

Professional Development Course
Case Studies in Biological Monitoring Workshop
Prepared for NASA by Larry K. Lowry, Ph.D
for a NASA program, June 7, 2010, San Antonio, TX

Course Description

The objective of the "Case Studies Workshop in Biological Monitoring" PDC is to encourage the appropriate use of biological monitoring techniques. The course presumes the students have some experience in biological monitoring. The goal of this course is to explore specific applications of biological monitoring, including interpretation of results in light of existing guidelines. The course will present topics to enable industrial hygienists and medical professionals to implement biological monitoring programs. The format of the course will include a review of pertinent biological monitoring topics followed by breakout groups in which case studies will be analyzed. Case studies will be real-world practical examples that will challenge the practicing industrial hygienist. Each case study scenario will include pertinent information. The attendee will develop a solution for the case scenario.

Learning Objectives

This is a practical, hands on 4-hr workshop in the application of biological monitoring. By the end of the workshop, participants should be able to:

- (1) Recognize the importance of a multidisciplinary team approach to exposure assessment with biological monitoring
- (2) Select the most appropriate biological monitoring determinant or marker and understand the factors influencing the selection of that marker
- (3) Discuss pre-analytical issues of importance in biological monitoring such as sampling, sample preservation, sample stability, and the importance of sample integrity on results
- (4) Discuss laboratory issues of importance such as quality assurance, proficiency testing, limits of detection, interferences, etc.
- (5) Gain insight in the interpretation of biological monitoring results, both in the presence of guidelines as well as in the absence of interpretation guidelines.
- (6) Understand the advantages and limitations of biological monitoring using real-world examples
- (7) Understand the importance of risk communication in the reporting of biological monitoring data to health professionals, workers, and management.

Course Outline

INTRODUCTORY LECTURE (15 min): Brief review of the basics of biological monitoring, significant issues associated with biomonitoring as well as successes, and ethics.

LECTURE ON SPECIFIC CHEMICALS TO BE DISCUSSED IN BREAKOUT SESSIONS (1 hr): Use and potential exposure to the specific chemicals, pharmacokinetics of exposure to specific chemicals, specimen collection, laboratory methods, including contamination control and proficiency testing, industrial hygiene assessment of data, pitfalls and caveats of the laboratory findings, assessment of possible means to reduce exposure, and interpretation of biomonitoring results utilizing guidelines, such as BEIs, and interpretation in the absence of guidelines, discussion of confounders such as smoking, ethanol, second jobs, hobbies, diet, and other exposures,

BREAKOUT SESSIONS (FOUR CASES, SELECT ONE) (1 hr 30 min): Benzene, Carbon monoxide, Arsenic, or Carbaryl.

PRESENTATION OF FINDINGS TO THE GROUP AND DISCUSSION.(50 Min)

Instructional Methods

The course will begin with a general lecture on biological monitoring. Then there will be a series of four lectures on specific biological monitoring topics, benzene, carbon monoxide, arsenic and Carbaryl. Each topic will address relevant biological monitoring information on these four compounds. The class will then breakout into four roundtable groups where they will be given a specific case on one of the four compounds discussed above. Each group will discuss the case, and develop a plan of action to address the questions asked. Pertinent reference material will be provided to each group. At the end of the breakout session, a representative from each group will present the proposed solution to the scenario to the whole group. Discussion will follow.

Your Instructor

Dr. Lowry received a Ph.D. in Comparative Biochemistry and for twenty years, served as the NIOSH expert on the application of biological monitoring to the health hazard evaluation program. He has been a member of the ACGIH BEI committee since its inception and is current chair. He has been involved nationally and internationally in biological monitoring most recently as a technical advisor to the WHO. He is active in International Commission on Occupational Health, being a member of the Scientific Committees on Rural Health and on Occupational Toxicology. Dr. Lowry is currently Professor, Occupational Health Sciences, The University of Texas Health Science Center at Tyler and is the Director, Graduate Environmental Health Programs. He also directs the Southwest Center for Pediatric Environmental Health.

Schedule

- 8:00 am Course introduction and logistics
- 8:05 am introductory lecture
- Brief review of the basics of biological monitoring
 - Significant issues associated with Biomonitoring
 - Ethics
- 8:40 am Lecture on specific chemicals to be discussed in breakout sessions
Benzene
Carbaryl
Carbon Monoxide
Arsenic
Topics to be discussed:
- Use and potential exposure to the specific chemicals
 - Pharmacokinetics of exposure to specific chemicals
 - Specimen collection
 - Laboratory methods, including contamination control and proficiency testing
 - Industrial hygiene assessment of data
 - Pitfalls and caveats of the laboratory findings
 - Assessment of possible means to reduce exposure
 - Interpretation of biomonitoring results utilizing guidelines, such as BEIs, and interpretation in the absence of guidelines
 - Discussion of confounders such as smoking, ethanol, second jobs, hobbies, diet, and other exposures
- 9:40 am Breakout sessions (Four cases, select one) Specific handouts will be provided.
Benzene (occupational)
Carbaryl (occupational)
Carbon monoxide (occupational and environmental)
Arsenic (environmental)
- 10:00 am Break
- 10:15 am Continue with breakout sessions
- 11:00 am Presentation of findings to the group and discussion (Each of the 4 groups would have 15 minutes for their group presentation questions)
- 12:00 pm Course conclusion and evaluation

What is Biological Monitoring?

- Systematic collection of body fluid or tissue
blood, urine, breath, saliva, hair
- Analysis for parent compound, metabolite, or other biomarker of exposure or effect
- Gives an indication of exposure /body burden
- Complement to environmental monitoring

Relationship to Exposure and Body Burden

Sample and determinant must reveal information about absorbed dose

Venous blood: drains all internal organs

Voided urine: reflects composition of blood, contains metabolites

Breath: volatile agents in blood equilibrate with alveolar gas; alveolar gas difficult to obtain

Justification for Biological Monitoring

Additional information needed on individual exposure:

- effects of physical work rate,
- personal protective equipment,
- idiosyncratic metabolism,
- non-occupational exposure

