Biomarkers in Public Health: Development and Applications

Irina Stepanov, Ph.D.

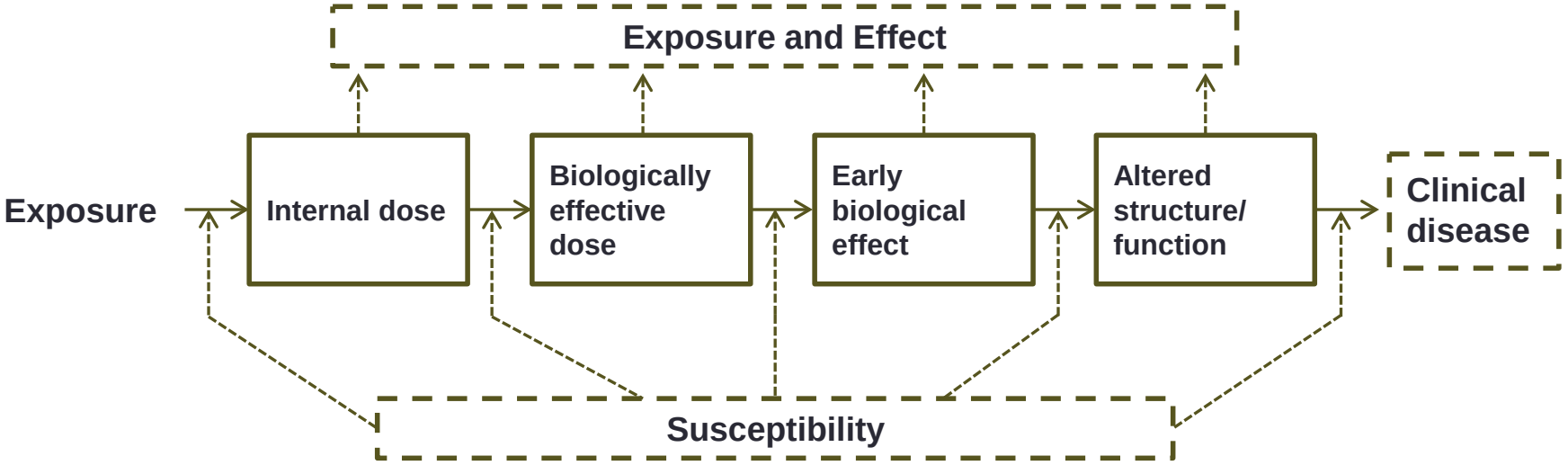
Assistant Professor

Division of Environmental Health Sciences and Masonic Cancer Center
University of Minnesota

Biomarker paradigm

1987, U.S. National Research Council

Biomarker paradigm



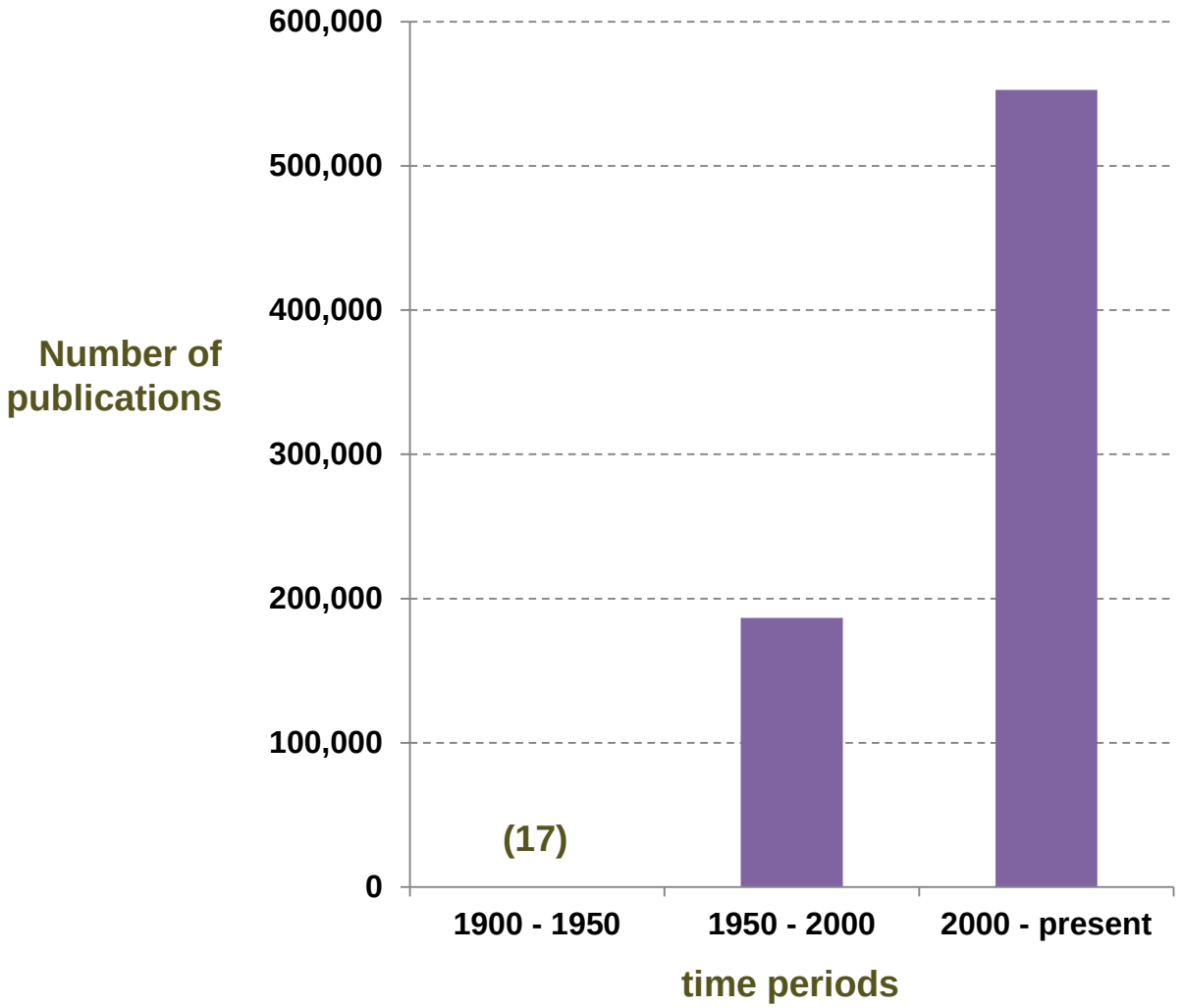
1987, U.S. National Research Council

What is a biomarker?

An indicator of exposure, effect, susceptibility, or clinical disease that can be measured in a biological system

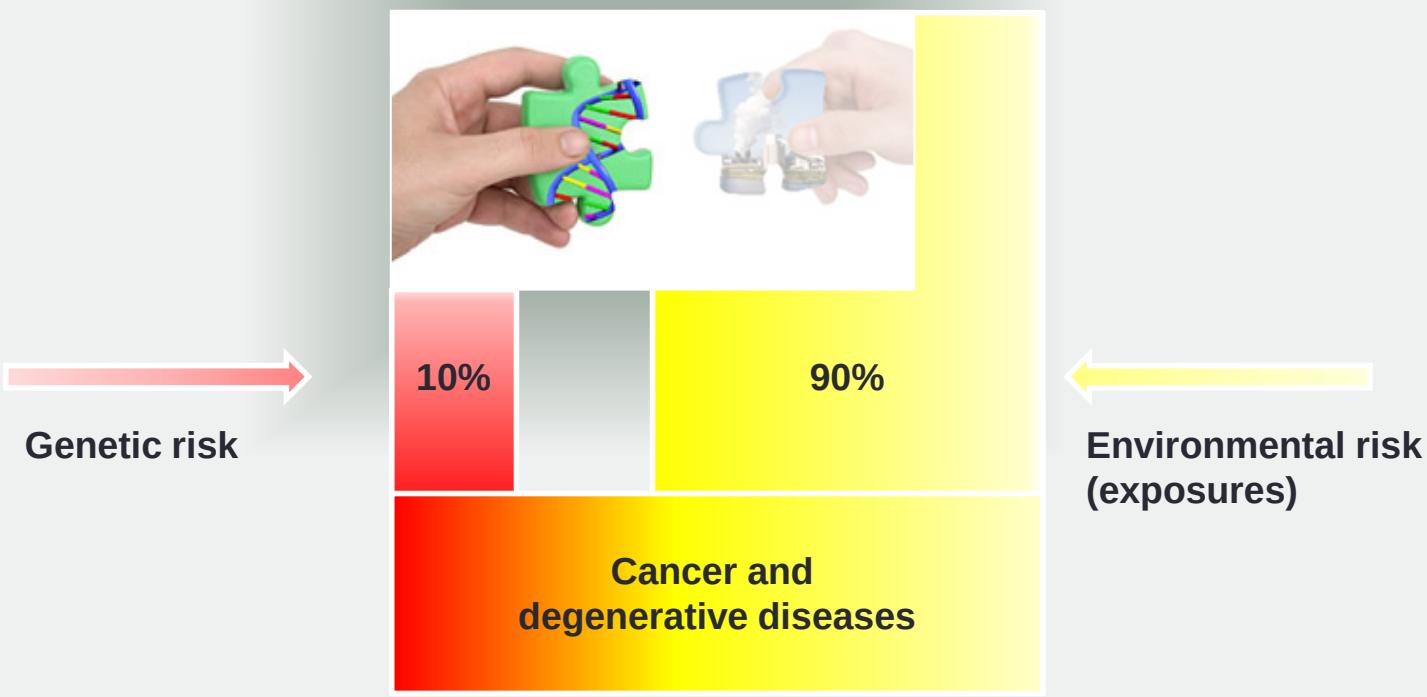
Category	Examples
Unchanged exogenous agents (exposure)	Asbestos fibers, metals
Metabolized exogenous agents (exposure, susceptibility)	Phenol for benzene, cotinine for nicotine, metabolite ratios
Endogenous molecules exposure effect susceptibility	 Porphyrin ratios (metals, dioxins) DNA and protein adducts Gene polymorphisms
Cellular/tissue changes (effect, susceptibility)	Neutrophil to lymphocyte ratios, sperm counts

