January 20, 2011

Helmut Greim, Antonia Calafat, Kirstin Macey, Janine Murray, Stephanie Wille, Hans Yu

Biomonitoring

Biomonitoring is defined as the analytical measurement of biomarkers in specified units of tissues or body products (blood, urine, etc.). This includes the measurement of chemicals, or their metabolites and reactants, in the blood, urine, or body tissues. It has become a major tool to evaluate human exposure to chemicals, to link individual exposure to effects and is also applied to identify susceptible individuals or groups of individuals within a population. Because it allows direct measurement of exposure and effects in single individuals it is an optimal tool to correlate exposure and its potential impact on human health. These advantages have resulted in the broad application of biomonitoring to monitor exposure at workplaces, to evaluate whether specific populations are exposed to a specific chemical including the identification of susceptible individuals or populations.

The many uses for biomonitoring data center on their application to address one or more specific questions, which include the identification of a specific exposure source such as lead in paint, or a potential health risk e.g. by measuring a marker of genetic change. In fact there is a continuum from the identification of a special external exposure source to the internal exposure to the human health risk via a biomarker of effect.

Biomarkers

Biomarkers are any substances, structures, or processes that are measured to indicate an exposure or susceptibility or that predict the incidence or outcome of disease. The measurement of biomarkers, in combination with other data, plays an integral role in identifying exposure (sources, trends, etc), potential human health effects, and/or the effectiveness of public health measures introduced to control exposures.

The World Health Organization (WHO 2001) has defined 3 types of biomarkers:
Biomarkers or biological indicators for identifying exposures, effects or susceptibility.
For assessing exposure, biological monitoring and biochemical effect monitoring can be applied. The former determines the chemical or its metabolites in biological materials, whereas the latter determines the interaction of a specific chemical with a biological target such as DNA or protein.

Biomarkers of exposure provide a direct measure of body burden and are applicable for volatile and nonvolatile compounds. It can be the parent compound, metabolite, protein and DNA adduct that is measured in a compartment in an

organism. A biomarker of exposure integrates all routes of exposure and uptake to give a single measurement which can provide unequivocal evidence of exposure; it can be used to indicate extent or magnitude of exposure, but may not be sensitive or specific enough for use in some investigations and is often difficult to relate to risk of adverse outcome. Adducts of chemicals with various proteins can also be used as biomarkers of exposure. Considering the half life of the protein the exposure over time can be estimated. Measures of a compound bound to hemoglobin will reflect exposure for up to 180 days before the red blood cell is cleared from the circulation; for serum albumin up to 20 days, and for histones years in non-dividing cells.
In using any biomarker of exposure the normal (background) level of the biomarker needs to be known in order to determine if and to what extent a deviation from this background range has occurred. There is also the need to know the background variability. A number of factors may influence an individual person's level of the biomarker in the body such as age, sex, lifestyle including smoking, alcohol consumption, etc.
Biomarkers of exposure are also indicators for biological change such as conjugation with glucuronic acid or binding of a reactant with a protein or DNA.

Biomarkers of effect include measurable biochemical, physiological, behavioral, or other alteration in an organism that, depending on the magnitude, can be recognized as associated with an established or possible health impairment or disease. For instance, small and reversible departures from normal values - for example in packed cell volume - may not signal an adverse health effect. Other biomarkers may indicate an early stage in the disease progression; examples of such early biomarkers of effect are somatic mutations, changes in tumor suppressor genes, cytogenetic changes (aberrations, micronuclei, and aneuploidy). Whereas later indicators are of altered structure and function, and such biomarkers include changes in mutational spectra.

Some biomarkers such as DNA adducts cross the above definitions and may be indicators of exposure and possibly of effect, while others are specific to exposure (hemoglobin adducts) or to effects (chromosomal aberrations). Ideally, biomonitoring links these markers of exposure, effect and susceptibility to understand the implications of exposure to chemicals in occupational settings or among the general population.

Biomarkers of effect reflect a change of a cellular function such as an enzyme activity. The latter are biomarkers of susceptibility when used to identify susceptible (sensitive) subpopulations. Biomonitoring in its broadest terms can therefore mean measurement of one or more of these biomarkers as well as the actual chemical itself.

Biomarkers of susceptibility are indicators of an inherent or acquired ability of an organism to respond to the challenge of exposure to a specific chemical substance. These biomarkers provide an indication of the extent to which an individual may be prone to progress from exposure to developing an adverse

health effect. An example is glutathione S- transferase M, a Phase II conjugation enzyme, which depending on high or low activity may increase or impair metabolic inactivation of a reactant.