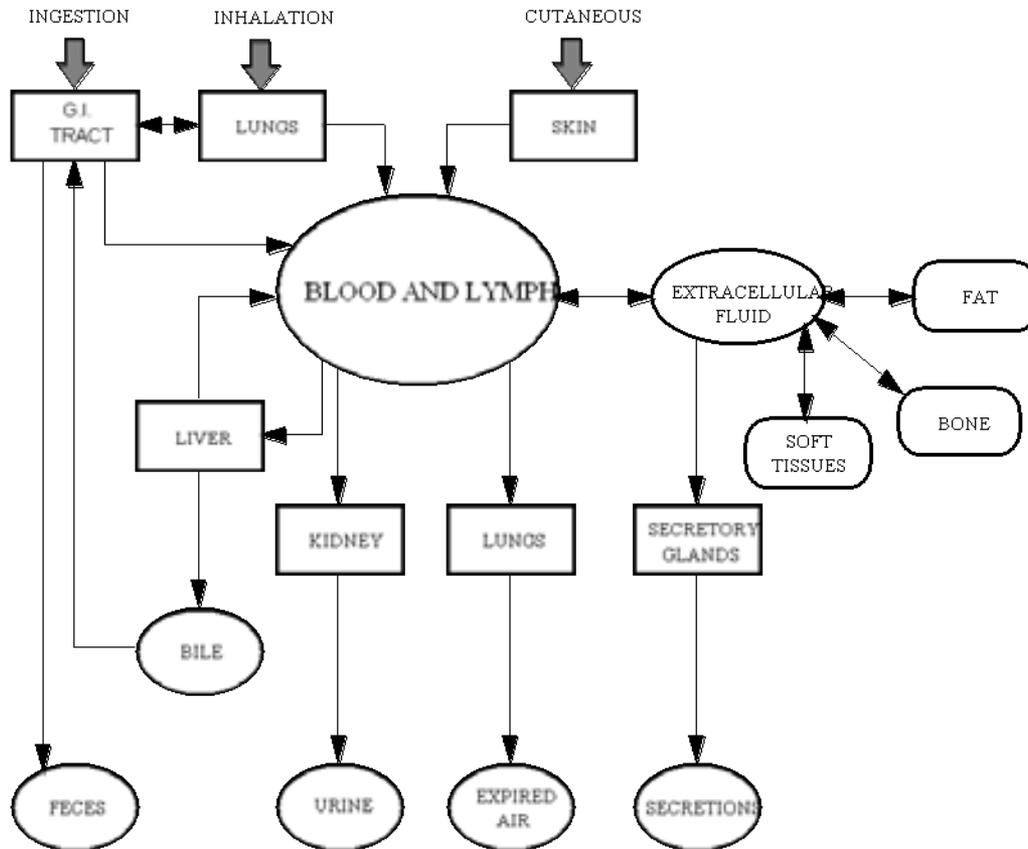
Data on early biological effects
may be obtained

Role of Toxicokinetics in Interpretation

Toxicokinetics: time-dependent processes affecting the relationship between external exposure and dose of agent delivered to the target organ or tissue

Toxicodynamics: processes that mediate biochemical response at target site: altered cell function, cell death, DNA adduct formation

Distribution Pathways



Analytical Issues

Breath: high levels of CO_2 (50,000 ppm), water vapor (40,000 ppm) may interfere

Background from non-occupational exposure: hobbies, diet, consumer products

Progressive lowering of TLV/PEL: background levels become very significant fraction of total dose.

Selection of Appropriate Indicator

What biological specimen should be taken?

What indicator species should be analyzed?

When should specimen be collected?

Further considerations

Sample timing relative to end of exposure

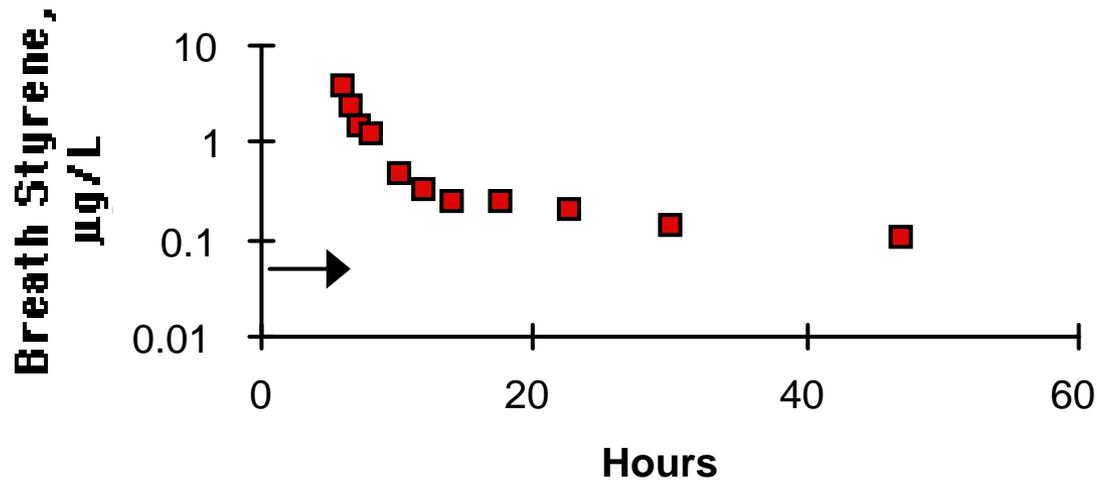
Volatile solvents: rapid elimination in early post-exposure period may lead to serious errors

Later samples reflect adipose tissue storage and long-term average

Heavy metals: slow elimination means sampling can be done at any convenient time, eg cadmium and lead

Sample Timing relative to Exposure

Styrene in breath after 6 hr at
80 ppm (Ramsey, 1980)



More considerations

Influence of personal characteristics

Physical work rate - enhances absorption

Genotype - inherited differences in metabolic capability

DNA analysis reveals presence of genetic variant: potential

Phenotype - expressed genetic differences

Eg induced enzyme activity, "slow acetylators"

Ethical Considerations

1. Potential Invasion of Privacy
medical procedures, safety, informed consent
careful identification of target compound(s)
2. Results may be treated as medical records
confidentiality, data access for risk management
3. Obligations to Employees - reporting

Biomonitoring Records

Employer must retain for 30 years
after termination

Must be made available to:

- workers
- authorized worker representative
- authorized OSHA representative

Annual summary: injury/illness,
compensation as part of OSHA
recordkeeping standard - link to
biomonitoring data may be needed

Positive attributes

May reveal unsuspected exposures

May identify atypical persons at risk

May reveal interactions among chemicals

May provide useful guidance for individual prescription medications

Negative Attributes

Biomarker may appear after damage has occurred

Time-dependence: cause-effect relationship difficult to know

Inter-individual differences may be substantial

Cost and (perhaps) liability issues

Arsenic Uses

"Arsenical" insecticides

Wood preservation: chromated
copper arsenate (CCA)

Semiconductor doping agent

Glass manufacturing

Sulfuric acid manufacturing

Arsenic Potential Exposures

Primary production - refining of
arsenic ores: arsenopyrite, realgar,
auripigment

Smelting of non-ferrous ores rich
in arsenic, especially copper ores

Semiconductor fabrication

Timber industry

Drinking water, seafood

Pharmacokinetics

Intake: inhalation of water-soluble forms: arsenic trioxide, pentoxide; ingestion of oxides, methylated forms, arsenobetaine, arsenocholine, arsenosugars.

Metabolism: inorganic arsenic converted to methylated forms: methyl arsonic acid, dimethyl arsinic acid

Pharmacokinetics

Elimination: Dimethyl arsinic acid plus small amounts of methyl arsonic acid and oxides; arsenobetaine and choline excreted unchanged; arsenosugars excreted as dimethyl arsinic acid.