Research publications on biomarkers



Results of PubMed search

Most diseases are believed to result from a complex interaction between an individual's genetic make-up and environmental agents



Adapted from M.T. Smith and S.M. Rappaport, *Environ Health Perspect*, 117(8): A334-A335 (2009).

Exposure assessment approaches

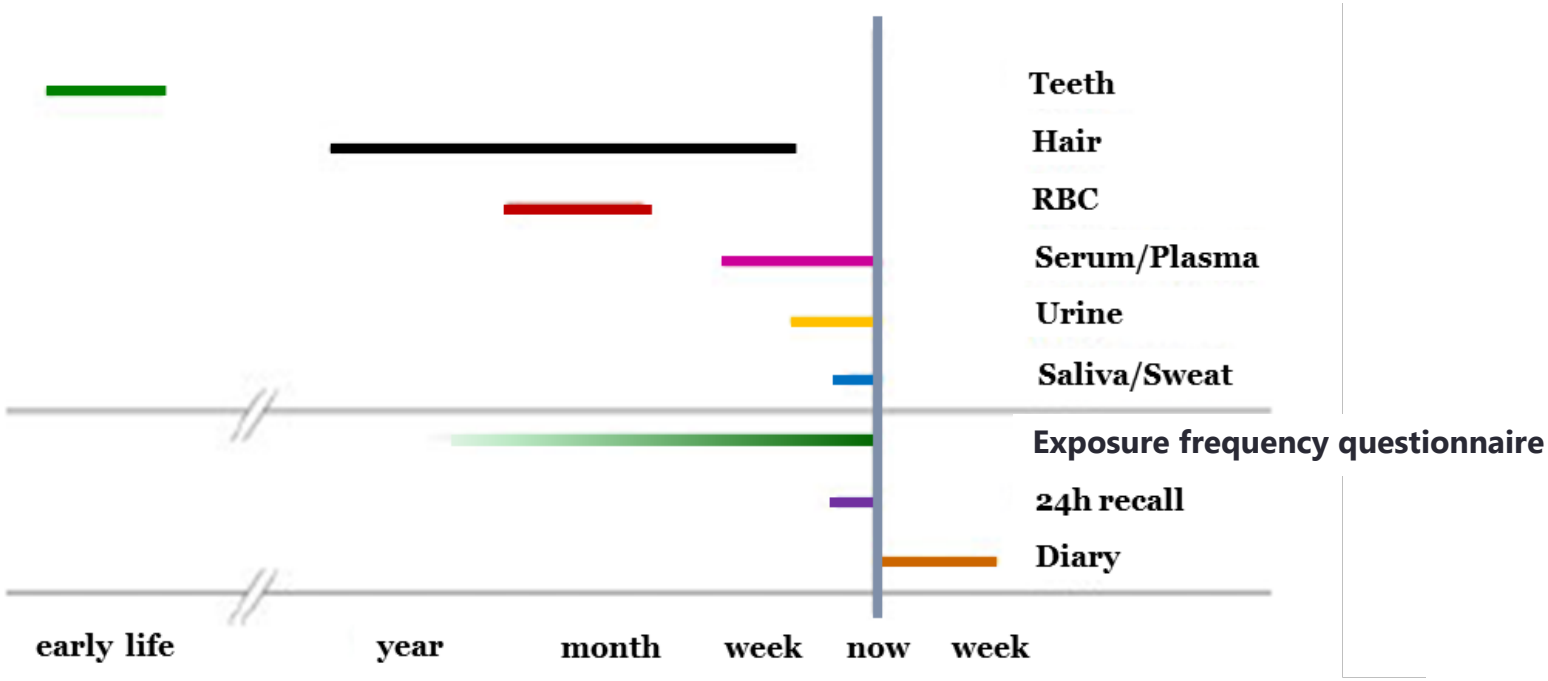
Questionnaires

Interviews

Diaries

Biomarker measurements

Different biological sample types for the biomarker-based exposure assessment



Kuhnle G. Nutritional biomarkers for objective dietary assessment. J Sci Food Agric 2012; 92: 1145–1149

Advantages and disadvantages of different biological sample types

Type	Advantages	Disadvantages
Blood	Easy to collect, regularly replenished, a way for chemicals to travel within the body	Invasive, cultural rejection, short lifespan
Urine	Non-invasive, large amounts	Metabolites are measured, more than a single time point or 24-h collections are often needed
Adipose tissue	Good matrix for analysis of non-polar chemicals	Invasive
Breast milk	Ease of collection, may reflect historical exposures to lipid-soluble chemicals	Limited application
Hair	Non-invasive, temporal distribution	Not many methods are validated, trace levels, potential of external incorporation
Nails	Non-invasive, reflect long-term cumulative exposure	Not many methods are validated, trace levels
Saliva	Non-invasive	Not widely used
Exhaled air	Non-invasive	Limited application (volatile compounds)

Characteristics of a good biomarker?

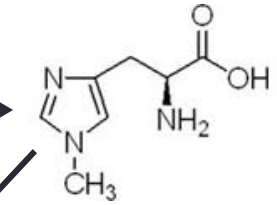
- ✓ **Specificity (to exposure, disease)**
- ✓ **Sensitivity (in response to changes in exposure, disease)**
- ✓ **Technical feasibility (collection, measurement)**
- ✓ **Reproducibility (frame of reference)**
- ✓ **Biological relevance (timeframe, mechanism of effect and disease)**
- ✓ **Predictive value**

Biomarker development

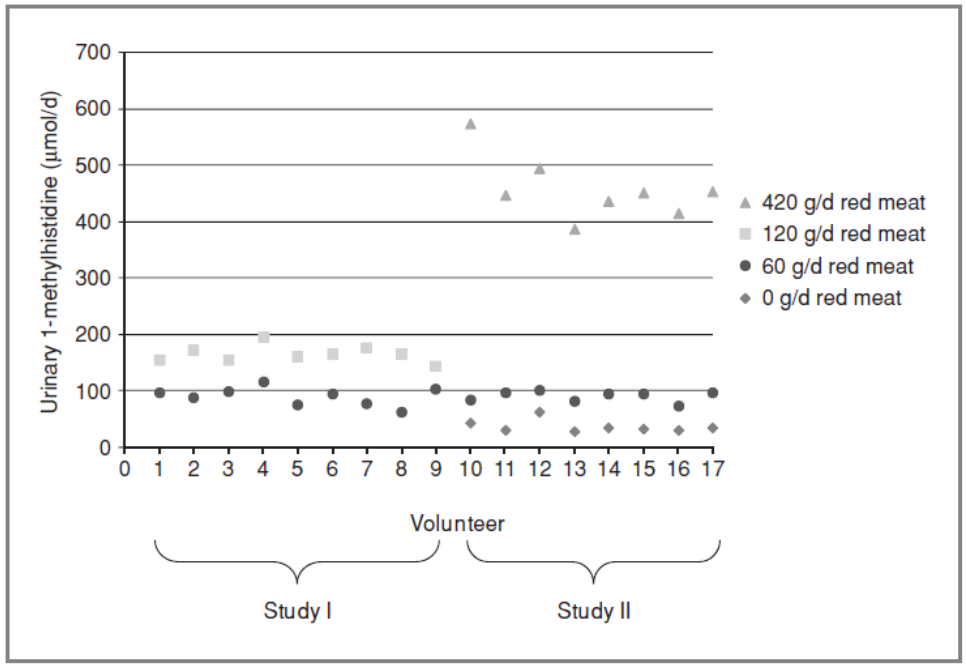
Biomarker development strategies

Hypothesis driven

- **Food**
- **Food composition data; Bioavailability; Metabolism**
- **Identification of likely candidate markers**
- **Development of specific analytical method**
- **Dietary intervention with extremes (nil/high) of food**
- **Analysis of candidate biomarkers in collected specimens**
  - Technical feasibility
  - Specificity
  - Sensitivity
- **Confirmation of suitability of candidate biomarker**
- **Application in epidemiological or clinical settings**
- **Biomarker validation**