Activities in Europe and North America

After its preferential use in occupational medicine, biomonitoring became an increasingly relevant tool for environmental epidemiology programs to assess chemical exposure of populations in addition to programs to evaluate concentrations of chemicals in environmental biota such as drinking water, food, fish or air.

In Europe, the discovery of high blood lead levels resulted in the European Commission directive on “Biological Screening of the Population for Lead” in 1977 (77/312/EEC). In 2004 the “Coherent Approach to Biomonitoring through the European Environment and Health Action Plan 2004-2010” started to define further biomonitoring activities (EU 2004) to:

(1) identify policy-relevant objectives
(2) develop protocols
(3) integrate biomonitoring within environmental and health monitoring
(4) develop communication strategies with stakeholders.

To achieve these objectives a pilot biomonitoring project started in 2006, which included the biomonitoring of lead, methyl mercury, and nicotine. Inclusion of phthalates and acrylamide has been discussed.

As part of the European Enviromental and Health Strategy (COM 2003) about 100 biomonitoring activities in children have been described and evaluated. These include activities in Belgium (WVC 2005), France (Huel et al 2002), Germany (GerES 2005, Pesch et al 2002, LGA 2005), Portugal and Poland ((Indulsky et al 1999, Heinrich-Ramm et al 2000, Jakubowsky and Trizcinka-Ochocka 2005), which include breast milk nutrition. Results of Research activities in the Fifth EU Framework Programme are also available (ChildrenGenoNetwork 2005, PINCHR 2005, AIRNET 2005, Plutocracy 2005, MENDOS 2005, BIOMONECES 2005).

In Germany reference values for background exposure levels based on German environmental surveys between 1985 and 1998 (GerES I-III) have been established as well as human biomonitoring values (HBM I and III) by the Human Biomonitoring Commission (2006). HBM I defines the concentration, below which there is no risk, HBM II approximates the no-observed-effect level.

ECETOC (2005) in its Guidance for interpretation of biomonitoring data described case studies on aflatoxin, DDT, fluazifop-butyl, hexachlorobutadiene, parabens, PFOS, vinclozolin, and for select parabens also reported the case studies performed by the World Wildlife Fund (WWF 2005), Greenpeace Netherlands (2005), and CDC (2005). Further the aflatoxin B1 biomarker study in a population of 18,244 men in Shanghai is reported. In this study urinary aflatoxin B1

metabolites and its nucleic acid adduct aflatoxin-N-7-guanine have been determined to evaluate the association between aflatoxin exposure and hepatocellular carcinoma. A highly significant correlation between presence of urinary aflatoxins metabolites and liver cancer risk has been found whereas no association between estimated dietary aflatoxin and hepatocellular carcinoma has been seen (Ross et al 1992, Quian et al 1994).

In the United States by using biomonitoring the National Health and Nutrition Examination Survey (NHANES) has been used to evaluate exposure to select chemicals in the general population. The *Fourth National Report on Human Exposure to Environmental Chemicals* published in 2009 (US CDC 2009) provided data for 212 chemicals in people's blood or urine—75 of which had never before been measured in the U.S. population including metals, polycyclic aromatic hydrocarbons, dioxins, phthalates, polyfluoroalkyl chemicals, polybrominated diphenyl ethers, bisphenol A and other phenols, organophosphates and pyrethroids. In July 2010, CDC released additional data from the NHANES 2005-2006 survey period for 51 of the chemicals previously reported in the *Fourth Report* and the new addition of four parabens and two phthalate metabolites in 2005-2006 (US CDC 2010).

In 2010, the Canadian Health Measures Survey (CHMS) released data on more than 80 environmental contaminants and chemical substances (e.g., heavy metals, pesticides, herbicides, PCBs, polyfluoroalkyl chemicals, bisphenol A) that were measured from 2007 to 2009 in about 6,000 people aged six to seventy-nine years randomly selected across Canada (CHMS 2010).

Previously the National Human Adipose Tissue Study collected data between 1970 and 1987, and the Human Exposure Assessment Survey (NHEXAS) multi-pathway, multi-media exposure data on certain chemical classes.

The Arctic Council of the governments of Canada, Denmark, Finland, Iceland, Norway, Sweden, USA and the Russian Federation support the Arctic Monitoring and Assessment Program to continuously monitor substances in the arctic environment (www.arctic-council.org).

The Health and Environmental Science Institute (HESI) has evaluated the use of biomonitoring data in exposure and human health risk assessment (Albertini et al 2006, Angerer 2006, Doerrer 2007) and has performed case studies on inorganic arsenic (Hughes 2006), methyl eugenol (Robison and Barr 2006), the organophosphorus pesticides chlorpyrifos and malathion (Barr and Angerer 2006), PFOS (Butenhoff et al 2006), phthalate esters (Calafat and McKee 2006), and polybrominated diphenyl ethers (Birnbaum and Cohen Hubal 2006). Recently, Lew et al (2010) reported a study on children who were exposed to arsenic from a contaminated playground.