Single dose (As_2O_3) is eliminated in three phases with half lives of 2 days, 9.5 days, 38 days.

Pharmacokinetics

Routes of elimination:

Urine is predominant

Hair, nails, sloughed epidermal cells

Specimen Collection

End of shift at end of work week

Avoid contamination from ambient environment

Urine analysis requires measurement of creatinine to assess level of physiologic dilution.

Laboratory Methods

Preliminary separation : HPLC, ion exchange

Hydride generation: reaction with sodium borohydride, produces volatile hydride only from inorganic arsenic and methylated forms.

Proficiency testing and evaluation: accuracy and precision are acceptable at urine concentrations $\geq 5 \mu\text{g/L}$.

Industrial Hygiene Assessment

Airborne exposure measurements should use a size-selective method. The respirable particulate fraction is preferred: use a sampling method that preferentially collects particles with aerodynamic diameters less than $4 \mu\text{m}$.

Current TLV: $10 \mu\text{g}/\text{m}^3$, TWA, A1
carcinogen

Analytical Issues

Background from non-occupational exposure: hobbies, diet, consumer products

Progressive lowering of TLV/PEL: background levels become very significant fraction of total dose.

Laboratory Findings

Speciation depends on ability to separate the compounds, or to place them accurately in groups:

"organic" arsenic: arsenobetaine, arsenocholine, arsenosugars

"inorganic" arsenic: trioxide, pentoxide
methylated oxides: MMA, DMA

Total arsenic: generally the sum

Interpretation of Biological Monitoring

BEI: 35 μg inorganic As/L in urine,
end of shift at end of work week.

Creatinine concentration in same
specimen should be between 0.3
g/L and 3.0 g/L

Results outside this range should
be discarded.

Other tissues: blood, hair, nails

Confounding Factors

Lack of analytical specificity

Non-occupational exposures -

 dietary intake - seafood

 drinking water - geographic variation

 smoking- may be small contribution?

 air pollution - industrial emissions

General guide for communication of results

Distinguish uncertainty from variability; focus on what is being done to reduce the former.

Explain what we know and with what accuracy and confidence.

Explain what we don't know and why.

Explain what we could know with more resources.

Explain what we must know to take action.

Use comparisons to reference values or guidelines where they exist.

Benzene Biomonitoring Case Studies Workshop in Biological Monitoring

**American Industrial Hygiene Conference
and Exposition**

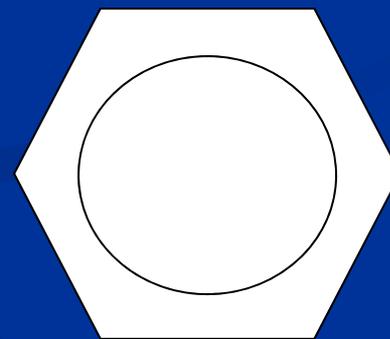
**Lee M. Blum, Ph.D.
National Medical Services
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Benzene

- **Physical/Chemical Characteristics**
- **Health Effects**
- **Sources of Potential Exposures**
- **Routes of Exposure**
- **Pharmacokinetics**
- **Specimen Collection**
- **Interpretation of Laboratory Results**

Benzene

- Clear, colorless, highly flammable, non-polar liquid with a pungent aromatic odor
- Vapor pressure: 100 mm Hg at 26.1°C, highly volatile



Health Effects

- **CNS**

Euphoria, drowsiness, dizziness, headaches, fatigue, irritability, confusion, unconsciousness

- **Cardiac**

Sensitization and arrhythmias

- **Pulmonary**

Bronchial irritation, cough, hoarseness, pulmonary edema

- **Dermal**

Erythema, blistering, dermatitis

- **Hematologic**

Blood dyscrasias, aplastic anemia, and specific types of leukemia

Sources of Potential Exposures

- Formed from natural processes, such as volcanoes and forest fires.
- A component of cigarette smoke.
- Industrially used primarily as an intermediate in the synthesis of other chemicals.
- Found in a number of hazardous waste sites in the U. S.

Routes of Exposure

- **Inhalation**

- **Dermal**

 - Gaseous form

 - Liquid form

- **Ingestion**

Pharmacokinetics

- **Absorption**

- ~Primarily through inhalation

- **Distribution**

- ~From animal studies, maximal concentration in tissues within 3 hours.