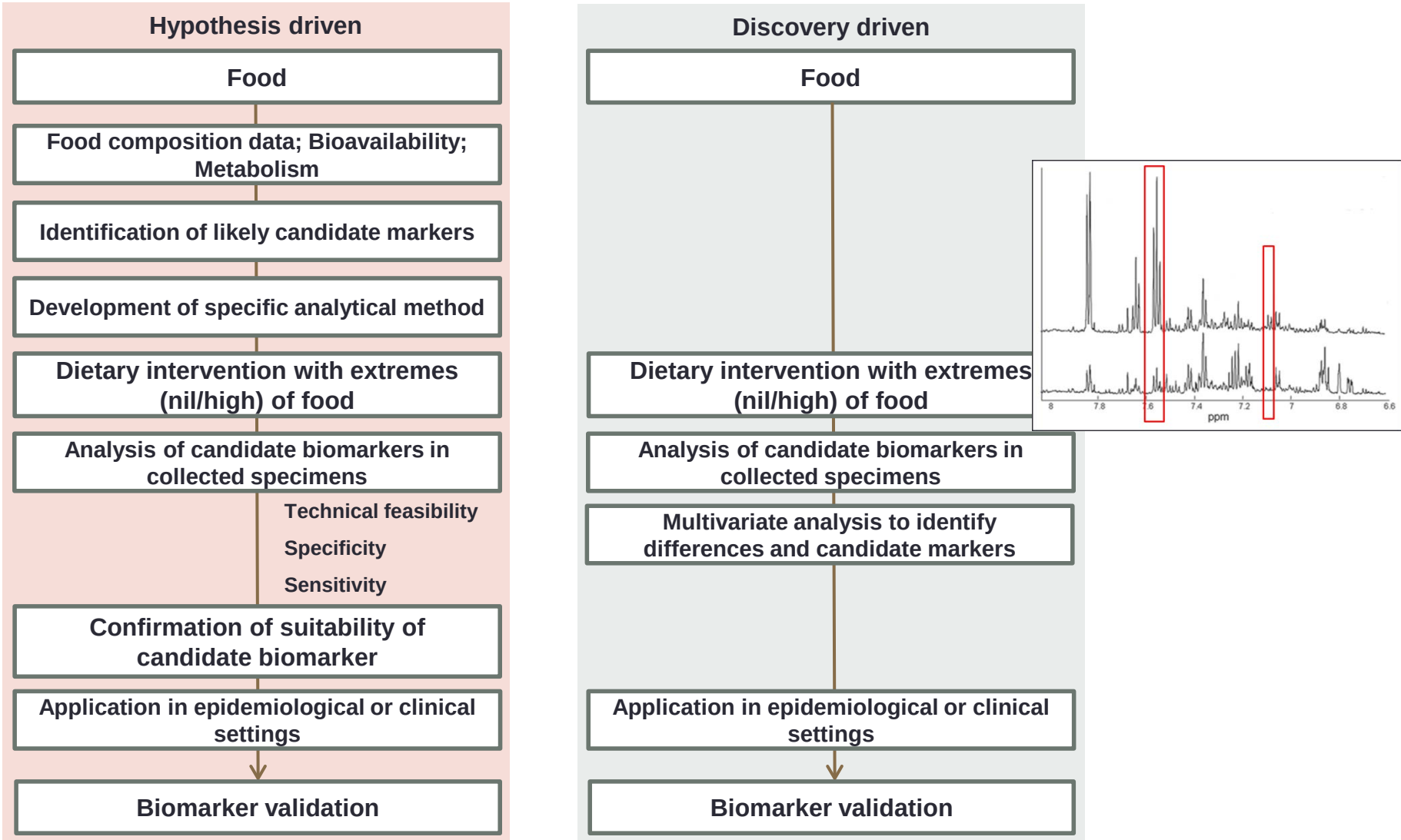


1-methylhistidine



Kuhnle G. J Sci Food Agric 2012;92:1145–1149; Gross et al. CEBP 2011;20(6):1107-1111

Biomarker development strategies

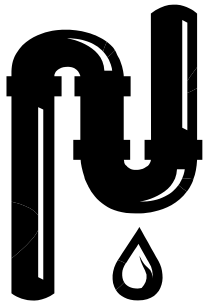


Kuhnle G. J Sci Food Agric 2012;92:1145–1149;

Examples of biomarker applications in Environmental Health

Lead (Pb)

banned



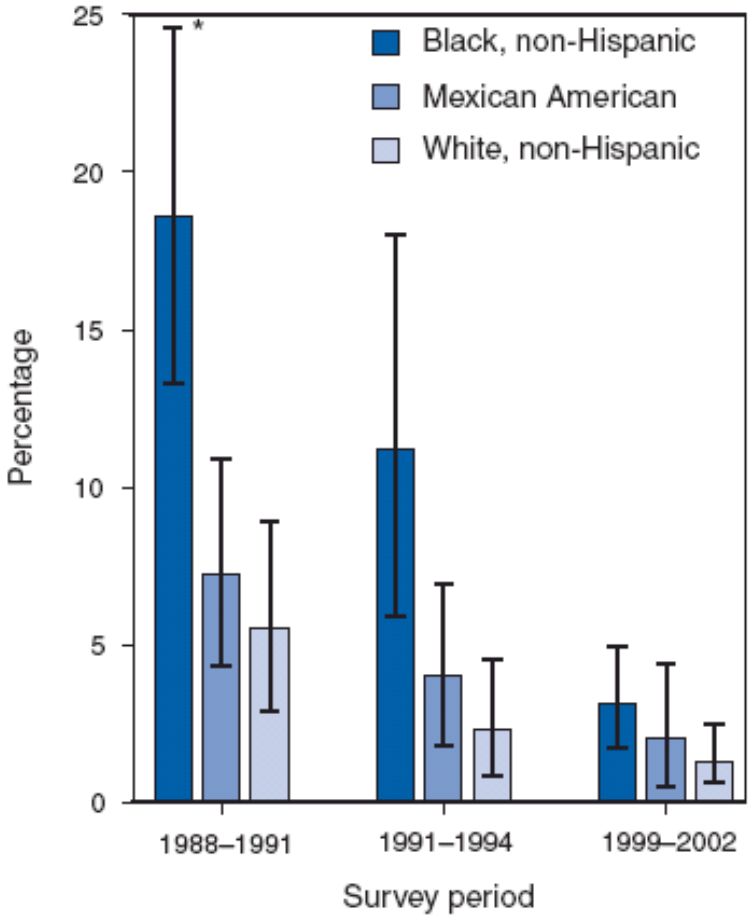
banned, copper is used instead

outlawed since 1978

however…

Temporal changes in blood Pb levels

FIGURE. Percentage of children aged 1–5 years with blood lead levels ≥10 µg/dL, by race/ethnicity and survey period — National Health and Nutrition Examination Surveys, United States, 1988–1991, 1991–1994, and 1999–2002

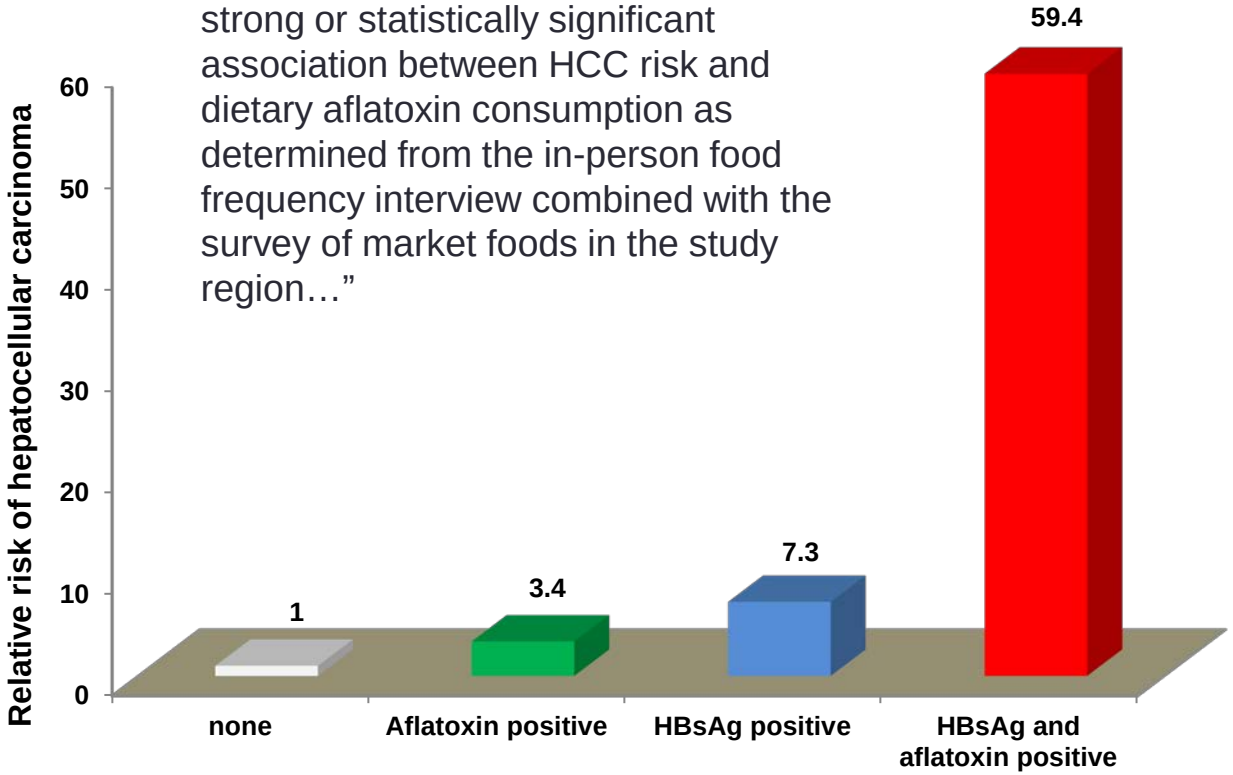


*95% confidence interval.

