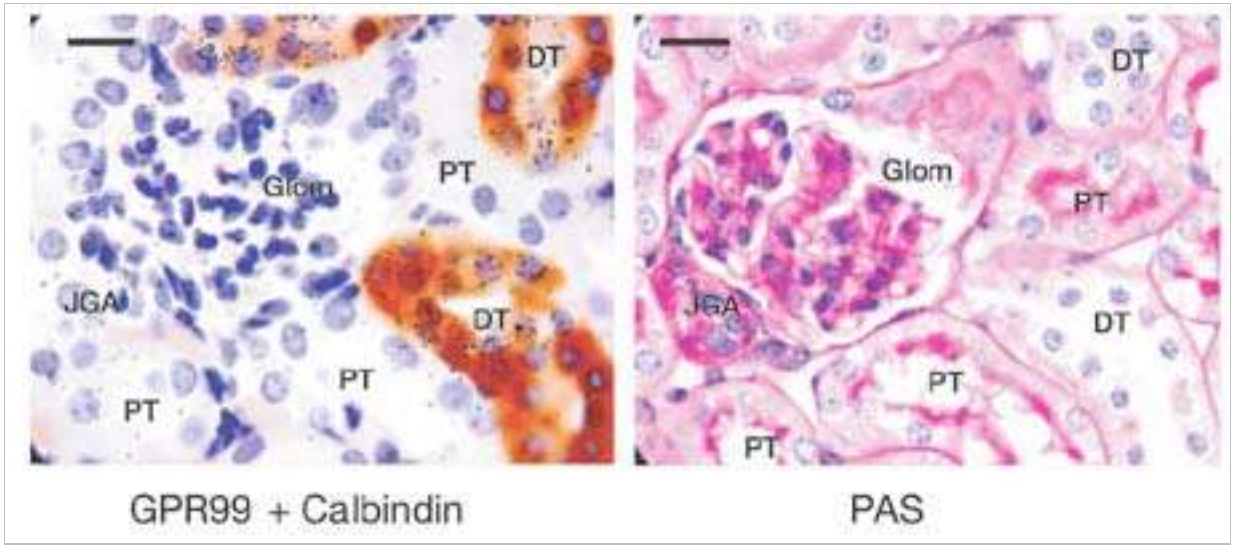







transcription
translation
cellular processing
structure & function
enzymes
PHENOTYPE

Unanticipated Role of “Intracellular” Metabolites

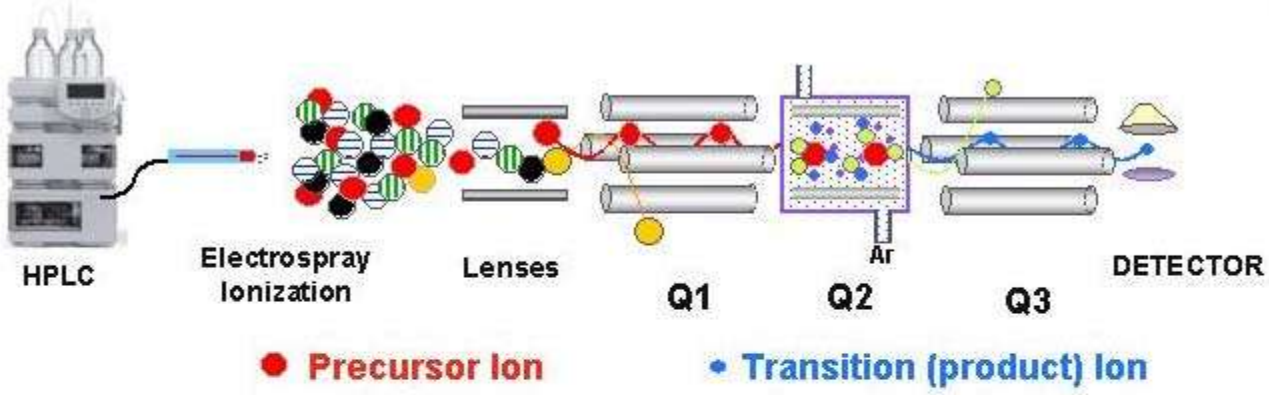


GPR 99 highly expressed in the renal cortex
Postulated to play a role in BP control
Ligand unknown