These activities indicate that biomonitoring is successfully applied in risk assessment and risk management. This includes:

- Integration in risk assessment
- Exposure assessment to evaluate need for risk management
- Evaluation of regulatory measures
- Assessment of trends of exposure
- Exposure control at workplaces

This is exemplified by the following eamples.

Examples for the different application of biomonitoring

1. Integration in risk assessment: methyl eugenol

Given to the natural occurrence of methyl eugenol a majority of the population is likely to be exposed. The compound is considered to be non-mutagenic and non-clastogenic at biological relevant concentrations, most likely due to rapid O-demethylation via P450 1A2. At high concentrations induction of unscheduled DNA synthesis has been shown, and at high bolus exposures life-time studies in rats and mice showed hepatic neoplasms. At high exposure P450 2C9 and 2C19 also contribute to the metabolism and result in a 1'-hydroxymethyl eugenol metabolite, which is involved in DNA adduct formation.
However, biomonitoring studies indicate that human exposure is several orders of magnitude lower than the lowest dose used in rodent bioassays.

2. Exposure assessment to evaluate need for risk management: PAH in parquet glue

In the 1950s and 1960s a tar oil based parquet glue containing concentrations of polycyclic aromatic hydrocarbons (PAH)s up to 12,000 mg benzo(a)pyrene BaP)/kg glue was common practice in Germany. Broken or loose boards, cracks, or clefts in these parquet floors have been expected to lead to high PAH-contamination in house dust. Infants and small children have been considered to be at risk of an elevated PAH exposure when ingesting PAH-containing dust during playing and crawling.
To evaluate whether measures needed to be taken to eliminate this potential source of children's exposure to PAHs, biomonitoring was offered to all children of a housing estate built in Frankfurt with parquet flooring in 1955/56 (Heudorf and Angerer 2000)
Spot urines of 347 children up to 6 years of age were analyzed for hydroxy-phenanthrenes and 1-hydroxypyrene by HPLC and flame ionization detector. The detection limit of the different metabolites was 5 ng/L. At the same time, researchers measured the BaP content of the glue used in the different apartments.
In about one third of the apartments tested, the parquet glue was a tar-free bitumen material. In another third of the flats the glue contained 10-3000 mg

BaP/kg and in the last third of apartments, the BaP content was above 3000 mg/kg with a maximum value of 12,000 mg/kg. However, the median urinary concentrations of the PAH-metabolites in the children living in these apartments did not differ. Because an increased internal PAH exposure among children living in homes,where coal tar based parquet glue was used was not found, an increased health risk was excluded. Therefore, the health office of the City of Frankfurt, which initiated this investigation, concluded that no measures to reduce exposure needed to be taken. However, for reasons of prevention removal of the parquet and the glue was recommended in cases with high PAH concentrations in the dust.

3. Evaluation of regulatory measures: Lead

According to the U.S. Centers for Disease Control and Prevention, government legislation on the use of lead in gasoline resulted in a decline of mean? blood lead levels in the US population between 1976 and 2001 from 16 to 2 ug/dl.
In Europe the report on lead exposure of children from the Danish Institute of Epidemiology of 1978 described blood lead levels in children from various areas in Denmark. The highest levels were found in children living near major traffic routes and whose fathers have been working in secondary lead smelters (90-290 microg/l). Children living in non-contaminated areas without intense traffic had blood lead levels between 60 and 170 microg/l (Bach, 1978). This agrees with Swedish data of 1978 as reported by Skerfving et al (1999). The concentrations of lead in the blood of Swedish children have steadily decreased in the following 15 years (Skerfving et al, 1999) and up to the year of 2000 the concentrations had not reached a new steady state level (CSTEE 2000). Similar to Denmark a Swedish study has shown that blood lead levels continuously declined and that food became the main source of lead exposure even in young children living in areas with high soil lead concentrations, i.e. downtown Stockholm (<10-330 mg/kg in soil) and mining areas (20-5,000 mg/kg). CSTEE (2000) concluded that lead even in soil and dust contributes little to the total intake.

4. Assessment of trends of exposure
Presently the inclusion of the following examples are being discussed:

- Decline in serum cotinine levels as a result of measures in the USA in limiting smoking in public places
- Decline in serum PCBs levels or in PBDEs levels after regulatory action
- Decline in serum PFOS levels in the USA after 3M discontinued production of PFOS and PFOS-precursors in the early 2000s.
- Decline in blood lead levels after reduction of lead in gasoline

5. Exposure control at workplaces

To determine individual exposure at work-places the Deutsche Forschungsgemeinschaft has established standard procedures and Biological Exposure Indices for about 130 compounds (see DFG 2010).