CDC, MMWR, May 27, 2005 / 54(20);513-516

Aflatoxin exposure an hepatocellular carcinoma (HCC)

"… On the other hand, cohort analysis using all cases of HCC revealed no strong or statistically significant association between HCC risk and dietary aflatoxin consumption as determined from the in-person food frequency interview combined with the survey of market foods in the study region…"



G S Qian, R K Ross, M C Yu, et al. *Cancer Epidemiol Biomarkers Prev* 1994;3:3-10.

Use of biomarkers in environmental health

Identifying priority exposures

Identifying at-risk populations

Recognizing time-trends in population exposures

Evaluation of exposure reduction and prevention activities

Checking the validity of traditional exposure models

Establishing reference ranges for comparison

Application of biomarkers in tobacco carcinogenesis studies

Stepanov Lab research focus:

Applying biomarker-based approach to characterize and quantify the links between exposures and health outcomes

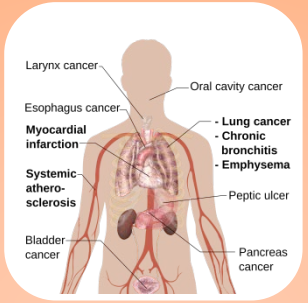
Chemical toxicants and carcinogens in environmental sources

Biomarkers:

Exposure

Metabolism

Effect



Cancer

Inflammation-driven diseases

Neurobehavioral abnormalities

Quantitative links

Biomarker expertise

Tobacco exposures

- Tobacco-specific *N*-nitrosamines
- Cotinine (nicotine)
- Polycyclic aromatic hydrocarbons (1-HOP)
- HPB-releasing DNA adducts

Oxidative DNA damage

- 8-oxo-dG
- 8-oxo-dA
- M_1dG

Trace elements (sample collection and processing)

- Manganese
- Iron
- Cadmium

Why do we study tobacco?

100,000,000

people died from tobacco use in the 20th century

1,000,000,000

people will die from tobacco use in the 21st century

Main health consequences of smoking

Over 7,000 constituents

- **Nicotine**
- **Numerous toxicants**
- **More than 70 carcinogens**

Addiction

Toxicity

- **Respiratory**
- **Cardiovascular**

Carcinogenicity

- **19 types of cancer**

Why do we study tobacco?

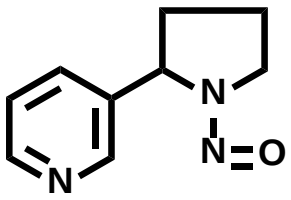
Understand the diversity of tobacco products and related risks

Understand mechanisms of diseases caused by tobacco use

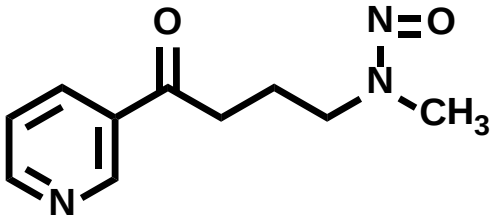
- **Identify susceptible individuals**
- **Develop preventive measures**

Build science base for tobacco product regulation by the FDA

Focus on NNN and NNK



NNN

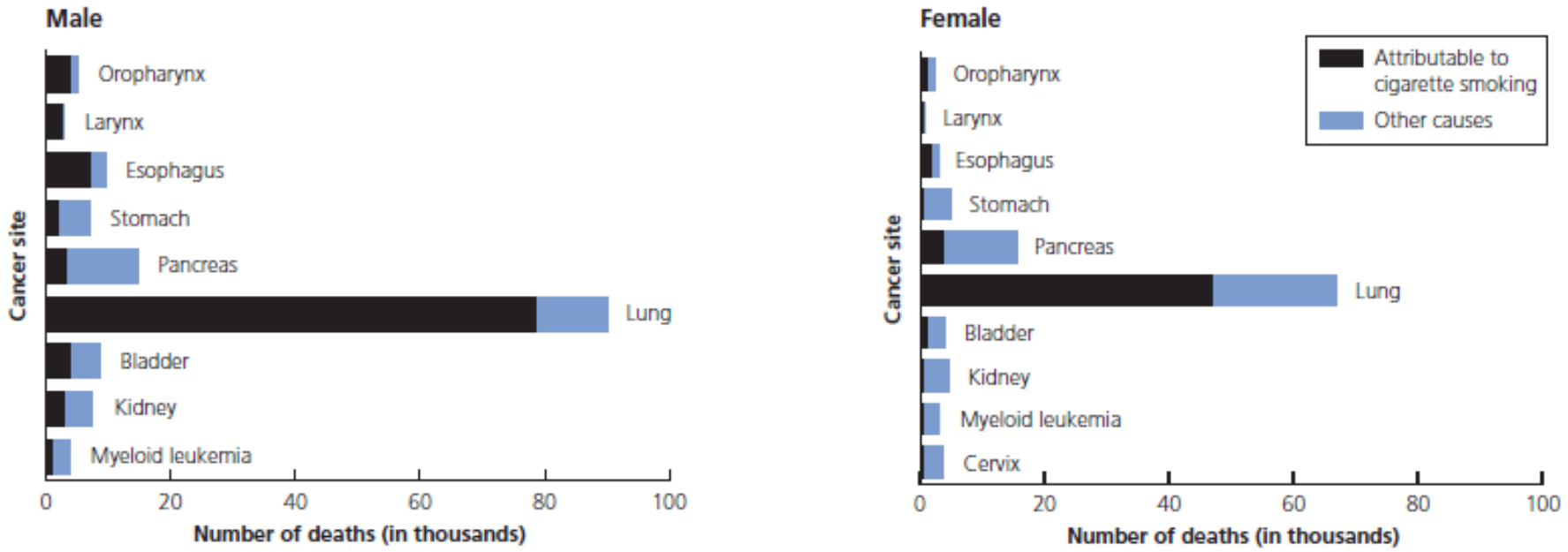


NNK

- The most important carcinogens in smokeless tobacco
- NNN: oral and esophageal cancer
- NNK: lung and pancreatic cancer

Cancer deaths due to smoking (US)

Annual Number of Cancer Deaths Attributable to Smoking by Sex and Site, US, 2000-2004