He et al. *Nature* 2004

Metabolomics Platform

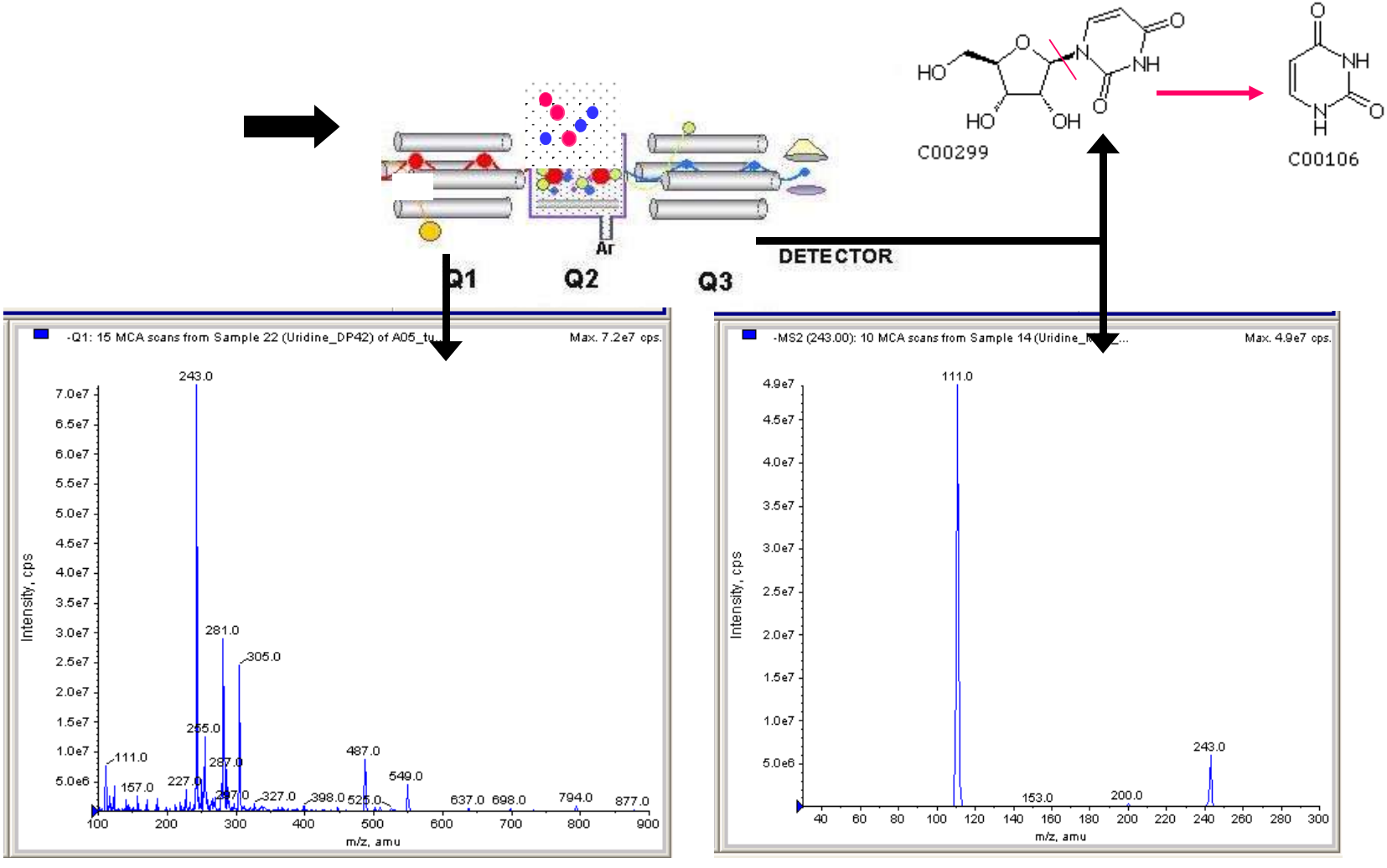
Organic acids, amino acids, nucleotides, carbohydrates, lipids



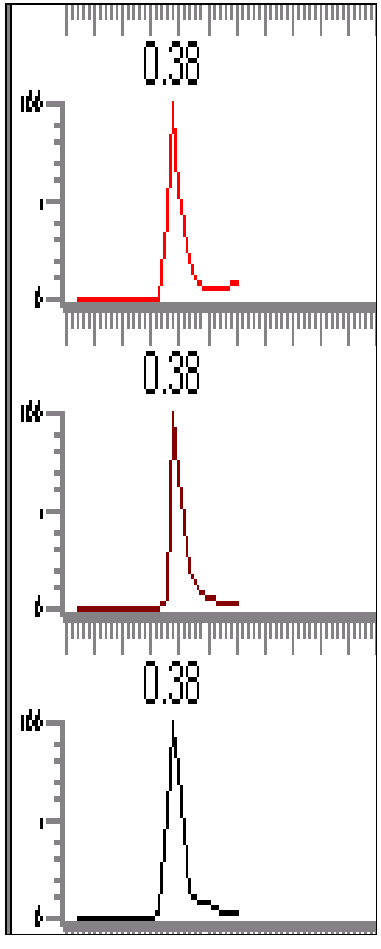
- **Q1 selects and passes only precursor of interest**
- **Q2 selected precursor is fragmented**
- **Q3: monitor specific fragment ions**

- **Sensitivity**
- **Specificity**
- **Throughput**
- **Small sample volumes**
- Absolute Quantitation
- Flexibility/Multiplexing
- Utility across species

