Opportunities for pathway discovery in childhood cancer using metabolomics

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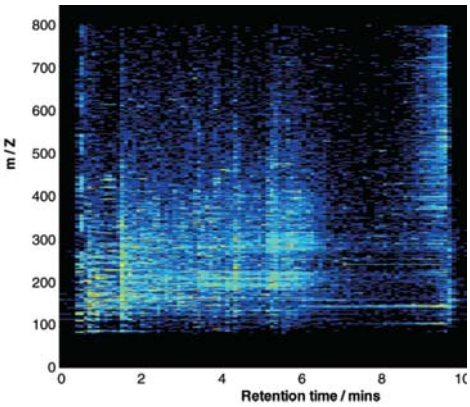
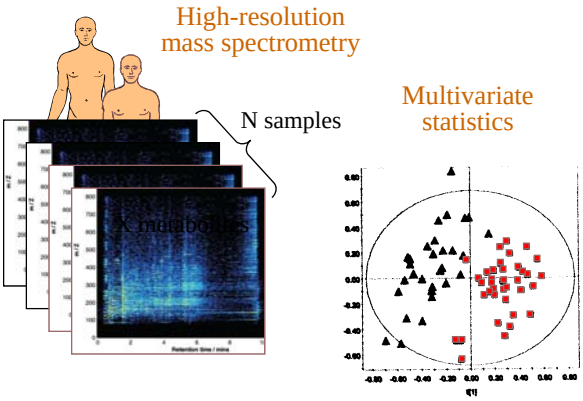
Metabolomics
A considerable information in biospecimens

Plasma sample

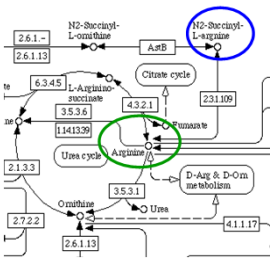


- >10,000 chromatographic features
- Low volumes of sample (plasma, 20 uL; urine, 1 uL)

Metabolomics – Looking for biomarkers



Data
interpretation

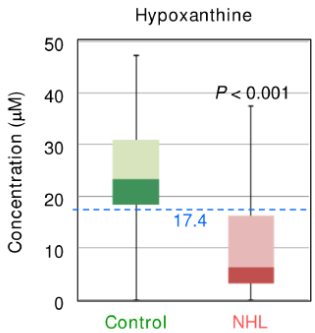


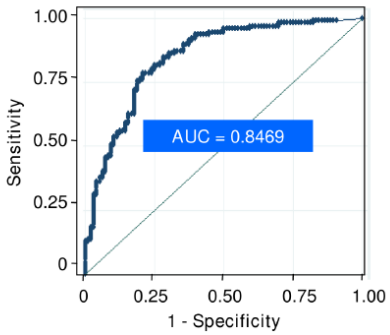
→ New putative biomarkers
  → New risk factors for cancer
  → New dietary biomarkers
→ New mechanistic hypotheses

Metabolomics and non-Hodgkin lymphoma

Discovery phase
30 Non-Hodgkin lymphoma patients, 30 controls
Urine
MALDI-Tof mass spectrometry

Validation phase
96 Non-Hodgkin lymphoma patients, 121 controls
Enzymatic kit



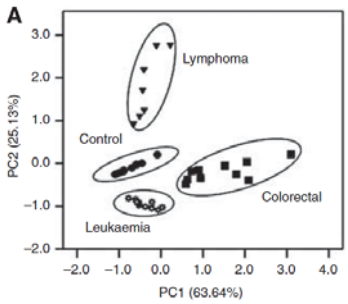


- Hypoxanthine as a biomarker for NHL

Yoo B.C., 2010, *BMC Cancer*

Urine volatiles and cancer patients

14 leukaemia, 12 colorectal and 7 lymphoma patients
21 healthy controls
Urine
Volatiles (SPME in headspace mode)
GC-MS