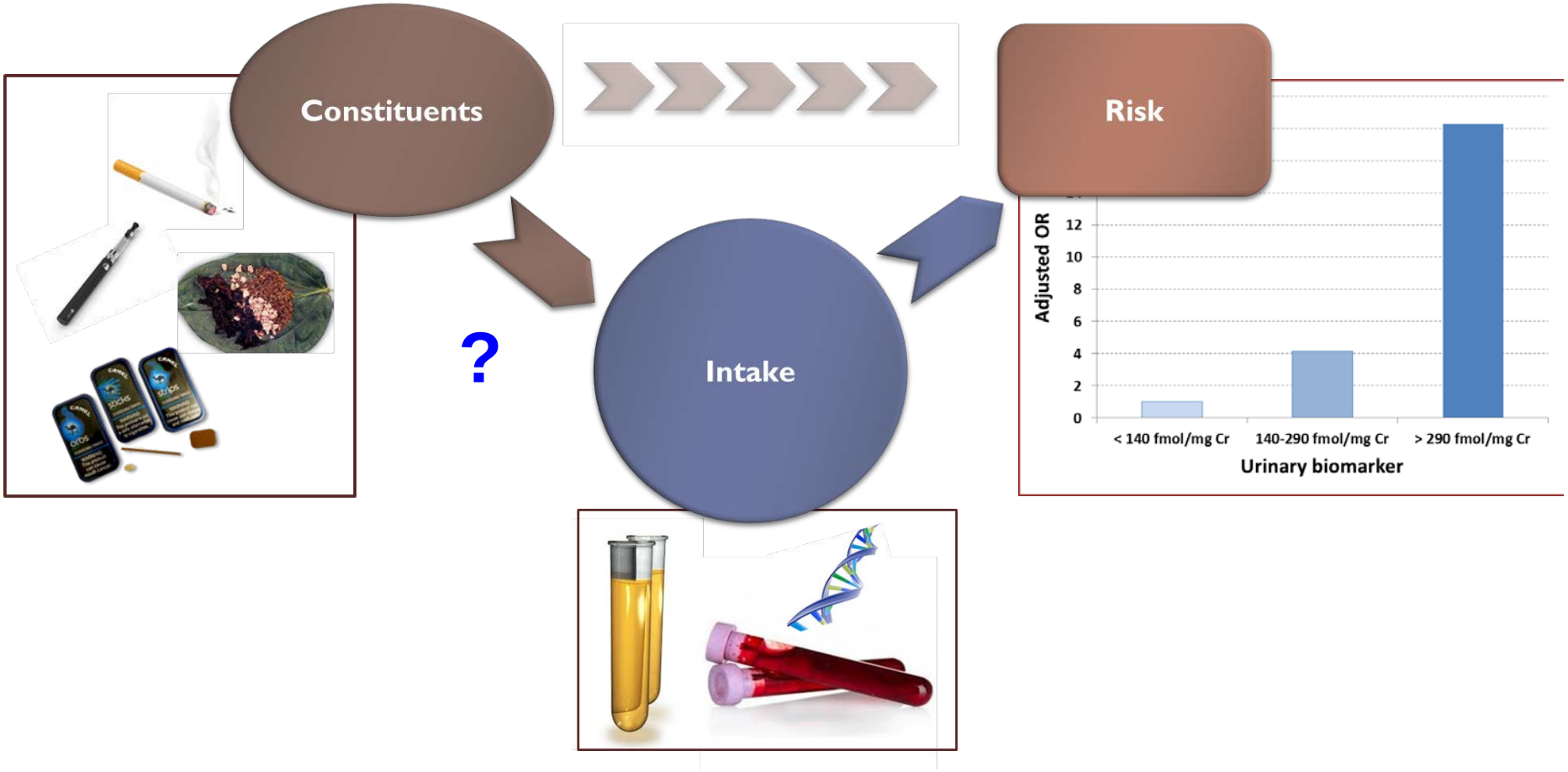


Source: Centers for Disease Control and Prevention. Smoking-attributable mortality, years of potential life lost, and productivity losses – United States, 2000-2004. *MMWR Morb Mortal Wkly Rep*. 2008;57(45):1226-1228.

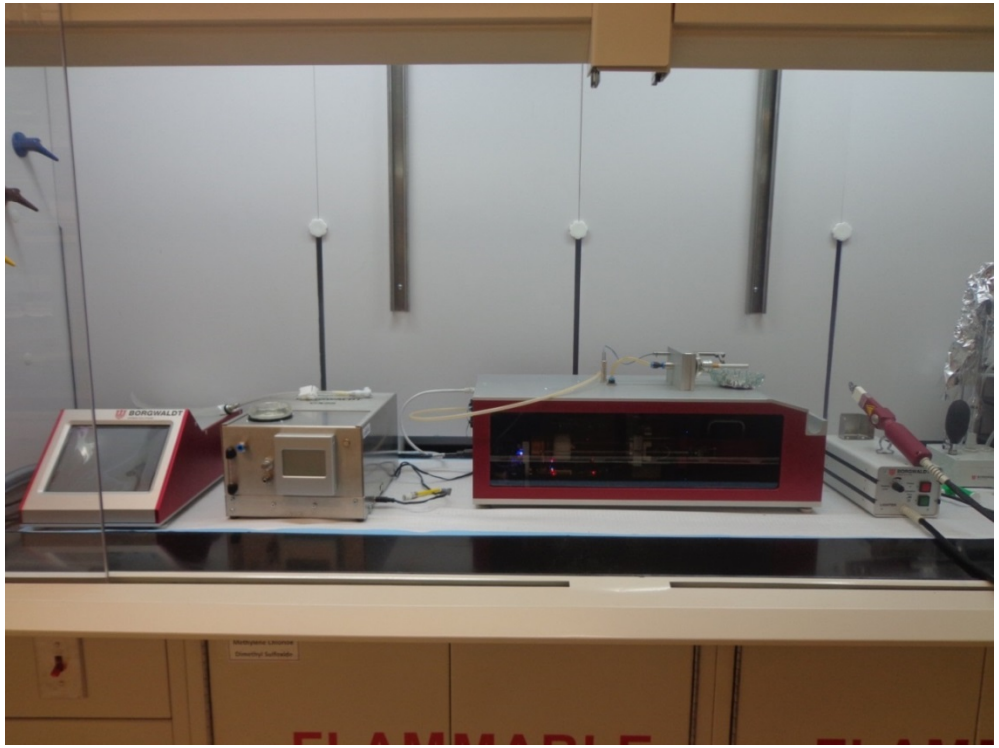
American Cancer Society, Surveillance and Health Policy Research, 2010