- ~Highly fat soluble

- **Metabolism**

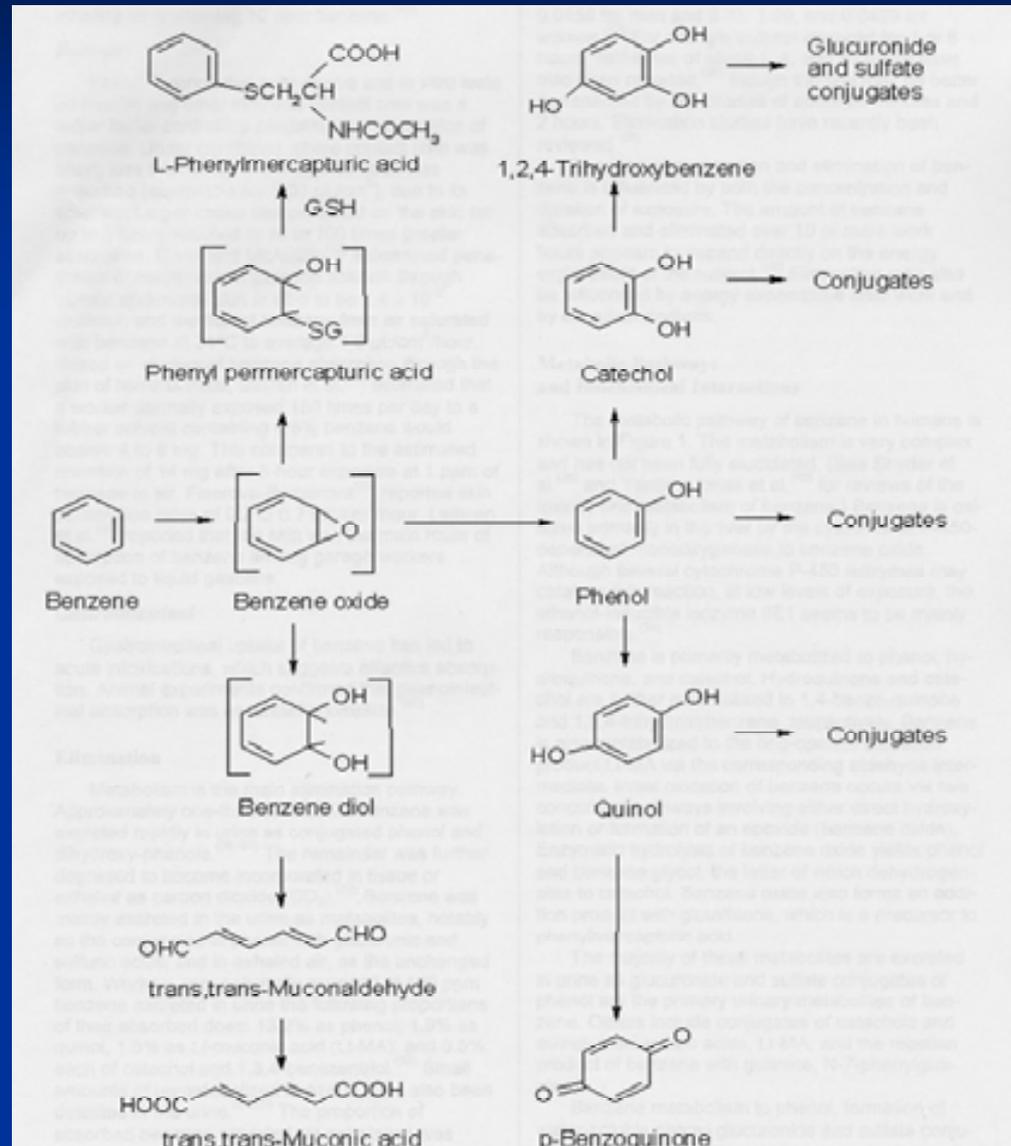
- ~Main elimination pathway

- **Elimination**

- ~Exhaled breath as unchanged benzene

- ~Urine as metabolites

Metabolism



Specimen Collection

- **Biomarkers of occupational exposure to benzene**

Measure unchanged benzene in:

Blood

Sensitive and specific

Invasive and affected by sampling time

Breath

Reflects concentration in the blood

Non-invasive

Same interpretive problems as testing blood

Sampling and analysis is not yet standardized

Urine

Lower dependence on sampling time

Non-invasive

Fluctuations in urinary flow

Insufficient data to interpret benzene result

Specimen Collection (cont'd)

Measure urinary metabolites:

Determination of phenol and some of the other metabolites (e.g. phenol, catechol, quinol, and 1,2,4-benzene-triol)

Non-specific

Not sensitive enough for low level exposures

trans,trans-Muconic Acid (t,t-MA) and S-Phenylmercapturic Acid (S-PMA)

At the end of shift, indicators of exposure during the last shift

Influenced by long-term retention of benzene

Interpretation of Laboratory Results

■ Phenol - Urine

Normally non-exposed: < 10 mg/L

0.5 - 4.0 ppm benzene: < 30 mg/L

25 ppm benzene : avg 200 mg/L

OSHA limit: 75 mg/L

Interpretation of Laboratory Results (cont'd)

■ Phenol - Urine

Endogenous production by bacteria in GI

Effected by antibiotics, GI disorders, liver and kidney diseases

Exogenous exposures

Chemical and industrial

Disinfectants

Medicinals

Foods

Tobacco smoke

Interpretation of Laboratory Results (cont'd)

■ t,t-MA - Urine

ACGIH BEI: 500 $\mu\text{g/g}$ creatinine (end of shift)

Non-exposed, non-smoker:

geo mean = 67 $\mu\text{g/g}$ creatinine

(95% upper limit: 296 $\mu\text{g/g}$ creatinine)

Non-exposed, smoker:

geo mean = 207 $\mu\text{g/g}$ creatinine

(95% upper limit: 563 $\mu\text{g/g}$ creatinine)

Interpretation of Laboratory Results (cont'd)

■ t,t-MA - Urine

Tobacco smoke

over 50 cigarettes/day: 450 $\mu\text{g/g}$ creatinine

Sorbic Acid 6 - 30 mg/day

Non-smokers: Accounts for 10 - 50% of background

Smokers: Accounts for 5 - 25% of background

Interpretation of Laboratory Results (cont'd)

■ t,t-MA

Tobacco smoke

Sorbitol absorption (sweetening agent)

Dermal exposures

Interindividual differences in kinetics

Pregnancy

Concomitant exposure to toluene

Interpretation of Laboratory Results (cont'd)

■ S-PMA - Urine

ACGIH BEI:

25 $\mu\text{g/g}$ creatinine (end of shift)

Non-exposed, non-smokers:

1.99 \pm 0.29 $\mu\text{g/g}$ creatinine

Non-exposed, smokers:

3.61 \pm 0.57 $\mu\text{g/g}$ creatinine

Interpretation of Laboratory Results (cont'd)

■ S-PMA

Tobacco smoke

Dermal exposures

Interindividual differences in kinetics

Concomitant exposure to other aromatic hydrocarbons

Interpretation of Laboratory Results (cont'd)

Possible non-occupational exposures to benzene

Two major sources

- ~Vehicle emissions (from gasoline evaporation and from exhausts)
- ~Smoking (especially side stream smoke which has been shown to contain more benzene than mainstream smoke).

Other sources

- ~Contaminated ground water

References

1. Agency for Toxic Substances and Disease Registry (ASTDR). 1997. Toxicological Profile for Benzene. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.
2. ASTDR. 1998. Toxicological Profile for Phenol. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.
3. 29CFR1910.1028. Title 29: Department of Labor, Chapter XVII: OSHA, Part 1910: Occupational Safety and Health Standards. Revised July 1, 1998. U.S. Government Printing Office.
4. ACGIH BEI for Benzene
5. Toxnet Benzene <http://toxnet.nlm.nih.gov>

Biomonitoring Carbaryl Pesticide Case Studies Workshop in Biological Monitoring

American Industrial Hygiene Conference and Exposition

**Lee M. Blum, Ph.D.
NMS Labs
Willow Grove, PA**

Carbaryl Pesticide

- Carbaryl
 - General use carbamate pesticide
 - Processed by many formulators into over 1000 different products
 - Controls over 100 species of insect on citrus, fruit, cotton, forests, lawns, shade trees and other crops.

Carbaryl Pesticide

- Exposure
 - General population
 - Residues on food is the major source of intake
 - Pesticide use in the home and recreation areas
 - During periods of application it can be occasionally found in the environment, such as in surface water or reservoirs.
 - Workers
 - During manufacture, formulation, packing, transportation, storage, and during and after application.