Mass spec optimization: Tuning



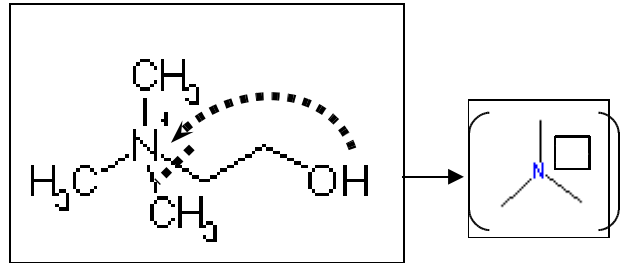
Specificity of the Mass Spectrometer



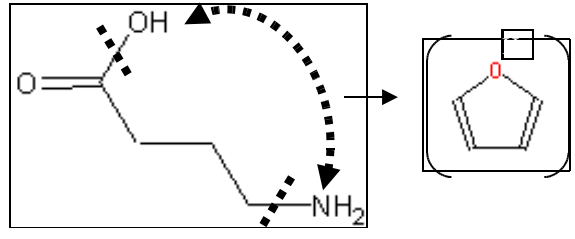
Reverse phase

Parent Daughter

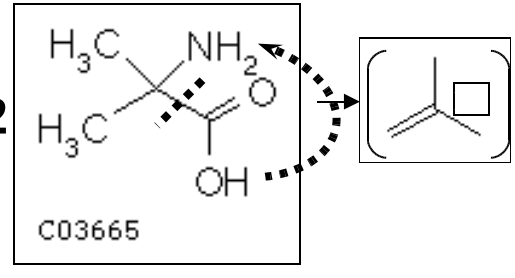
Choline: 104 -----→60.1