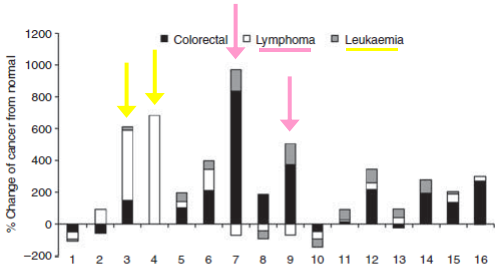
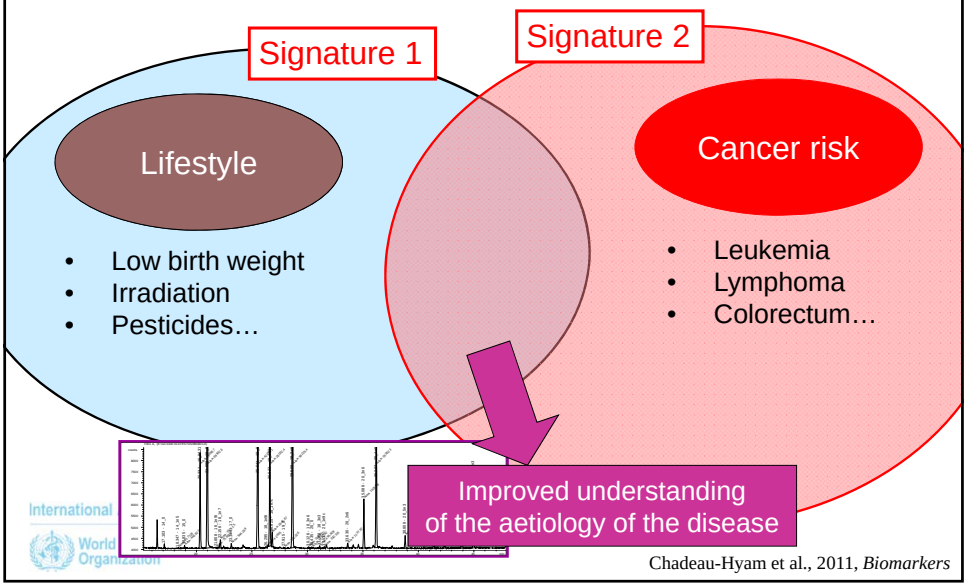


Figure 5 Percentage of change between cancer patients and normal subjects. Numbered bars correspond to: (1) hexanal; (2) heptanal; (3) 2-methyl-3-phenyl-2-propenal; (4) 3-heptanone; (5) 1,2,4-trimethylbenzene; (6) *p*-cymene; (7) anisole; (8) γ-terpinene; (9) bornylene; (10) dimethyl disulphide; (11) 2-methoxythiophene; (12) 4-methyl-phenol; (13) 1-octanol; (14) 1,2-dihydro-1,1,6-trimethyl-naphthalene; (15) 1,4,5-trimethyl-naphthalene; and (16) 2,7-dimethyl-quinoline.

- Metabolites characteristic of different cancer sites

Silva et al., 2011, *Br J Cancer*

Metabolomics and cancer aetiology
The meet-in-the-middle concept



Chadeau-Hyam et al., 2011, *Biomarkers*

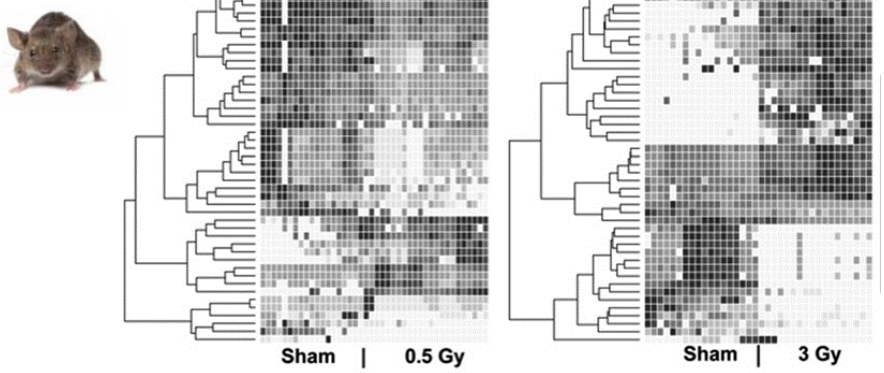
Metabolomics and risk factors for childhood cancers

Risk factor	Sample	Analytical method	Metabolites influenced	Reference
Radiation	Urine	MS		Coy, 2011
Pesticides	Urine	NMR	Glycine, threonine, lactate, glycerophosphocholine, citrate	Bonvallot, 2013
Low birth weight	Cord blood	NMR	Choline, proline, glutamine, alanine, phenylalanine, citrulline, glucose	Ivorra, 2012
	Cord blood	Targeted analysis	Choline, betaine, dimethylglycine	Hogeveen, 2013
	Cord blood	MS	Tryptophane, phenylalanine, isoleucine, valine, caffeine	Favretto, 2012
	Cord blood	Targeted analysis	Caffeine, paraxanthine	Grosso, 2006

- Methylation reactions, energy metabolism, xenobiotic metabolism, glucose intolerance, adipogenesis

Metabolomics and dosimetry

γ-irradiated wild type C57Bl/6 mice
Serum
ESI+ MS
50 top metabolites

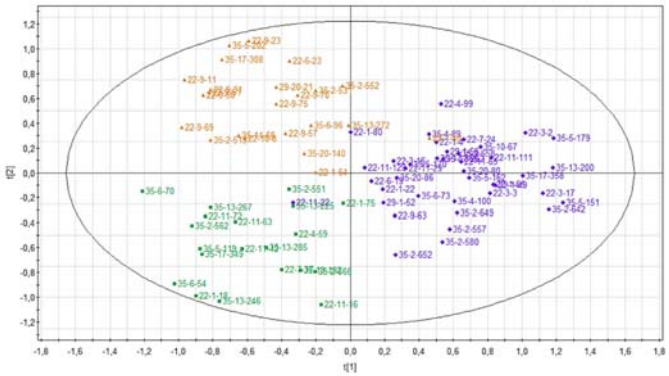


- In prostate cancer patients, correct classification of urine samples before and after irradiation