ACS, Cancer Facts and Figures 2010

Research approach

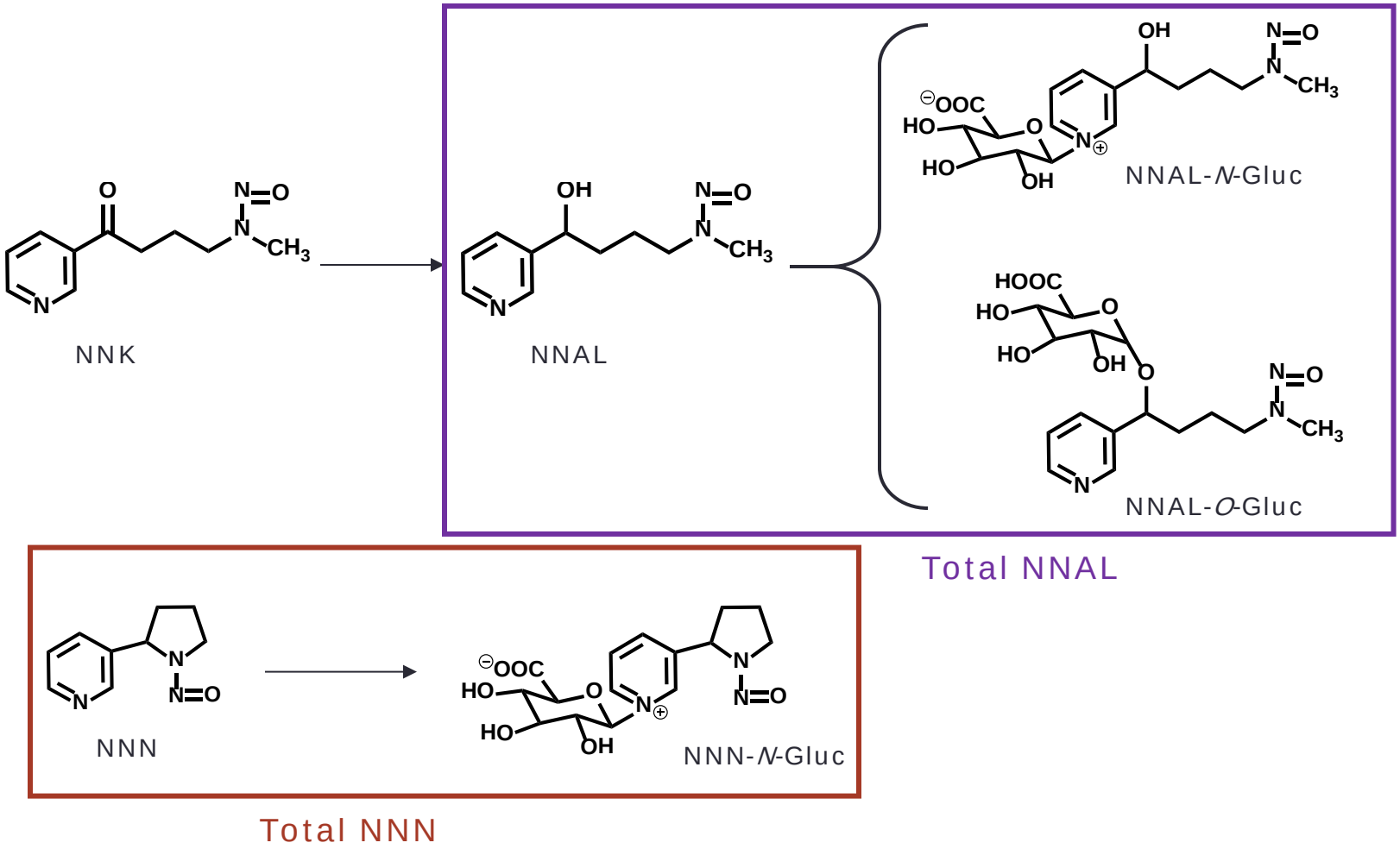


Exposure measurement



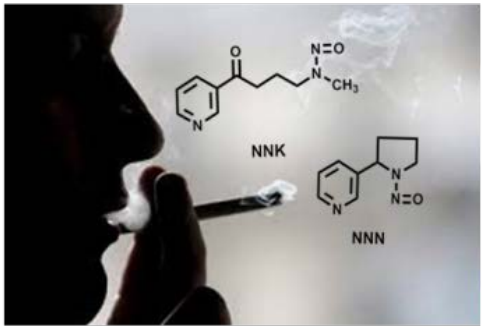
Product analysis

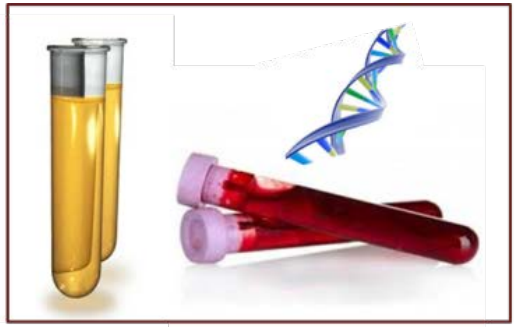
Biomarkers of NNK and NNN intake

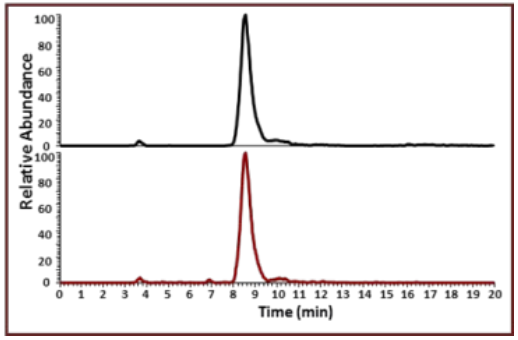


Measuring the relationship between product content and constituent intake

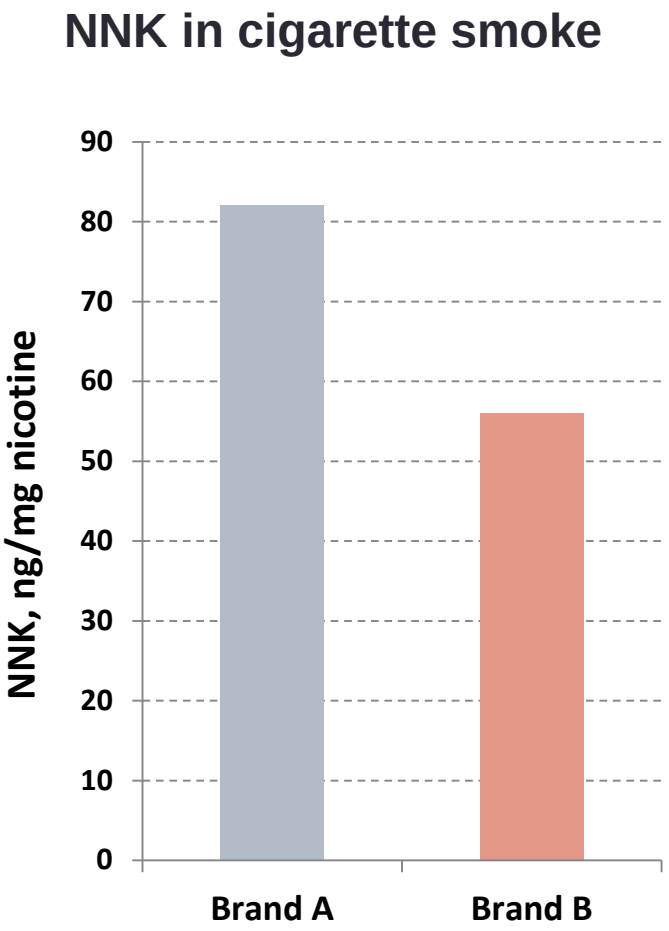
Habitual users of various brands

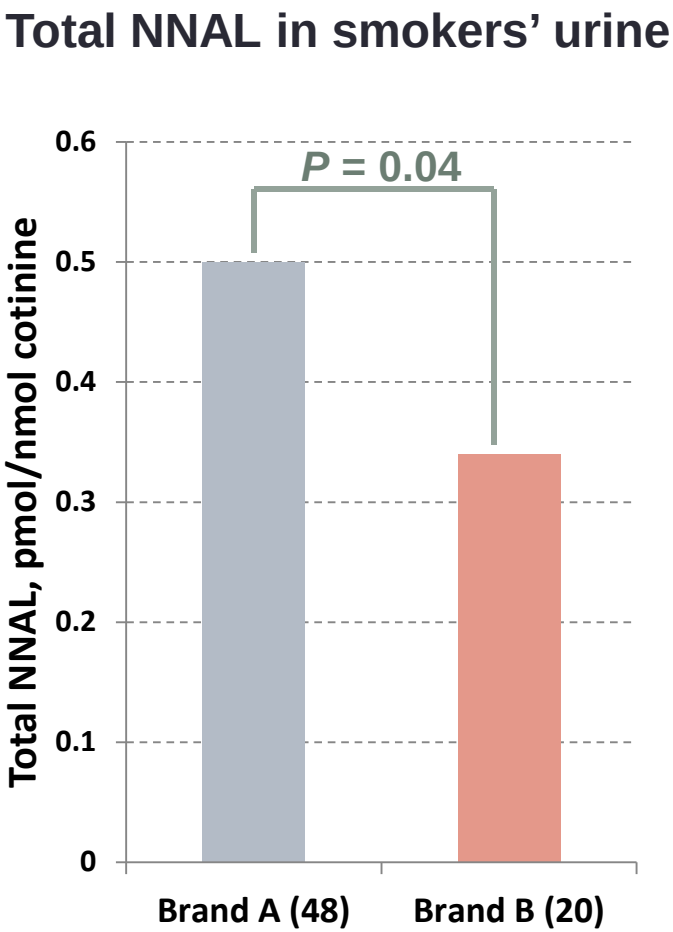






Urinary total NNAL in US smokers of different cigarette brands





Relationship between constituent levels and biomarkers of intake in smokeless users

Smokeless tobacco users (343)

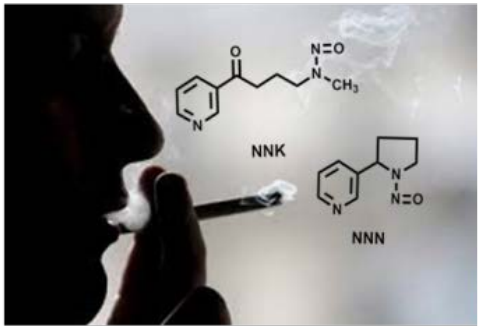
Products with wide range of nicotine and TSNA levels

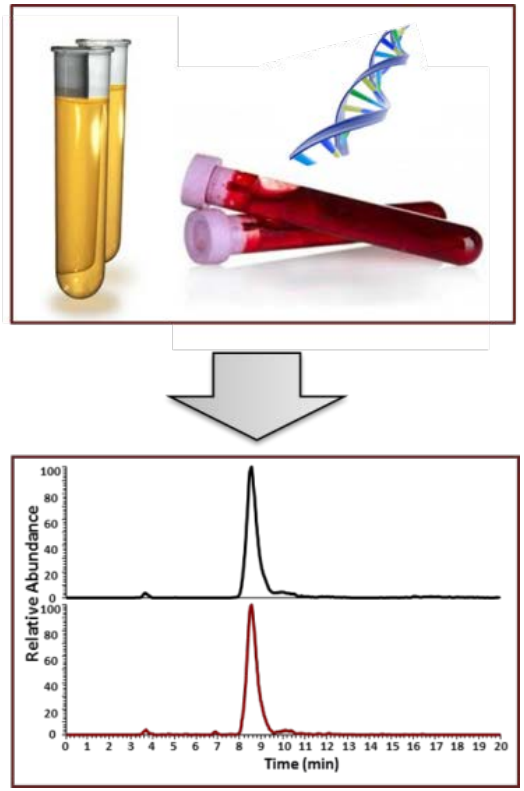
Multiple regression analysis for biomarkers (P-values)

Covariates	Urinary biomarkers		
	Total nicotine equivalents	Total NNN (NNN intake)	Total NNAL (NNK intake)
Constituent level in product	0.155	<0.001	<0.001
Dip weight	0.086	0.068	<0.001
Total daily dip duration	<0.001	<0.001	<0.001