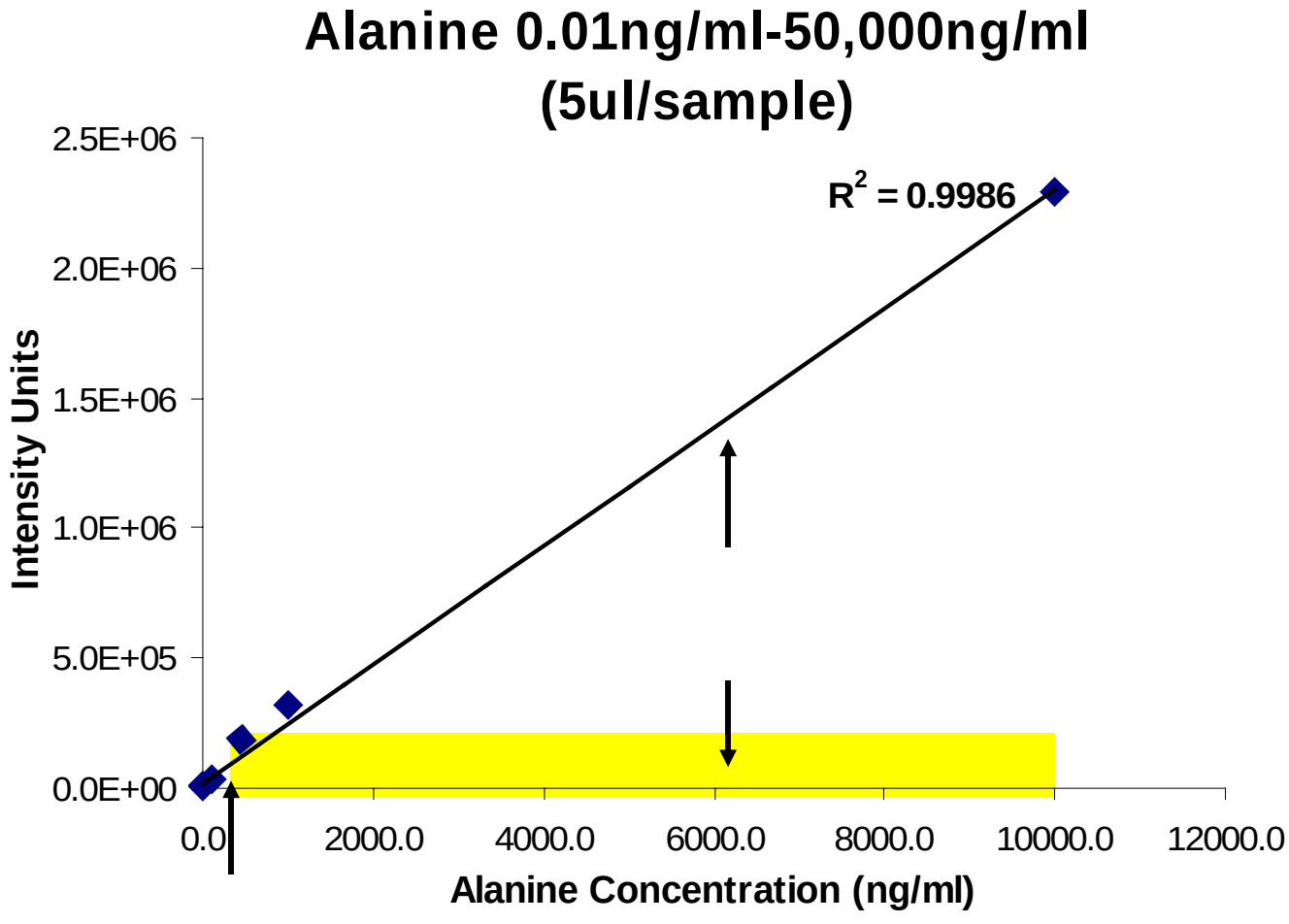
GABA: 104 -----→69.0



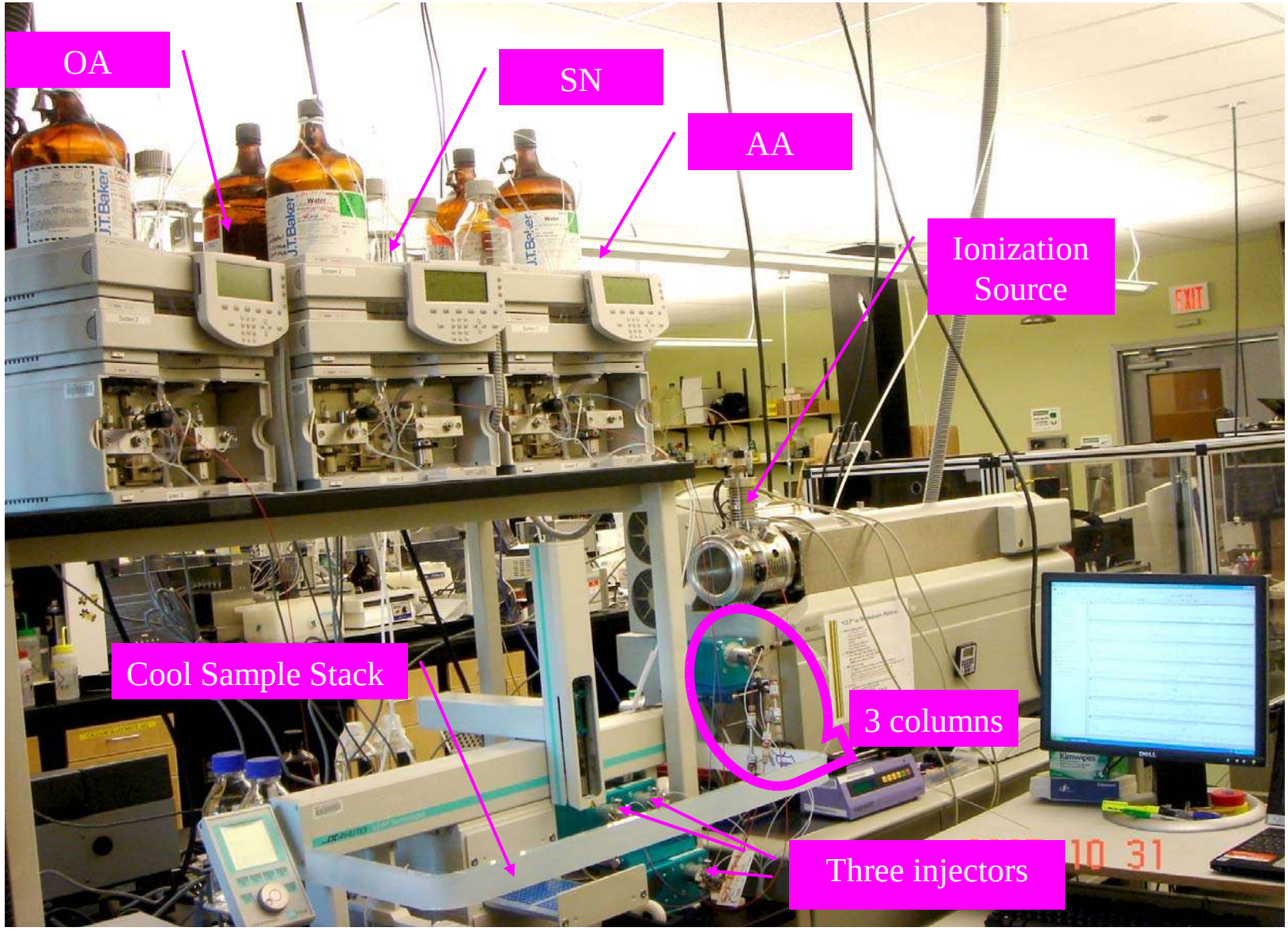
Aminoisobutyric acid : 104 -----→57.2



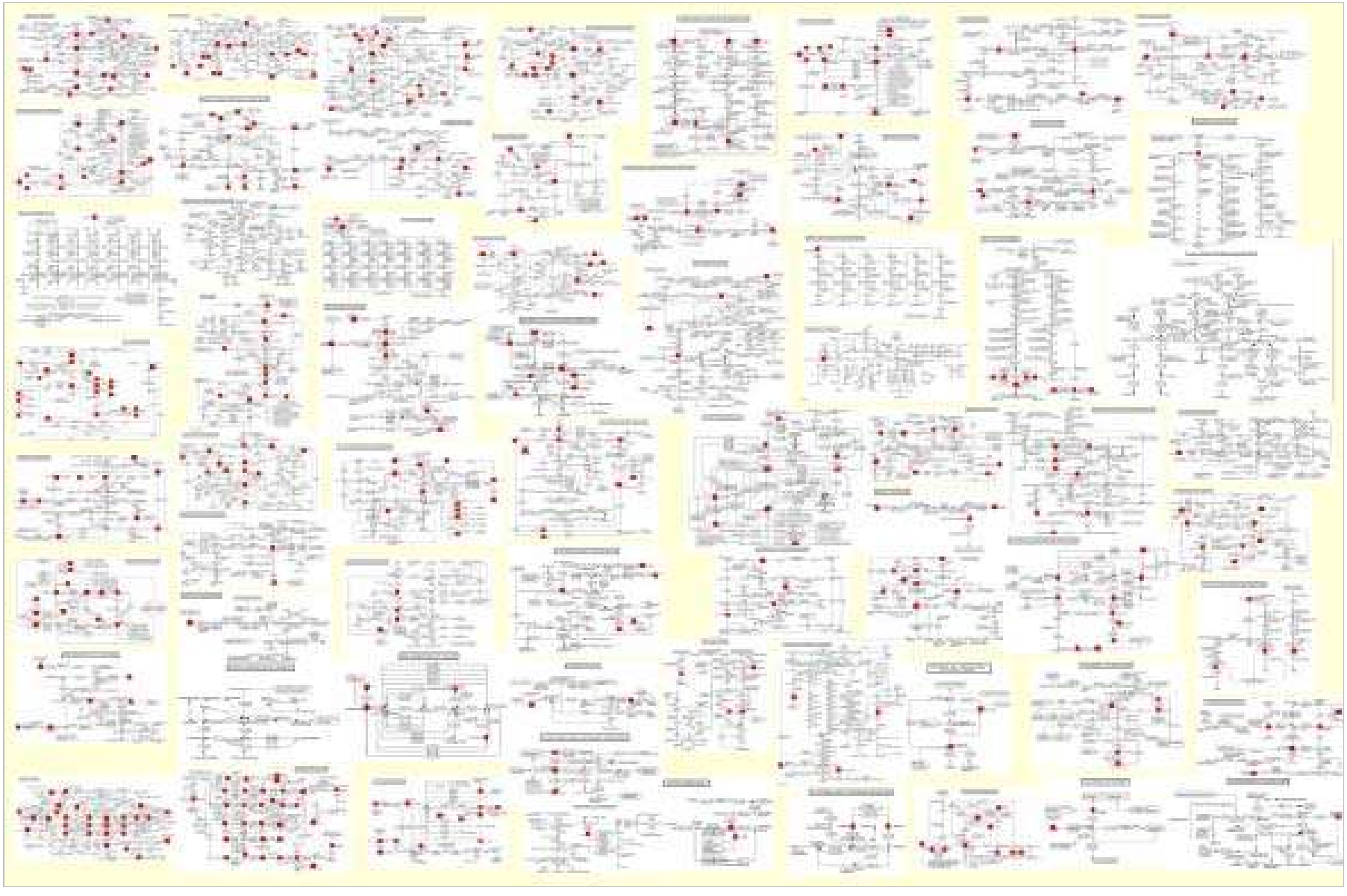
Sensitivity: Limit of Detection and Linear Range