Coy et al., 2011, *Int J Radiat Biol*

Metabolomics and exposure to pesticides

83 pregnant women from Brittain (France)
3 groups according to the surface of land dedicated to cereal crops in the town of residence
Urine
^{1}H-NMR
PLS-DA

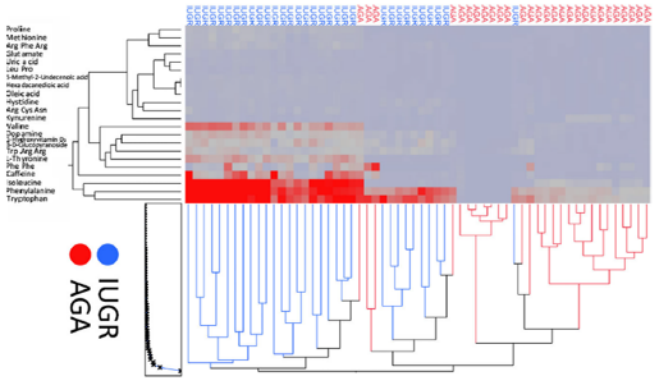


- Significant changes were observed for glycine, threonine, lactate, glycerophosphocholine and citrate

Bonvallot et al., 2013, *Plos ONE*

Metabolomics and low birth weight

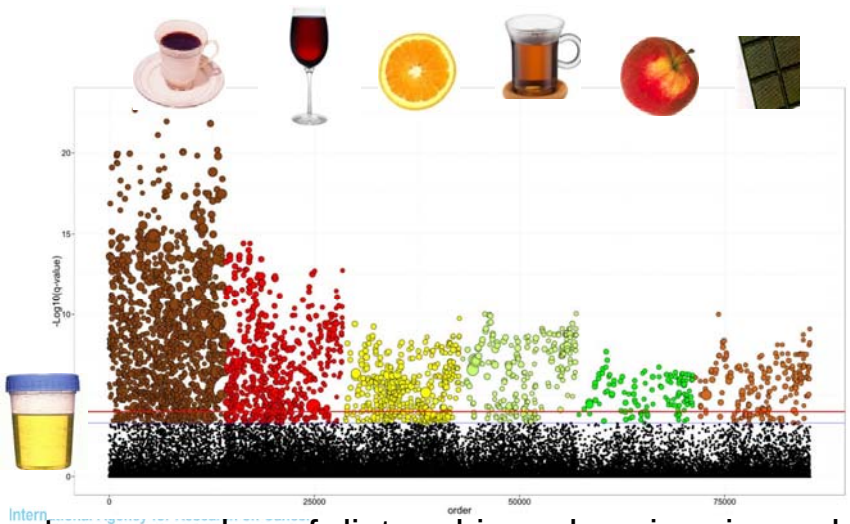
Cord blood
Fetuses with low (IUGR, n=22) or normal (AGA, n=21) birth weight
Orbitrap MS



- 22 metabolites associated to low birth weight
- Several amino acids (PheAla, Try, Isoleu, Val)
- Caffeine

Favretto et al.. 2012, *Anal Bioanal Chem*

Metabolomics and dietary exposure



- Large number of dietary biomarkers in urine or blood

International Agency for Research on Cancer
World Health Organization

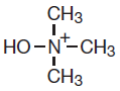
A new dietary risk factor for disease in a fully agnostic EWAS

Gut flora metabolism of phosphatidylcholine promotes cardiovascular disease

Zeneng Wang[1,2], Elizabeth Klipfell[1,2], Brian J. Bennett[3], Robert Koeth[1], Bruce S. Levison[1,2], Brandon DuGar[1], Ariel E. Feldstein[1,2], Earl B. Britt[1,2], Xiaoming Fu[1,2], Yoon-Mi Chung[1,2], Yuping Wu[4], Phil Schauer[5], Jonathan D. Smith[1,6], Hooman Allayee[7], W. H. Wilson Tang[1,2,6], Joseph A. DiDonato[1,2], Aldons J. Lusis[3] & Stanley L. Hazen[1,2,6]

Wang Z. et al., 2011, *Nature*

- Cases: myocardial infarction, stroke
- Controls matched for age and gender
- Plasma collected 3 years before
- LC-MS, m/z 50-200



Choline
Betaine
TMAO

Liver
Red meat
Poultry
Milk
Fish
Shell fish

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World Health Organization

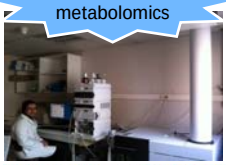
- 3 metabolites derived from the diet predict disease risk

Metabolomics and childhood cancers

- A powerful approach to discover new biomarkers and generate novel mechanistic hypotheses
- No prospective studies in the field of cancer so far
- Exposome-wide association studies to discover novel risk factors

BMA Group

Non-targeted metabolomics



UPLC-QTof

Computing capacity

Metabolite databases

Targeted metabolomics

UPLC-QqQ