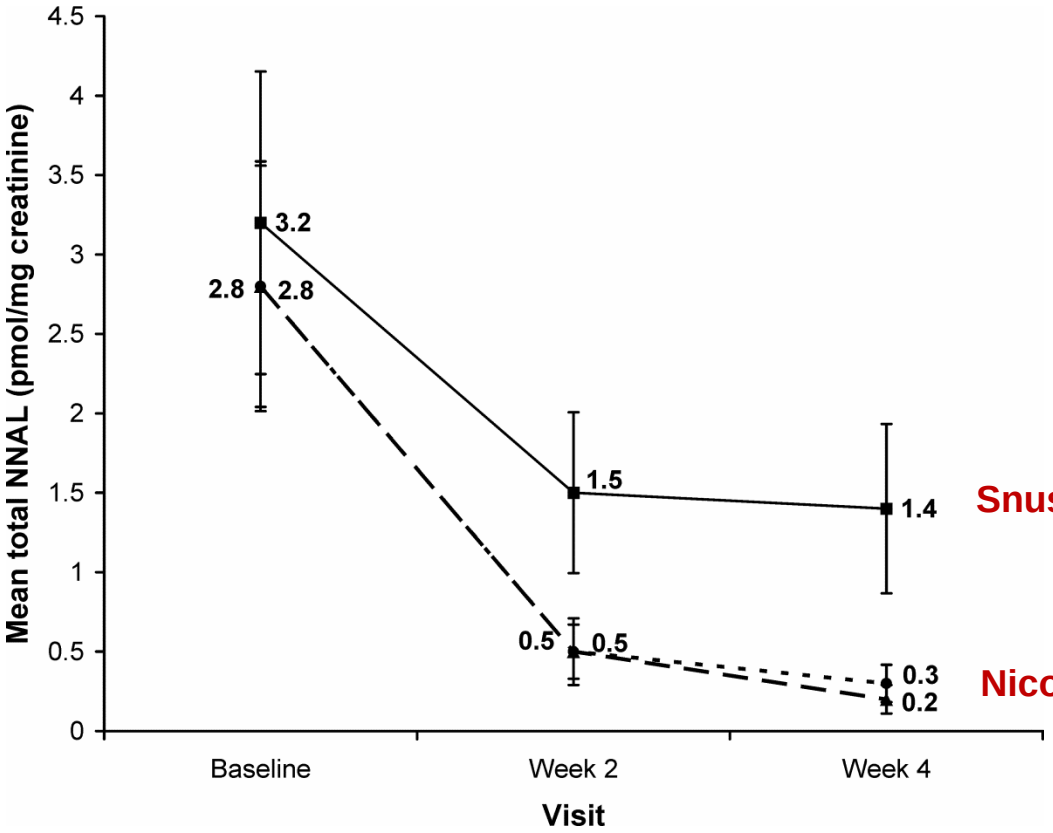
Measuring the relationship between product content and constituent intake

Switching studies



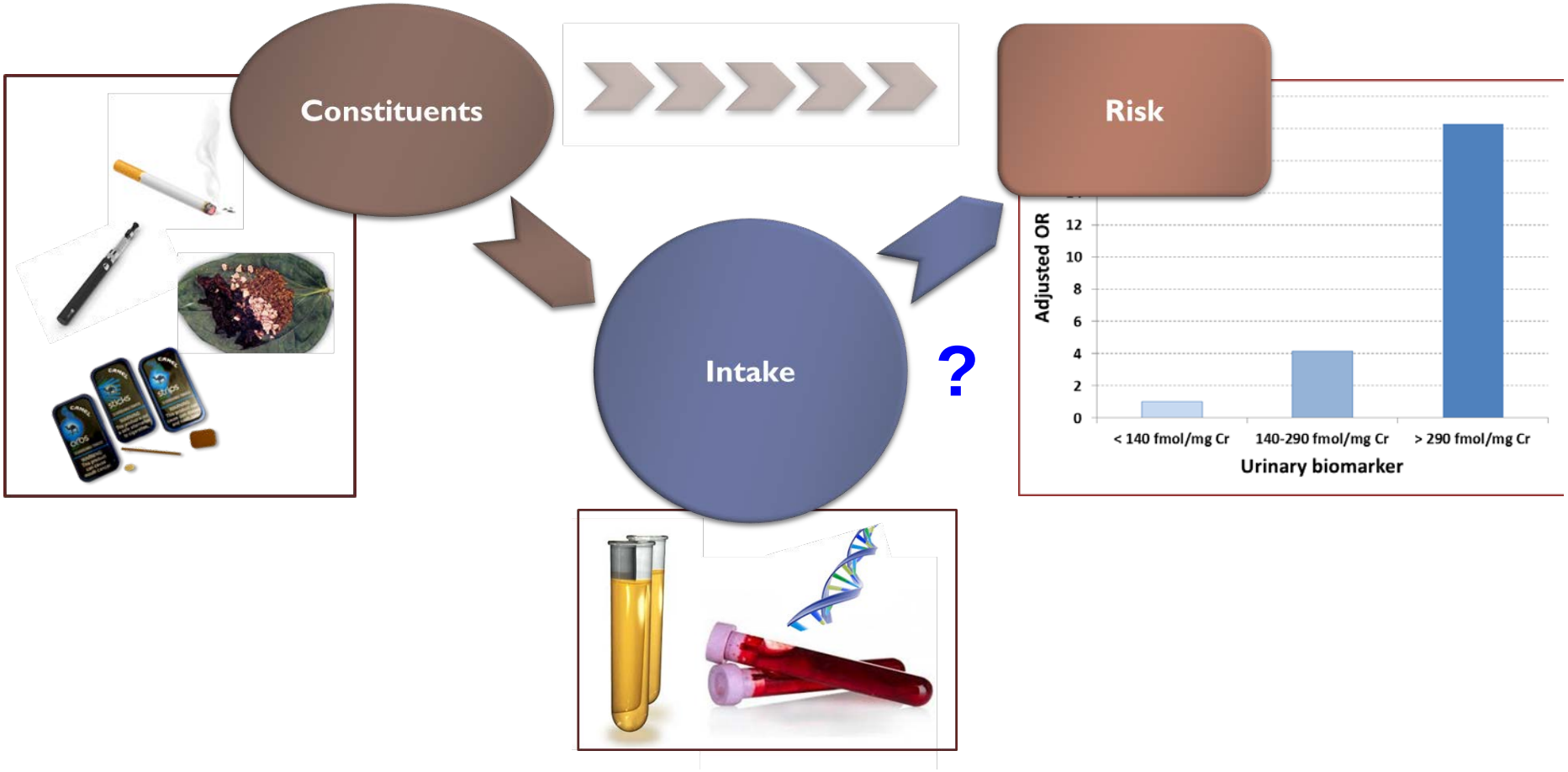


Total NNAL in urine of smokeless tobacco users assigned to low-TSNA products



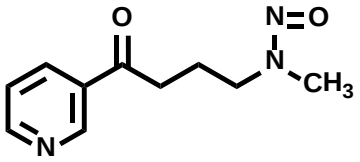
Hatsukami et al. J Natl Cancer Inst. 2004;96:844-852