Metabolomics Platform



• Platform Coverage



KEGG database of all known metabolic pathways

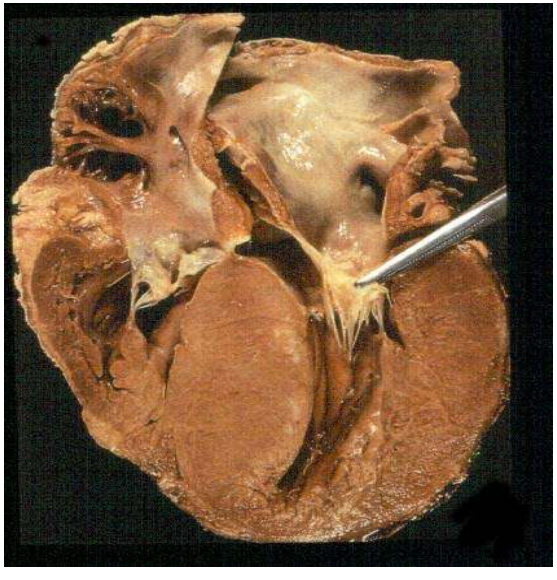
Progress in Platform Development

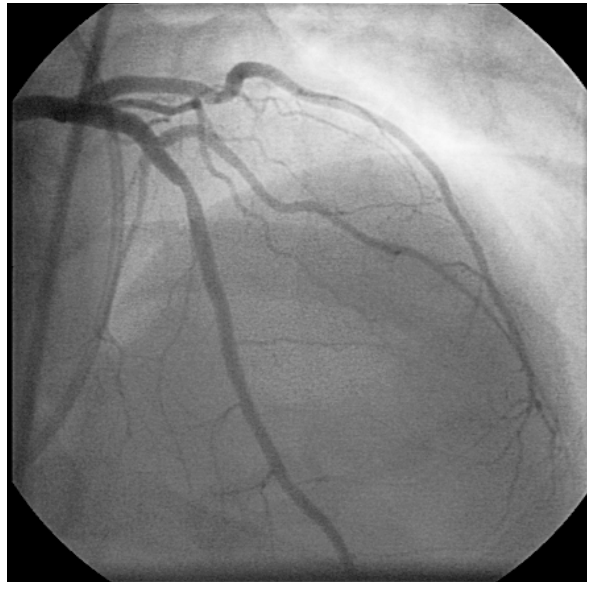
- Sensitivity
  - ng/ml range
- Reproducibility
  - CV <20%
- Comprehensive coverage
  - Approaching 500 metabolites

Challenges in Applying Metabolomics to Human Disease

- **Inherent unpredictability of the onset of pathological states**
- **Inter-individual variability**

