Development and Validation Dietary Methodology for N- Nitroso Compounds

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USDA/ARS

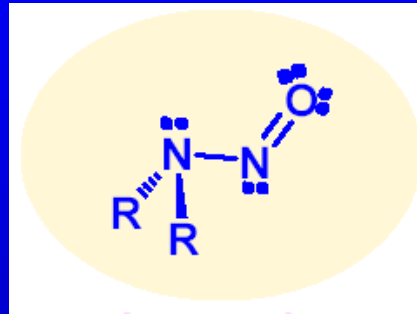Children's Nutrition Research Center

Houston, Tx

N-Nitroso Compounds associated with Cancer

- Evidence from Animal Experiments

- Biological Mechanisms

- Epidemiological Studies

  however, dietary assessment was incomplete

N-Nitroso Compounds

Ubiquitous in nature and are carcinogenic, mutagenic, and teratogenic

- Tobacco

- Occupational

- Dietary

Classification of N-Nitroso Compounds in Foods

Major class	Abbreviation minor class	High Source Foods
Volatile	NDMA	Beer, sausage, cured meats
	NDEA	Sausage, cheese
	NPYR	Fried Bacon, sausage, ham
	NPIP	Bologna, sausage
Non-volatile	NSAR	Cured Meats
	NPRO	Fried Bacon, cured meat
	NTCA	Smoked meat
	NHMTCA	Smoked meat
Precursors	Nitrate	Vegetables, grains
	Nitrite	Additive to meats, other foods
	Amines	Protein foods
	Amides	Protein foods

Food Processing Techniques Promote N-Nitroso formation

- Cured with added nitrites
- Pickled
- Smoked
- Stored in Air with high N content
- Aged

Evolution of dietary exposure to N-nitroso compounds

Level	Characteristic	# of Foods	Location	Type of Cancer
1	Several Questions	(1-7 items)	Germany, US Sweden	Adult brain, Pedi Brain, other cancers
2	Moderate	43, 53, 74 items	US, Canada, Germany	Other cancers
3	Intermediate	45, 59, 77, 79, 125, 146 items, ____, ____	Italy, US, Australia, Spain, Sweden, Finland, US-NCI, Italy, France, Germany	Adult Brain, Pedi brain, others
4	Superior	47 items	US	Maternal intake for pedi brain

N-Nitroso Recent Scholarly Reviews

Pediatric Brain tumors

- Review of chemistry & carcinogenicity
- Limitations of previous studies
- Meta analysis re Pedi Ca

- M Dietrich Cancer Causes & Control 16:619, 2005

Pancreatic Cancer

- **Risk linked to N-nitroso**
- Excess gastric acidity
- cytokine and immune functions
- H pylori
- Nitrate & above ↑ P Ca, and calls for more studies
- HA Risch J Natl Cancer Inst. 95:13,2003

Objectives

- To develop a comprehensive database of all N-nitroso compound concentrations in foods

- To validate the modified N-nitroso dietary modified FFQ instrument with 7-day diet records in 100 individuals.

N-nitroso Database

Data management ACCESS & SAS

	Initial	Final
Compounds	**36**	**23**
References	**300+**	47
Units	11	one
Food entries	4309	

Abbreviation - Compound Name

NDBA	N-Nitrosodibutylamine
NDBZA	N-Nitrosodibenzylamine
NDEA	N-Nitrosodiethylamine
NDMA	N-Nitrosodimethylamine
NHMT	N-nitroso-2-(hydroxymethyl)thiazolidine
NHMTCA	N-nitroso-2-hydroxymethylthiazolidine-4-carboxylic acid
NHPRO	N-nitroso-4-hydroxyproline
NMTCA	N-nitroso-2-methylthiazolidine-4-carboxylic acid
NPYR	N-Nitrosopyrrolidine
NPRO	N-nitrosoproline
NSAR	N-Nitrososarcosine
NTCA	N-nitrosothiazolidine-4-carboxylic acid