Research approach



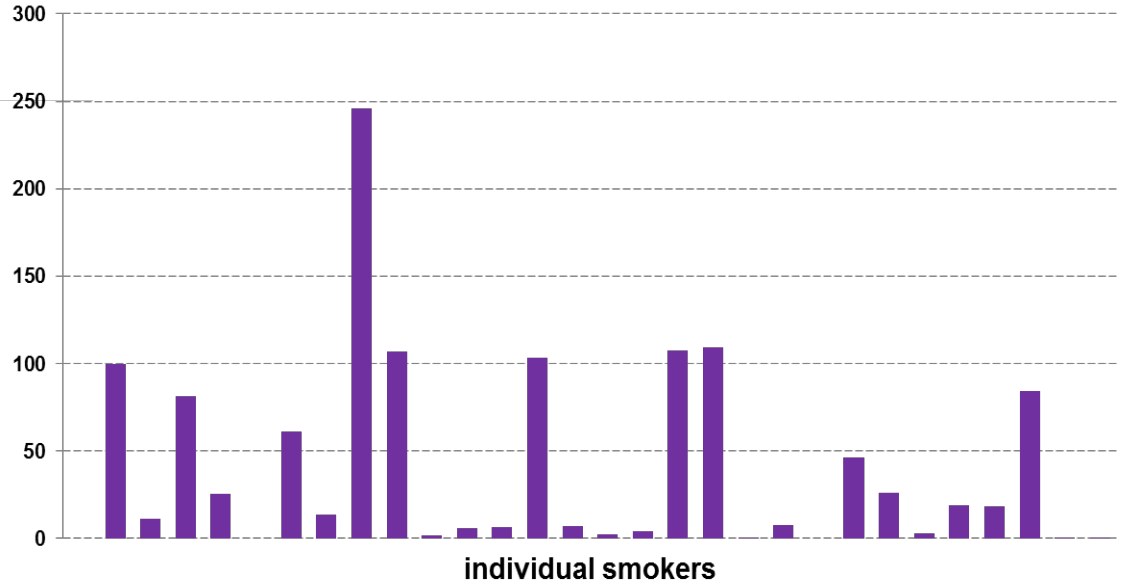
Understanding individual susceptibility

DNA damage



NNK

metabolic activation



Linking exposure to cancer risk

Shanghai Prospective Cohort

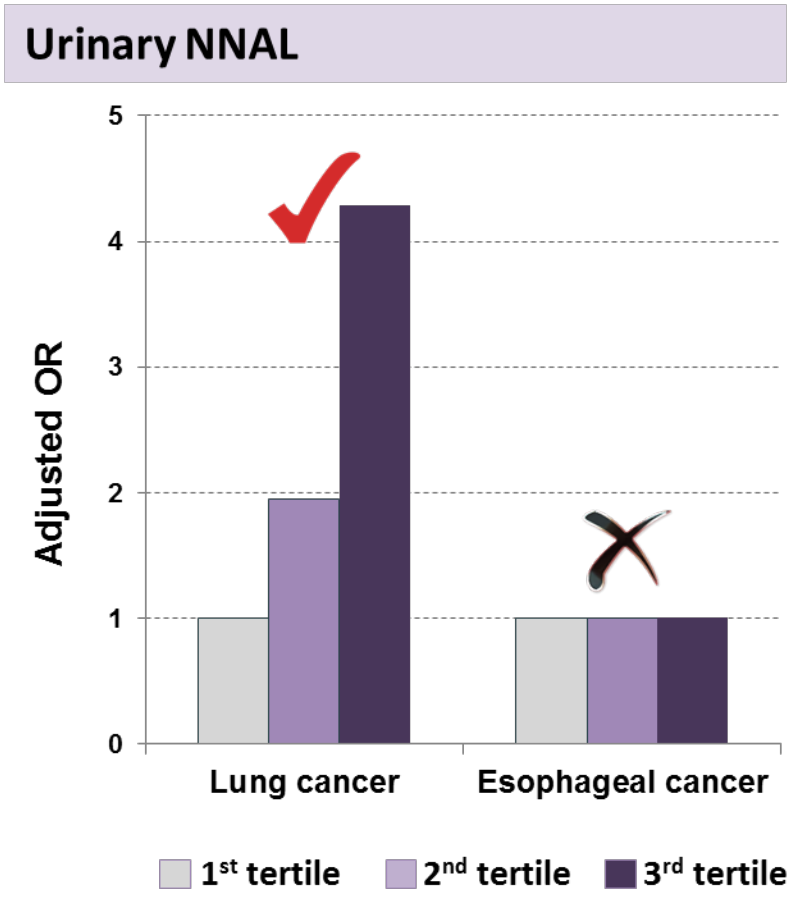
18,244 men (45-64 years old) enrolled between 1986 and 1989

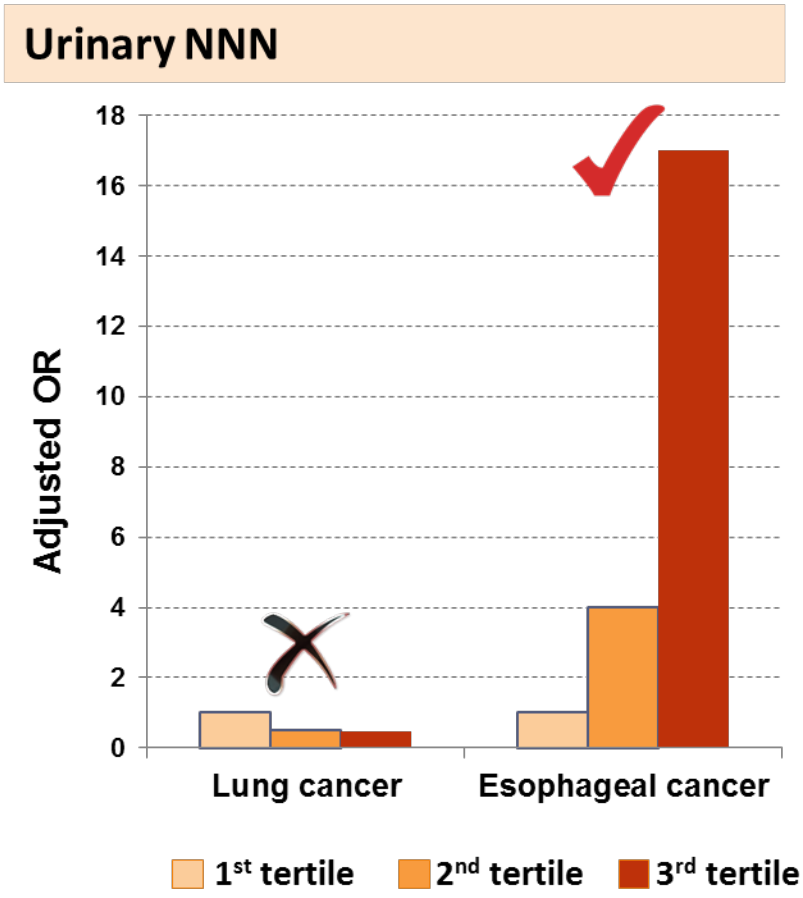
use of tobacco and alcohol, usual diet, and medical history

10-ml blood sample and a single spot urine sample

Yuan et al, *Carcinogenesis (2011);32:1366-1371*

Relationship between TSNA exposure and cancer risk in humans





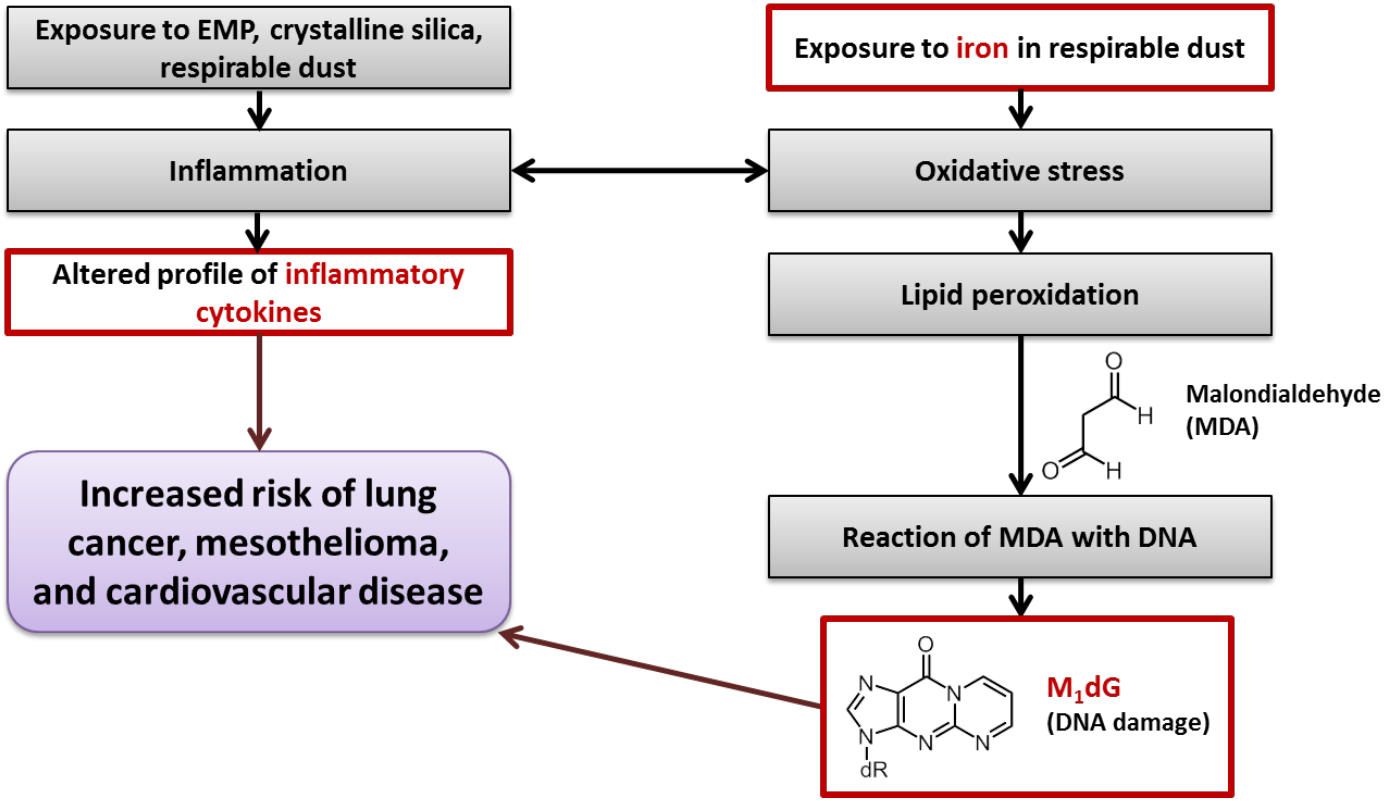
J.-M. Yuan et al, Carcinogenesis 32(9): 1366-1371 (2011);
I. Stepanov et al. Int J Cancer 134:2278-2283 (2014)

Ongoing non-tobacco studies

Oxidative DNA damage in Taconite Workers Health Study

Oxidative DNA damage in Taconite Workers Health Study

Oxidative stress and inflammation, induced by the dust from taconite operations and mediated by iron exposure, are important contributors to the development of cancer and cardiovascular disease in taconite workers.



Oxidative DNA damage in age-related macular degeneration (AMD)

- The leading cause of blindness among older adults in the developed world
- Approximately 30% of individuals over 75 years are affected
- Indications of increased oxidative mitochondrial DNA damage in the retinal pigment epithelium

11-55 ng mtDNA