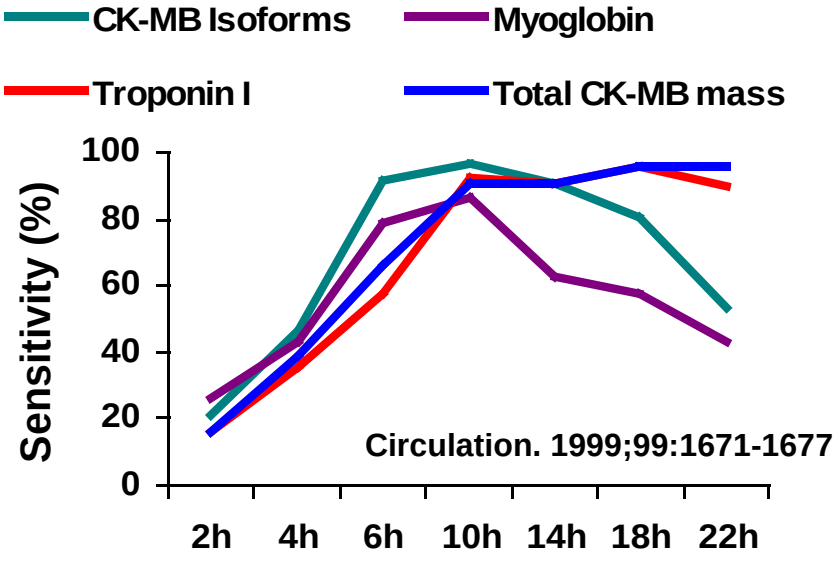
“Planned” Myocardial Injury





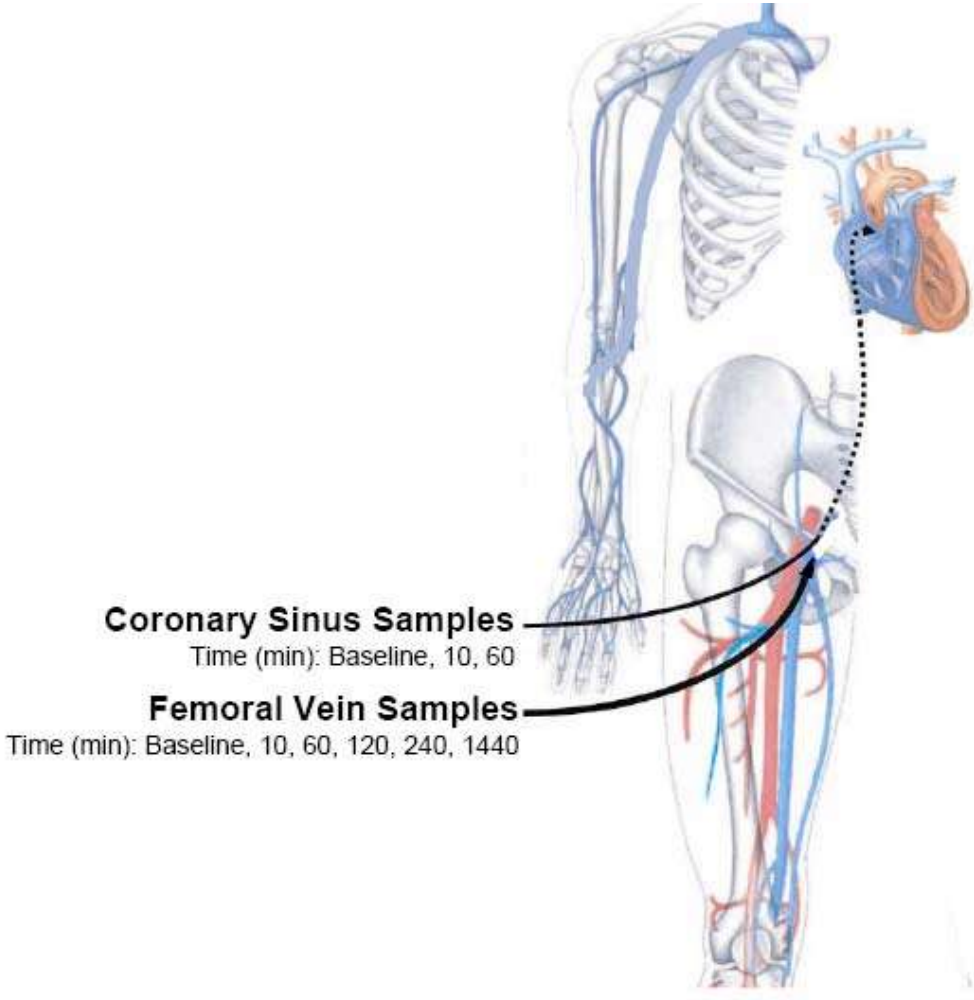
Current Blood Biomarkers

- None suitable for myocardial *ischemia*
- Existing biomarkers of myocardial *necrosis* (e.g., troponin) are not elevated until many hours after myocardial injury



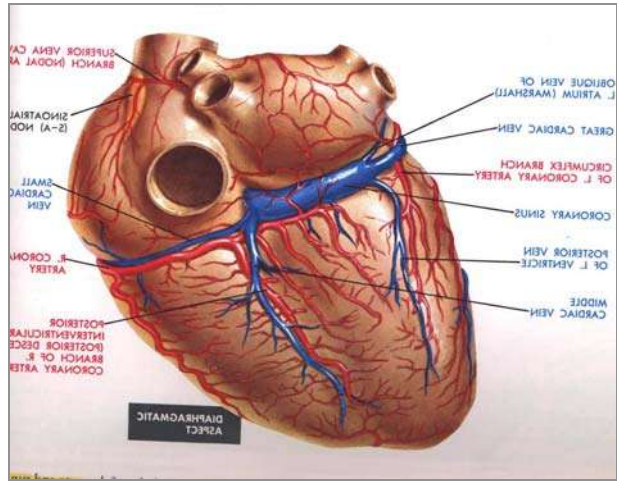
- Search for new biomarkers has been limited to proteins and constrained by a candidate marker approach

Blood Sampling during “Planned MI”