Carbaryl Pesticide

- Pharmacokinetics

- Absorption

- Inhalation
 - Ingestion
 - Dermal

- Metabolism

- The principle metabolic pathways are hydroxylation and hydrolysis resulting in numerous metabolites and subjected to conjugation with water soluble compounds in the body.
 - The principle human metabolite is 1-naphthol

- Excretion

- Mainly via the urine as 1-naphthol
 - Maximum level of 1-naphthol in applicators and formulators reached during the work day was in the late afternoon

Mechanism of toxicity

Inhibition of cholinesterase activity

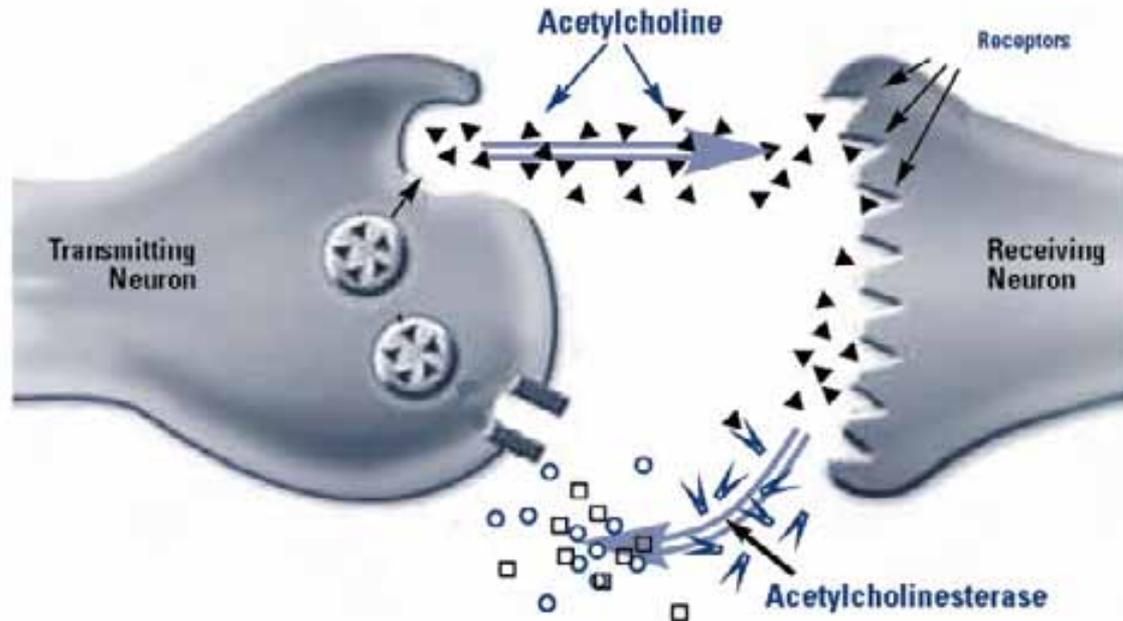


Fig. 1. After signalling, acetylcholine is released from receptors and broken down by acetylcholinesterase to be recycled in a continuous process.

Inhibition is not long lasting and reversible within a few hours

Health effects result from accumulation of acetylcholine at nerve endings

- Health effects

- Increased bronchial secretion
- Bronchoconstriction
- Difficulty in breathing
- Excessive sweating
- Salivation
- Lacrimation
- Pinpoint pupils
- Abdominal cramps
- Bradycardia
- Fasciculation of fine muscles
- Tachycardia
- Headache
- Dizziness
- Anxiety
- Mental Confusion
- Convulsions
- Coma
- Respiratory depression

Cholinesterase

- Three (3) basic types of cholinesterase
 - “True” cholinesterase
 - Found in the brain
 - Acetylcholinesterase
 - Found in the red blood cells
 - Closely related to “true” cholinesterase
 - Therefore a good biomarker of cholinergic effects
 - Pseudocholinesterase
 - Found in plasma

Medical surveillance

- Pre-exposure examinations and baseline acetylcholinesterase levels should be established
 - Considerable inter-individual variation
 - Evaluate the degree of inhibition in an individual
 - Variety of methods and units used in laboratory testing; therefore, should use the same laboratory for all subsequent testing

Medical surveillance

- Over-exposure to carbaryl can be monitored by the measurement of red blood cells acetylcholinesterase activity
 - Enzymatic activity returns to normal within a few hours
 - Blood samples need to be collected and tested within 4 hours after exposure
 - ACGIH BEI for Acetylcholinesterase inhibiting pesticides is 70% of individual's baseline activity.

Medical surveillance

- Levels of 1-naphthol in the urine have been used as a biological indicator of exposure.
 - No clearly established relationship between external exposure of carbaryl and urinary 1-naphthol level.
 - However, when the 1-naphthol level in urine does not exceed 10 mg/L at the end of the exposure period, the risk of signs and symptoms of intoxication is low.

Medical surveillance

- The 1-naphthol hazard level is >10 mg/L and the symptomatic level is >30 mg/L (WHO, 1994)
 - U.S. population 6-59 years (n = 1998)
1999-2000 NHANES
 - Geo mean = 0.002 mg/L
 - 95% of population = 0.012 mg/L
 - 1-Naphthol also a metabolite of Naphthalene

References

- Acetylcholinesterase Inhibiting Pesticides BEI Document. ACGIH. Cincinnati, OH. 2001
- Carbaryl. Environmental Health Criteria 153. International Program on Chemical Safety. World Health Organization, Geneva, 1994; accessed www.inchem.org/documents/ehc/ehc/ehc153.htm
- 1-Naphthol. NHANES 1999-2000 Survey. CDC. 2003

Carbon Monoxide Biomonitoring

Case Studies Workshop in Biological
Monitoring, PDC 602, Toronto, 2009

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Carbon monoxide

- ◆ Use and potential exposure
- ◆ Pharmacokinetics
- ◆ Specimen collection
- ◆ Methods of analysis
- ◆ Quality control
- ◆ Interpretation of results and confounders

Uses and exposure

- ◆ A product of incomplete combustion
 - Found in many workplaces as well as in environment
- ◆ A product of cigarette smoking
- ◆ A metabolite of dichloromethane (methylene chloride)
- ◆ Endogenous product of metabolism
- ◆ Inhalation is sole route of exposure

Pharmacokinetics of CO

- ◆ Preferential binding of CO (200 x greater than O₂ to hemoglobin)
- ◆ Rapid absorption into the blood reaching a steady state after 3 hrs.
- ◆ Elimination half-life of 5 hrs following cessation of exposure

Specimen collection

- ◆ Venous heparinized blood specimen collected, mixed and kept in dark
- ◆ Specimen collection within 10-15 minutes after end of exposure
- ◆ Place in crimp-top vials and seal
- ◆ Keep cold and analyze promptly
- ◆ Cannot analyze clotted blood

Methods

- ◆ Standard ER methods do not have adequate sensitivity to distinguish smokers from non-smokers.
- ◆ Variety of sensitive methods available
 - Oximeters (automated spectrophotometric instruments at three wavelengths)
 - Gas chromatography plus hemoglobin

Quality control issues

- No interlaboratory testing schemes
- Utilize controls provided by manufacturer
- Prepare standards by equilibrating known CO standard gas in blood
- Always include smokers and non-smoker controls in your specimen set sent to laboratory.