Protocol to assign Values to FFQ

- FFQ food matched to NOC database food
  - Where possible (Direct Match) and (Imputed Value)

- FFQ food(s) matched to several NOC database food
  - Mean value for several foods (Direct Match) and (Imputed Value)

- Descriptors of foods aggregated to a Food line item are known, documented, and modifiable

FFQ Results

Assigned N-Nitroso values
to 137 of 207 FFQ items

N-Nitroso compounds with the highest number
assigned
– NDMA   NAA
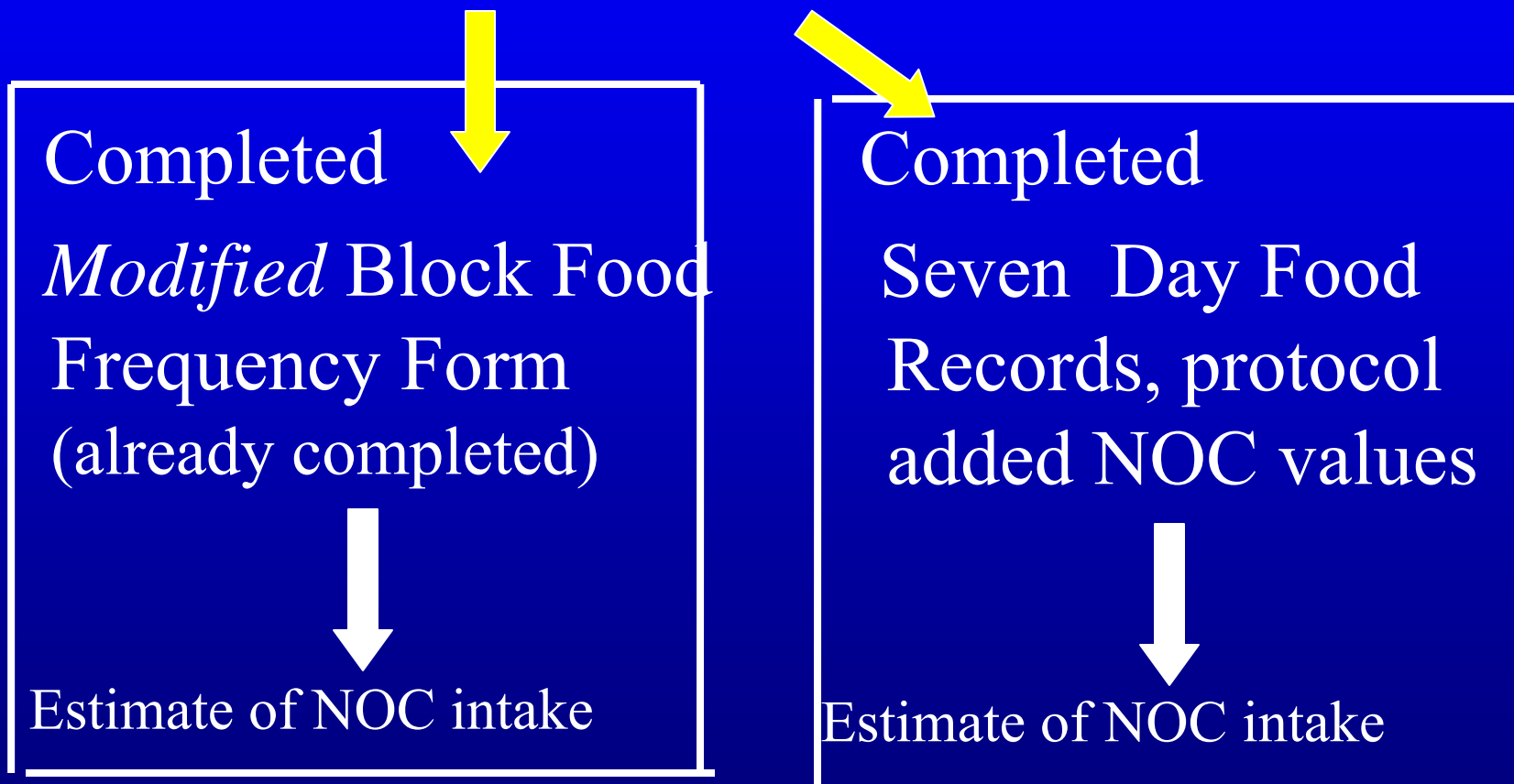– Nitrates   NPYR

FFQ Matching Results (cont.)