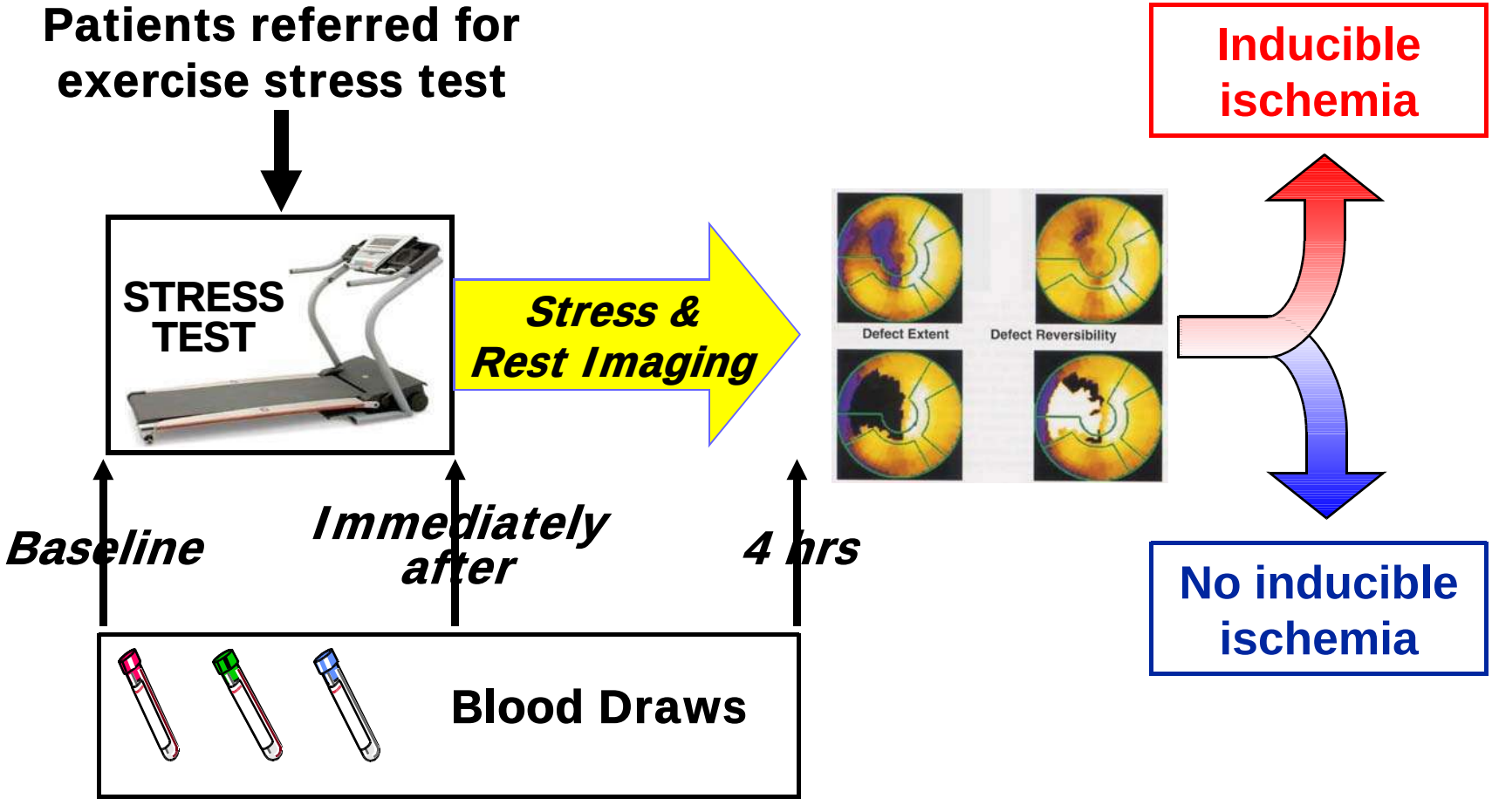


Coronary Sinus Samples
Time (min): Baseline, 10, 60

Femoral Vein Samples
Time (min): Baseline, 10, 60, 120, 240, 1440



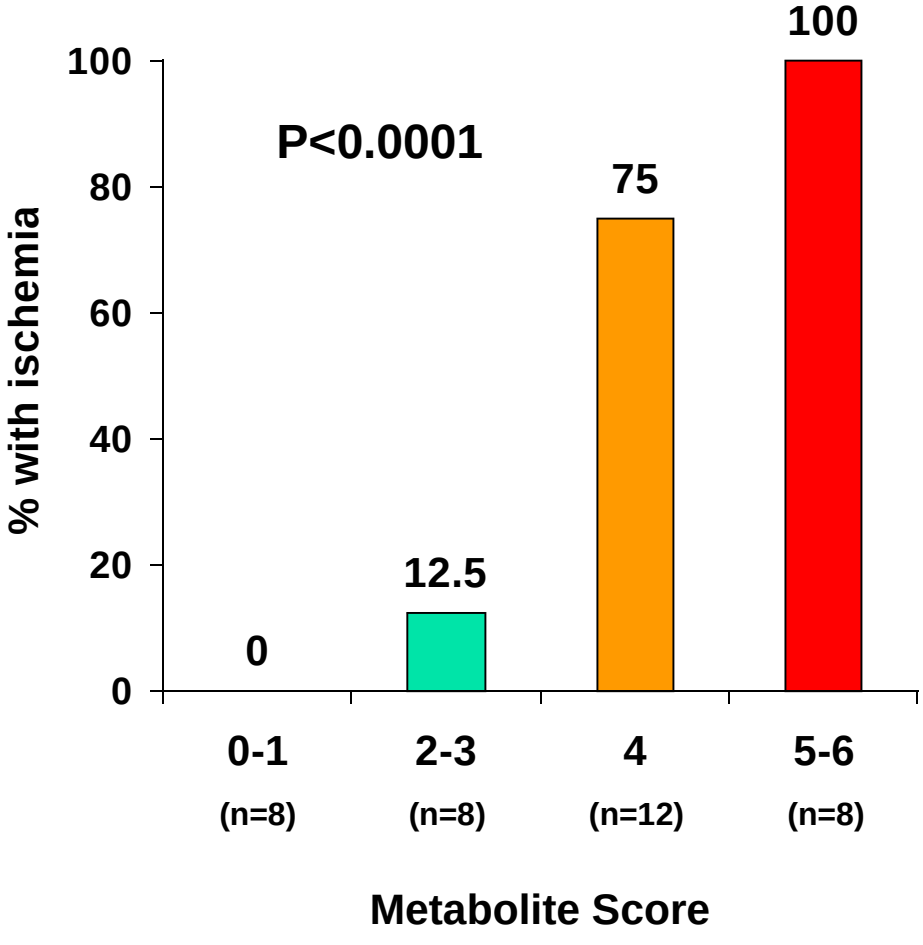
Exercise Stress Testing Protocol

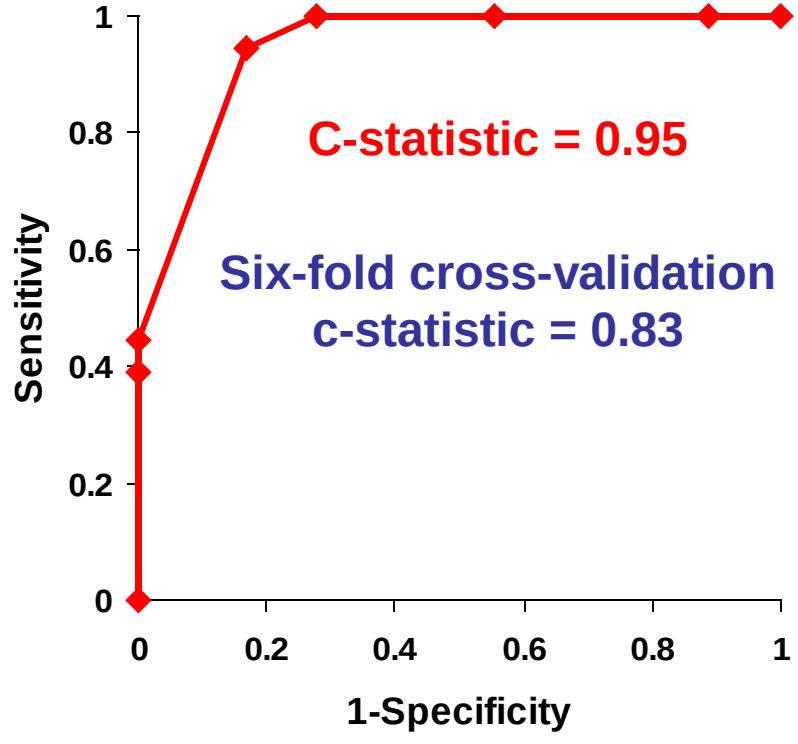


Each patient his or her own control
Critical advantage in data analysis

Sabatine et al.
Circulation 2005;112:3868-75.

Predictive Ability of Metabolite Score





Sabatine et al.
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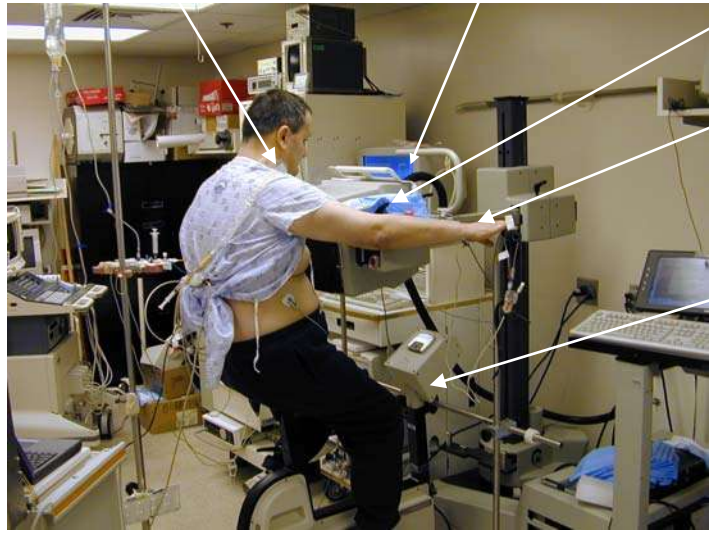
Studies of Extreme Exercise: Boston Marathon

Blood Draws

Maximum Exercise Testing in Normal Individuals

	ETT Cohort (N=26)	CPET Cohort (N=13)
Age, years	51 ± 11	52 ± 12
Male sex, (%)	62	60
Weight, (lb)	189 ± 40	184 ± 48
Body Mass Index	29 ± 5	27 ± 5
Exercise Duration (min)	12 ± 2.7	8 ± 0.8
Peak VO_2 (METs)	12 ± 4	9 ± 2



Baird Radionuclide Camera

Arterial Line

Upright Bicycle Ergometry
-continuous ramp protocol