Interpretation issues

- ◆ Other sources of CO
 - Smoking cigarettes
 - ◆ 1 pack/day – 5-6% COHb
 - ◆ 2 packs/day 7-9% COHb
 - Cigars – up to 20% COHb
- ◆ Urban pollution – 1-2% COHb
- ◆ Commuting on urban freeways – >5% COHb
- ◆ Endogenous production - 1%
- ◆ BEI – 3.5% COHb

Methylene chloride (dichloromethane)

- ◆ Metabolized to CO
- ◆ Pharmacokinetics – delayed by metabolism to CO with a half-life of about 12-14 hrs.
- ◆ 50 ppm exposure to MeCl for 8 hrs – expect 2% COHb

Interpretation

- ◆ Background levels in non-smokers usually less than 2%
- ◆ Smokers levels dependent on amount of smoking and time since last smoke
- ◆ ETS? An influence of unknown proportion

Suggested approach to BM

- ◆ Pre- and post-shift specimens for COHb
- ◆ Smoking history recorded
- ◆ Other exposures recorded
 - Methylene chloride, urban pollution, commuting, etc.
- ◆ Time of last exposure and lag before specimen collection
- ◆ Attention to specimen collection detail
- ◆ Prompt analysis

Alternatives

◆ CO in end-exhaled air

- Not widely used
- Subject to workplace contamination
- Specimen collection tricky
- Influenced by water content of expired air
- Analysis by Miran or other IR direct reading instrument
- Kinetics are different

References

- ◆ ACGIH BEI for Carbon Monoxide
- ◆ Common texts on biological monitoring
 - Lauwreys and Hoet, 3rd edition, 2001

Inorganic Arsenic Case Study

The Situation

The Old Lace Specialty Chemical Company (headquarters in St. Paul, Minnesota) has retained you to advise them on exposure assessment of the workers in their plants at several locations and one planned location around the world. The company refines arsenopyrite ore to inorganic arsenic compounds for the semiconductor industry. The principal product is arsenic trioxide, but a significant fraction of the product is arsenic pentoxide. Because of the obvious health risks associated with excessive exposure to these compounds, exposure monitoring and some employee health monitoring are desired by the management. You have been hired to help design and implement a biological monitoring program, to be operated in conjunction with routine air monitoring in the workers' breathing zones.

The company has refining facilities in St. Paul, in Port Arthur, Texas (on the Gulf of Mexico) and in Osaka, Japan. It is giving serious consideration to purchasing an existing arsenic refining plant located near Dakka, Bangladesh as well. A preliminary biomarker survey was recently conducted by another consulting firm, who reported urinary concentrations of total arsenic (that is, all arsenic-containing compounds in urine) from one-time specimens collected from "several" workers (the actual number of samples is not available) at each of the three existing facilities. The results are given below, together with some additional data obtained from local environmental regulatory authorities.

Location	Drinking Water Arsenic, mean $\mu\text{g/L}$	Typical Dietary Habits of Workers	Total Arsenic in Urine, mean $\mu\text{g/L}$
Port Arthur	less than 10	some seafood: mussels Predominant	200
St. Paul	less than 10	modest seafood	35
Osaka	less than 10	much seafood, including Seaweed	500
Dakka	55, some samples Up to 200	unknown	unknown

Old Lace management is very concerned about the results of the survey, since they know that the ACGIH Biological Exposure Index is $35 \mu\text{g/L}$, but they were not given much advice about how to interpret the data with reference to the BEI. Management plans to conduct regular air monitoring for particulate phase arsenic at each location (about 35 workers at each facility have regular exposure to airborne arsenic), and your task is to design a biological monitoring plan to complement air monitoring.

The specific questions you must address in your plan are:

1. Which tissues or body fluids should be sampled? Choose any or all from venous blood, exhaled air, voided urine, scalp hair, fingernail clippings.
2. a. Can total arsenic concentration (all species) be used, or should only specific arsenic species be analyzed in these samples?
b. If speciation is required, which chemical species of arsenic should be analyzed?
3. When, relative to the work shift, should the samples be collected from the workers?
4. What would be the expected mean values of concentration in urine, at each location, if the airborne arsenic levels were found to be $10\mu\text{g}/\text{m}^3$ (the current TLV), or $50\mu\text{g}/\text{m}^3$?
5. What non-occupational sources of arsenic will need to be ascertained at each location?
6. a. What long-term health effects might be expected in workers with continued excessive occupational exposure to inorganic arsenic?
b. What medical monitoring would you recommend, if any, to provide useful information to workers, health care professionals, and management?
7. Explain why the results of the preliminary survey given above may or may not suggest the workers are exposed to excessive airborne arsenic in the work places.

Useful Background Information

Arsenic in drinking water occurs primarily as the water-soluble inorganic compounds, arsenic trioxide and arsenic pentoxide. Occupational exposures in most industrial uses of arsenic are to the same two inorganic arsenic compounds, plus elemental arsenic and arsine, AsH_3 . Most living organisms metabolize inorganic arsenic taken up from water to the methylated compounds – methyl arsonic acid and dimethyl arsinic acid. Marine organisms further transform these compounds to the organic forms: fish and crustaceans contain significant amounts of “organic” arsenic compounds, primarily arsenobetaine, arsenocholine and arsenosugars, but only trace amounts of inorganic arsenic or methylated arsenic compounds. The exception is mussels, which contain considerable amounts of dimethyl arsinic acid as well as arsenobetaine and arsenocholine.

In the typical arsenic refinery producing inorganic arsenic compounds, airborne particulate matter has a mass median aerodynamic diameter of 3 to 5 μm , with a geometric standard deviation of 2.0 to 2.3.

In humans, arsine is the most toxic of the inorganic forms, arsenic trioxide is intermediate, and arsenic pentoxide is less toxic. The methylated metabolites are even less toxic, and arsenobetaine and arsenocholine are of very low relative toxicity. The biological half time for elimination of arsenobetaine and arsenocholine is less than 20 hours; the inorganic forms of arsenic are eliminated in a tri-phasic process with half times of 2 days, 10 days, and 38 days. Therefore, with daily exposure accumulation in the body is likely.

Analysis of arsenic in aqueous samples by hydride generation followed by atomic absorption spectroscopy without special treatment yields the total of all arsenic chemical species, including elemental, inorganic, methylated and organic compounds. Separation of the species requires prior treatment using HPLC, ion exchange or other high performance technique, followed by

hydride generation – AAS. Good quality analytical procedures used for urine can separate the sum of inorganic and methylated species from arsenobetaine, arsenocholine and the sugars.

References

ACGIH. Arsenic and soluble inorganic compounds. Documentation of the Biological Exposure Index. Cincinnati, OH: American Conference of Governmental Industrial Hygienists (1999).

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Case study for benzene:

It's winter time in a northern industrial company called Rubbersyn, Inc., which manufactures plastic household products. Several workers throughout the plant have been complaining of headaches, dizziness, and fatigue. Some had complained of an unusual increase in the number of colds and flu as well as feeling fatigued. They've been taking several over-the-counter medications, such as lozenges, and home remedies, including tea and honey, for the relief of these flu-like symptoms. Some of the workers have even used germicidal disinfectants throughout the work areas to help control the spread of these cold and flu type infections.