- 10 – 19 compounds
  - Cheese, bacon, sausage, cured lunch meats, sausage

- 5 – 9 compounds
  - Compounds dried meats, dairy, cooked meat, hot dogs

- 1 – 4 compounds
  - Soy, beans, pulses, grains, liquor, vegetables

Sample of N-Nitroso values to Block FFQ items

Block FFQ Food Item	N-nitroso Compound ug/100 g					
	NDMA	NPYR	NPIP	NDEA	NPRO	NTCA
Peas		0.025			0.300	
Whole milk	0.014	0.003	0.003			
Margarine	0.026					
French fries, fried potatoes	0.024	0.041				
White Wine	0.025	0.109				
Butter	0.026					
Refried beans or pinto beans	0.033					
Rolls,buns,muffins,bagels	0.050	0.009		0.023		
Cottage cheese	0.076	0.004	0.009	0.057		1.500
Fried fish	0.169	0.003		0.088	1.200	4.660
Beer	0.202					
Hot dogs or franks	0.221					8.950
Ham, boloney, lunch meats	0.490	0.534	0.004	0.149	5.752	46.130
Bacon	0.454	2.129	0.049	0.067	1.000	142.742
Virginia ham,smithfield ham	0.490	0.534	0.004	0.149	5.752	46.130
Sauerkraut	0.660	0.555	0.220	0.079	2.095	0.900
Oysters	1.139	0.038		0.109	5.416	0.864
Pizza	1.684	0.020		0.026	0.258	1.784
Sausage or chorizo	10.941	0.086	0.001	0.040	1.720	11.896

Study Design
100 Control Subjects, MDACC Brain Study

Completed

Modified Block Food
Frequency Form
(already completed)

Estimate of NOC intake

Completed

Seven Day Food
Records, protocol
added NOC values

Estimate of NOC intake

N-Nitroso Validation Study Subject Characteristics

Characteristic	n
Male	36
Female	61
White	73
Black	9
Asian	9
Hispanic	7
All	98

Comparison of FFQ ~ DFR Major Nutrients

|	FFQ	DFR
Calories, kcal	2300 ± 1300	1731 ± 513
Fat, g	92.9 ± 53.6	68.8 ± 24.4
Protein, g	97.0 ± 51.4	73.2 ± 25.3
Calcium, mg	1065 ± 565	756 ± 311
Vitamin C, mg	210 ± 190	128 ± 275