-work increased at 12.5 Watts/min

A cautionary tale….

Rapid and noninvasive diagnosis of the presence and severity of coronary heart disease using ^{1}H-NMR-based metabonomics

Joanne T. Brindle1, Henrik Antti1, Elaine Holmes1, George Tranter1, Jeremy K. Nicholson1, Hugh W.L. Bethell2, Sarah Clarke2, Peter M. Schofield2, Elaine McKilligin3, David E. Mosedale4 & David J. Grainger4

Published online: 25 November 2002; doi:10.1038/nm1202-802

nature medicine

TECHNICAL REPORTS

Proton NMR analysis of plasma is a weak predictor of coronary artery disease

Heide L Kirschenlohr[1], Julian L Griffin[1], Sarah C Clarke[2], Ranyl Rhydwen[2], Andrew A Grace[1,2], Peter M Schofield[2], Kevin M Brindle[1] & James C Metcalfe[1]

Metabolomics Platform

Metabolism of Complex Carbohydrates

Biodegradation of Xenobiotics

Metabolism of Complex Lipi

eotide bolism

Ru | Greg | Arvind | Oded

Carbohydrate Metabolism

Metabolism of Other Amino Acids

Amino Acid Metabolism

Vamsi

Energy Metabolism

Rob

Steve Carr