One day during the installation of new equipment, several drums of a liquid solvent were damaged creating a chemical spill of several gallons of liquid. Examination of the inventory records found that the liquid solvent contained in these drums consisted of benzene.

The clean up of the spill was promptly performed, and you as the plant industrial hygienist initiated air monitoring and medical surveillance on the 5 workers who were in the area of the spill, including urinary phenol testing at the end of the employees' work shift.

The results of the urinary phenol testing returned within 72 hours of the incident showed the levels to be above the 75 mg/L OSHA limit. You assumed the elevated phenol findings were the result of the chemical spill and you re-tested the same individuals. Again, each of the urinary phenol tests came back above 75 mg/L. You questioned the laboratory results, as the medical personnel at your facility have had trouble with the lab in the past on other issues. About a week post-accident, in addition to collecting urine samples from the same 5 individuals, you also obtained specimens from several other employees within the same department, but not in the area of the benzene spill to use as a control population.

The lab results from this third round of testing found that the values from the original 5 workers continued to be above the OSHA exposure limit, and those from the control population were also elevated.

You're somewhat perplexed by the urinary phenol levels of the 5 exposed employees about 7 days following the accident and of the unexposed workers since the air monitoring results declined to about 1.0 ppm within hours of the incident and then below 0.5 ppm within 24 hours. Review of records of air monitoring results from semi-annual testing in this area over the past couple of years have found the TWA to be less than 0.5 ppm with the last test performed about 3 months ago. You had no knowledge of any benzene spills since the time of your employment, 2 years ago, and have not yet established a biomonitoring program for benzene exposure in the employees at this facility, as OSHA requires urinary phenol testing only subsequent to a benzene spill.

What do you do now?

1. Do you continue to monitor the air for benzene? Why or why not?
2. Do you continue testing for urinary phenol? If you do continue testing for phenol in urine, do you switch to a new lab or do you continue using the same lab and why? How do you assess the lab testing, either at the same lab or at a new one?
3. With continued testing, you find that the urinary phenol tests are still elevated, what do think is (are) the cause(s)?
4. What additional testing (environmental and biological) can be performed? How do you evaluate the reliability of the test results? Are there any limitations in the findings of these new tests?
5. How do you satisfy governmental (i.e., OSHA) regulations in biomonitoring for benzene exposure with the known limitations in the test, and the budgetary constraints Rubbersyn, Inc. has on laboratory testing?

Carbaryl case:

A short time after lunch following a morning household chores, a 34-year-old previously healthy woman, who is a mother of three children woman, suddenly developed severe nausea, vomiting, and diarrhea. She called a neighbor for help who found her profoundly weak, with slurred speech, and complaining of a continual need to clear her throat because of abundant secretions. The neighbor called 911 for emergency assistance. When the paramedics arrived, they found the woman combative, displaying difficulty in breathing, incontinent of stool and urine, vomiting, salivating, and teary eyes. Her heart rate was low and her blood pressure stable. The paramedics administered medicine to increase her heart rate, blood pressure and respiration rate then transported her to a local hospital.

After several days in the hospital, the woman recovered from her illness; however, the doctor at the county health department calls you as a consultant because he suspects a chemical exposure with this woman from the signs and symptoms she initially displayed at the time the paramedics arrived at her house, her response to treatment, and the results of laboratory tests conducted while she was in the hospital. For example, her cholinesterase activity was measured as follows:

Cholinesterase Concentrations*

Time (Hours – days after onset of symptoms)	Plasma	Red Blood Cell
17:00 (4 hrs – day 0)	221	5.8
13:00 (48 hrs – day 2)	1878	...
11:00 (70 hrs – day 3)	2252	...
15:00 (98 hrs – day 4)	2193	...
18:00 (125 hrs – day 5)	2771	12.6

***Normal range, plasma (1700 to 5778 IU/L), red blood cell (11.2 to 16.7 IU/mL).**

The health department physician specifically wants to know the following:

1. What is the source of the chemical exposure?
2. Are others in the community affected by the exposure? If so, who?
3. Identify steps to control exposures and monitor their effectiveness.
4. To arrive at your conclusions, utilize biological monitoring data as part of your assessment.

The health department will provide you a nurse to assist you in gathering data including the collection of blood and urine samples, if necessary.

Further information provided by the patient included the following:

- She denied previous medical problems or taking medications. However, over the previous 3 months she had several similar episodes about once a week, one of

which required emergency treatment and resolved spontaneously over several hours.

- She also related that her children, especially the two year old, had periodic skin rashes and increased incidences of nausea, vomiting and diarrhea that she attributed to the usual colds children tend to get.
- Her husband works for Home Grown Fruits, a local farm known for its vineyards and fruit orchards that employs about 20 local residents from town.
- Home Grown Fruits utilizes various chemicals on its farm for fertilization and pest control. One of the pesticides used is carbaryl.
- Because of the economic downturn in the past year, the owner of Home Grown Fruits has cut costs by eliminating some labor positions resulting in consolidation of work duties including pesticide application, field re-entry and crop harvesting for all remaining employees. Employee perks on the farm such as employer purchased work boots and gloves, and free laundry services of employee work clothes also were abolished.

In your investigation, you have found that the local clinic has seen an increase in complaints of skin rashes and gastrointestinal symptoms including nausea, vomiting and diarrhea over the past few months among other women and children in the community, especially in those families whose husbands work at Home Grown Fruits.

From the information provided, you will need to respond to the concerns of the health department as outlined above. In your response, you were requested to use biological monitoring data as part of your assessment. In using biological monitoring data, you'll have to decide who to biomonitor and the biomarker(s) to use. Carbaryl is a carbamate pesticide that can inhibit the activity of the enzyme, acetylcholinesterase. There is a BEI (Biological Exposure Index) for acetylcholinesterase inhibiting pesticides. Carbaryl also metabolizes in the body to 1-naphthol, which is excreted in the urine and can be measured in a biomedical laboratory. A government funded study has evaluated urinary 1-naphthol levels found in the general U.S. population and the World Health Organization (WHO) cites a hazard level when urinary 1-naphthol concentrations are greater than 10 mcg/mL. Consider when you would collect specimens and how you would evaluate the results. You'll have to determine if either or both of these biomarkers have the required characteristics to evaluate exposure.

Carbon Monoxide Case Study

Background

You are the industrial hygienist (Joe Safety) for a large retail warehouse operation (Skymart) located in Columbus Ohio. The warehouse includes 10 acres of covered area with natural gas-fired space heating units to keep products from freezing in the winter. There are large roof-mounted fans in use to ventilate the warehouse. There are about 25 overhead doors associated with loading docks.