Protocol to Assign N-Nitroso values to Food Records

- Food and ingredient files, NDS
- Food and ingredient files, sorted, same sequence as food items on the FFQ
- NOC assigned lookup or matching function
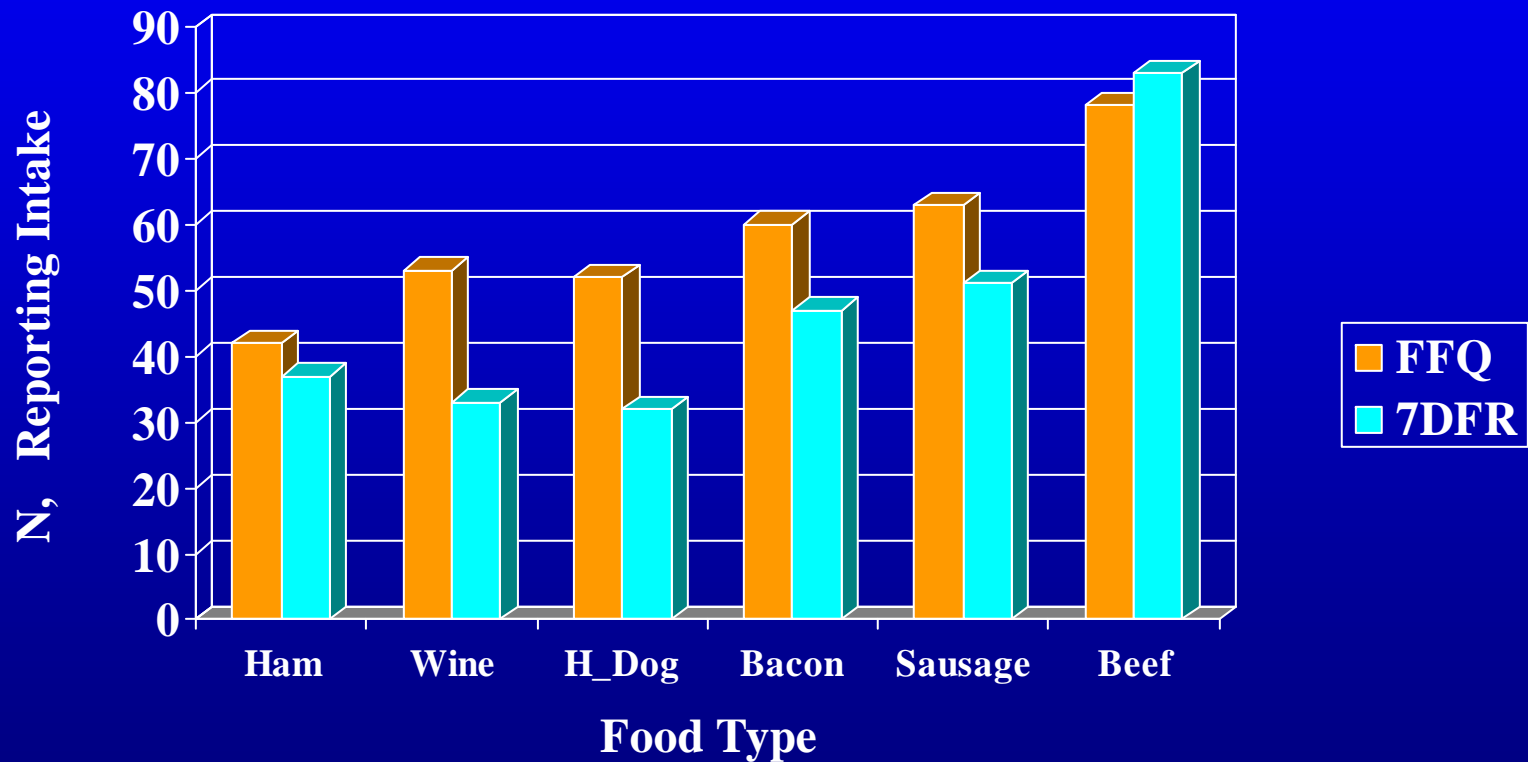
Daily Median Intakes
N-Nitroso Compounds FFQ vs. 7 DFR
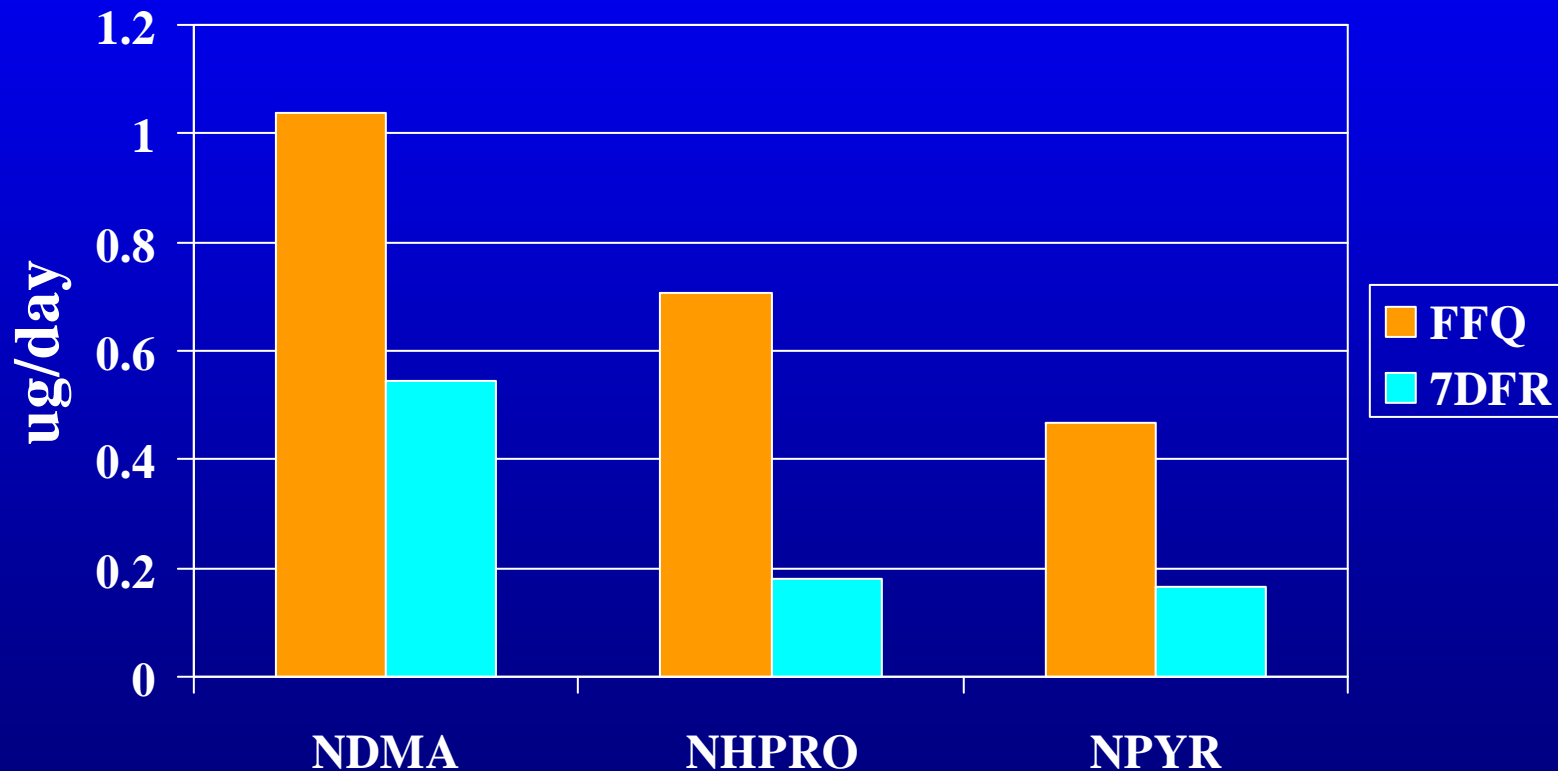
Daily Median Intakes (cont).
N-Nitroso Compounds FFQ vs. 7 DFR