There are 250 employees within the operation, including 200 who are involved in moving palettes of merchandise with propane fueled industrial trucks (fork lift trucks). There are three 8-hour shifts with equal numbers of workers at each shift. The company has a public image of providing good support for its workers, both financially and from a safety and health perspective. The company president (Fred Goodview) frequently appears in TV ads and at local events to promote the company image. The company is not unionized but privately, there has been some unrest among some of the employees regarding working conditions and lack of cost of living pay raises within the last 2 years. About half of the workers smoke cigarettes, including smoking on the job. The company has an on-site nurse (Ms. Goodbody) and small clinic equipped to handle first aid and required OSHA medical surveillance programs. The company uses a local family practice physician (Dr. Sue Ellen Quack) to help with required medical surveillance and to conduct needed physical exams.

The company has generally had a good safety record according to their OSHA 300 logs, with the majority of recordable events associated with ergonomic issues followed by cuts, bruises, and an occasional broken bone from a fall.

Changes in Company

In the fall of 2000, as a result of 9/11 and other events, the company experienced a business slowdown as consumer confidence fell and retail business declined. In an attempt to adapt its operation to the changing times, the company president, Mr. Fred Goodview, in consultation with his CFO, Mr. Pennypincher, initiated a number of cost-saving programs. There was no consultation with Joe Safety on the impact of some of these cost-saving programs. The following is a list of some of these steps, which were fully implemented by October 15, 2000:

- Layoff of about 20% of the employees (50 layoffs), based on maximizing savings. (highest paid were first to go)
- Hiring of minimum wage part time employees (50) to do some routine loading and unloading, thus saving the company from benefits costs (total number of ITO were 180, including the 50 part time employees)

- Initiation of an energy conservation program to include turning off the ventilation fans in the winter and a consistent program to keep all overhead doors closed except when actually loading or unloading trucks.
- Cutbacks in the safety and health program by reducing the industrial hygienist and nursing staff to three days per week.
- Elimination of routine physical exams not required by OSHA

The response by the employees was predictable. The employees contacted the local union and began a union organization program to provide worker job protection and restore safety and health programs eliminated in the cost-cutting steps. There were a few job slowdowns during the time. Within a month, a fully functional union has been established with more than 80% of the employees belonging to the union. The union steward was Bubba Woopper.

In the midst of this increased labor-management struggle, the company president (Fred Goodview) continued to apply the good image spin to various news reports, never acknowledging that his actions may have precipitated this strife.

The problem

In December, Nurse Goodbody notified you, Joe Safety, that she had noticed an increased number of ITO (industrial truck operators) reporting to the clinic with headaches. She reported the increased number of clinic visits to Bubba Woopper and Fred Goodview. The verbal exchange between Fred Goodview (malingering employees) and Bubba Woopper (the company is poisoning us) led to the following charge to you, Joe Safety. "Get to the bottom of this now; your future with the company depends on resolving this issue. Keep this out of the local press."

Assessment of the situation

Your initial assessment of the situation revolved about the following issues:

- There was significant labor-management unrest; the headaches could be the result of that unrest. (Can headaches be caused by stress?)
- There did not seem to be any interest in determining compliance with the PEL for CO.
- There seemed to be more reported headaches among regular employees than temporary employees.
- There seemed to be more reported headaches among ITOs
- The ventilation fans, being shut off and the overhead doors kept closed may contribute an additional burden from carbon monoxide emissions from the industrial trucks. (Note: propane fueled industrial trucks do emit carbon monoxide).
- About half of the apparent target population (ITOs) are current cigarette smokers.

- The reports of headaches seemed to peak after about 3-4 hours of work.
- Measurement of carbon monoxide in the breathing zone of the ITOs may assess workplace contributions to carbon monoxide exposure but would not assess the impact of carbon monoxide from smoking on possible headaches.
- After consultation with Nurse Goodbody and Dr. Sue Ellen Quack, you determine that a biological monitoring program for carboxyhemoglobin might be the most cost-effective means to address carbon monoxide exposure and its relationship to headaches.
- A biological monitoring program of ITOs during the day shift should give a representative assessment of the problem. There are about 60 ITOs including 18 part time workers during the day shift. About 50% of employees and part time workers smoke cigarettes on the job.

The approach

Your charge by, Fred Goodview, was to get to the bottom of this issue. You propose to him a biological monitoring program that would determine if headaches could be caused by exposure to carbon monoxide. He counters and says “make sure that you distinguish between carbon monoxide exposure in the warehouse and carbon monoxide arising from the foolish smoking habits of the employees. Do this at minimum costs.”

Your questions

- Design a biological monitoring program to assess the degree of carboxyhemoglobin elevation and determine if the levels are high enough to cause headache. (Generally headaches occur at carboxyhemoglobin levels above 10%).
- Include a means to determine the contribution of smoking on carboxyhemoglobin levels.
- Should temporary employees be included in the study? How many workers should be included?
- The local hospital laboratory offers a colorimetric carboxyhemoglobin test for \$10 that is primarily used to evaluate fire victims brought to the emergency room. The limit of detection is about 10% carboxyhemoglobin. Should you use this lab and method to reduce costs?
- Biological Monitoring Resources, a specialty laboratory in industrial toxicology offers a sensitive method for carboxyhemoglobin with a detection limit of 1% carboxyhemoglobin. The cost is \$30 per specimen and the lab is located in Cincinnati, an hour drive from the warehouse. Should you use this lab?
- Who would draw the blood specimens and when would the samples be collected?
- Include in your plan the means to interpret the results and answer the two basic questions imposed by Fred Goodview: Are carbon monoxide

exposures high enough to cause headaches? Does smoking contribute to the elevated levels?

Data Interpretation

Do not read this until after you have completed your plan

Assuming that your plan included pre-shift and post-shift CoHb levels as well as brief questions on number of cigarettes smoked, how would you interpret the following data?

ITO Operator	# Cigs before work	Pre shift CoHb, %	# Cigs during work	Post shift CoHb, %
E-1	NS	2	NS	5
E-2	5	6	20	11
E-3	1	2	5	7
E-4	5	5	5	9
E-5	NS	1	NS	6
E-6	3	3	4	7
E-7	10	7	40	18
E-8	NS	5	NS	6
E-9	NS	2	NS	5
E-10	NS	1	NS	7
CEO	5	5	10	6
CFO	NS	2	NS	2
Pt-1	3	3	5	8
Pt-2	NS	4	NS	6
Pt-3	5	6	20	13
Pt-4	NS	1	NS	5
Pt-5	10	7	10	13
JS	NS	2	NS	4
NG	NS	1	NS	2
BW	2	2	5	7

Some comments:

The following workers reported headaches during the day: E-2, E-4, E-7, E-9, Pt-3, Pt-5, BW. Are these related to CO exposure?

What is your conclusion and what do you report to Fred Goodview.

Epilog

The next morning you get a call from Bettie Sue Busybody, from the local TV station, WRAG-3. Ms Bettie Sue confronted you with the following question: "I understand that there has been a hazardous exposure to many of the workers at Skymart" Can you tell us what is going on? How would you respond to the reporter?

When all was done, do you still have your job?