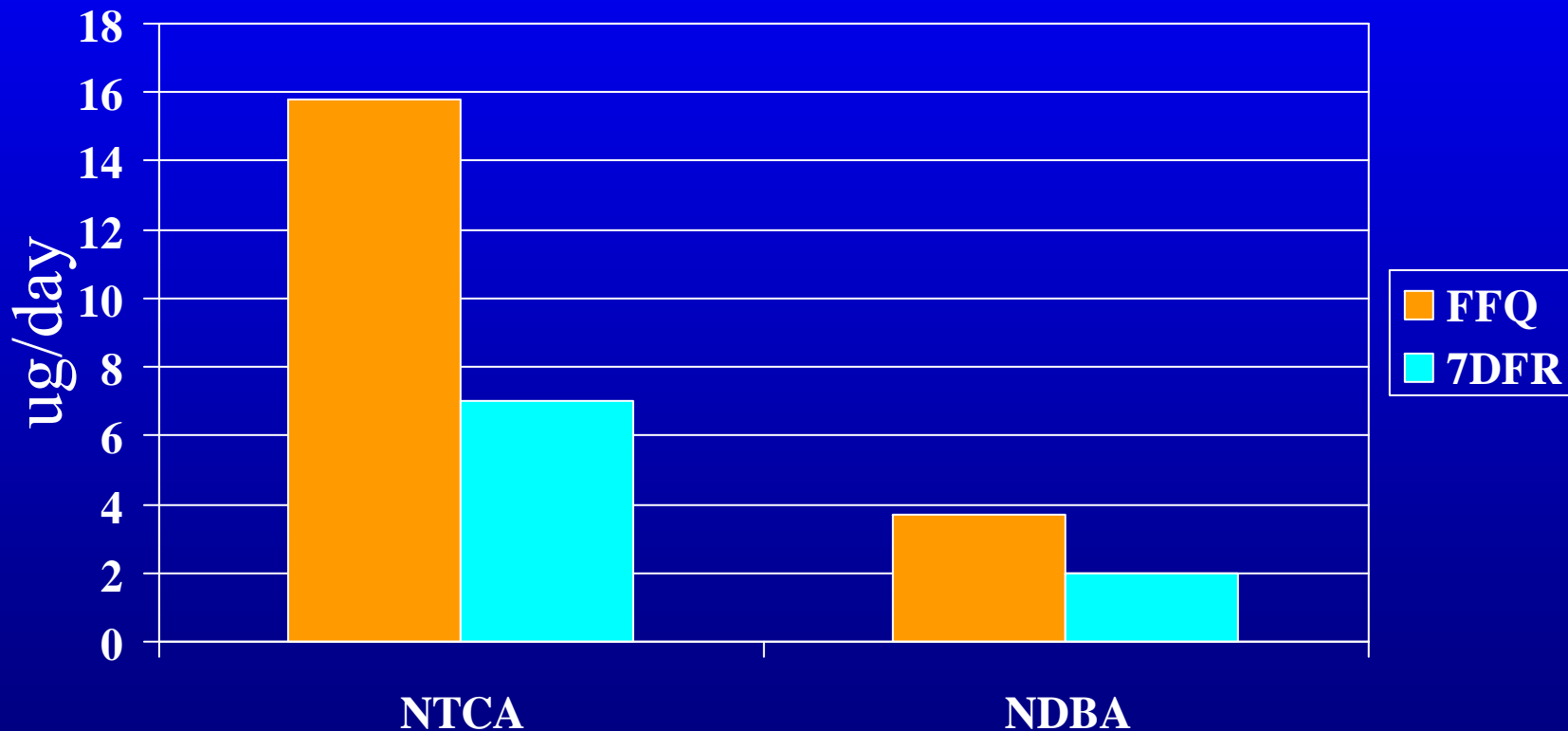
Frequency of Intake (N of persons) Food Groups FFQ vs. DFR

Agreement DFR vs. FFQ

High Sources of N-Nitroso Food Groups

Food Group	P value*
Ham	0.553
Wine	0.0001
Hot Dog	0.0001
Bacon	0.03
Sausage	0.03
Beef	0.663

*McNemar test

Advantages of N-Nitroso database

- More inclusive for all compounds (↑Number)
- More inclusive of all food sources (↑Number)
- Completed linkage of database to FFQ
- Verification of values in FFQ and DFR
- Validation of values and food sources in FFQ compared to DFR

Limitations of Database

- Many compounds and foods have not been assayed

- N-nitroso value may represent the maximum amount absorbed, but there are differences in metabolism, metabolic activity and action

- N-nitroso values may be based on only one analysis

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