In North America the American Conference of Governmental Industrial Hygienists (ACGIH) established “Biological Exposure Indices” for about 50 metals and inorganic and organic compounds (see ACGIH 2010).
Recently SCOEL (Scientific Committee on Occupational Exposure Limits) on behalf of the DG Employment started to define biological exposure indices to control exposure of metals and chemicals at the work place.

Brief description of available methodology referring to existing guidance documents and other guidelines.

Discussion
Conclusion on optimal design of specific studies to improve applicability for risk assessment and regulations
Recommendations to use available data for exposure assessment in risk assessment and regulations including use of data in one country for regulations in another.

Annex
Existing guidelines/methods to use biomonitoring

References

ACGIH (American Conference of Governmental Industrial Hygienists)TLVs and BEIs based on the Documentation of the Threshold Limit Values for Chemical Substances and Physical Agents & Biological Exposure Indices. Cincinnati, Ohio. www.acgih.org

AIRNET (2005) A thematic network on air pollution and health. Research project funded under the Fifth Framework Programme of the European Commission (contract number: QLRT-2001-00441). Airnet.iras.uu.nl

Albertini, R, Bird, MG, Doerrer, NG, Needham, L, Robison, S, Sheldon, L, and Zenick, H. 2006. The use of biomonitoring data in exposure and human health risk assessment. Environ Health Perspect. 114, 1755-1762

Angerer, J, Bird, MG, Burke, TA, Doerrer, NG, Needham, L, Robison, SH, Sheldon, L, and Zenick, H. 2006. Meeting Report. Strategic biomonitoring initiatives: moving the science forward. Toxicol Sci. 93, 3-10

Bach E: Borns belastning med bly Rapport om en befolkningsundersogelse I miljostyrelsens undersogelsesprogram for tungmetaller. Dansk institut for Klinisk Epidemiologi, 1978.

Barr, DB, Angerer, J. 2006. Potential uses of biomonitoring data: a case study using the organophosphorus pesticides chlorpyrifos and malathion. Environ Health Perspect. 114, 1763-1769

BIOMONECES (2005) Development of new BIOmarkers for monitoring occupational exposure to carcinogenic substances. Research project funded under the Fifth Framework Programme of the European Commission. www.biomonecs.com

Birnbaum, LS, Cohen Hubal, EA. 2006. Polybrominated diphenyl ethers: a case study for application of biomonitoring data to characterize exposure. Environ Health Perspect. 114, 1770-1775

Butenhoff, JL, Olsen, GW, Pfahles-Hutchens, A. 2006. The applicability of biomonitoring data for perfluorooctanesulfonate (PFOS) to the environmental public health continuum. Environ Health Perspect. 114, 1776-1782

Calafat, AM, McKee, RH. 2006. Integrating biomonitoring exposure data into the risk assessment process: phthalates (diethyl phthalate and di[2-ethylhexyl] phthalate) as a case study. Environ Health Perspect . 114, 1783-1789

ChildrenGenoNetwork (2005) European network on children's susceptibiloity and exposure to environmental toxicants QLK4-CT-2002-02198. www.pubhealth.ku.dk/cgn

COM (2003) Baseline report "Biomonitoring of Children" in the framework of the European Environment and Health Strategy. COM (2003) 338 final. www.brussels-conference.org/Download/baseline_report/BR_Biomonitoring_final.pdf

CSTEE (Scientific Committee on Toxicity, Ecotoxicity and the Environment): Opinion on Lead – Danish Notification 98/595/DK. Brussels 5th of May 2000

Doerrer, NG. 2007. Integration of human biomonitoring exposure data into risk assessment: HESI initiatives and perspectives. Int J Hyg Environ Health 210, 247-251

ECETOC (European Center for Ecotoxicology and Toxicology of Chemicals, Brussels 2005) Guidance for the interpretation of biomonitoring data. Document No. 44

EU (European Union) 2004: European Environment and Health Action Plan 2004-2010. Brussels, Belgium. Available: http://www.eu-humanbiomonitoring.org/doc/ta_vol2.pdf

GerES (2005) www.umweltbundesamt.de/survey-e/index.htm

German Human Biomonitoring Commission (2006) Avalable:

http://www.umweltbundesamt.de/uba-info-daten-e/daten-e/monitor/pub.htm

Heinrich-Ramm R, Jakubowski M, Heinzow B, Molin Christensen J, Olsen E, Hertel O (2000) Biological monitoring for exposure of volatile organic compounds (VOC). Pure Appl Chem 72, 385-436

Heudorf U, Angerer J: Urinary levels of 1-hydroxypyrene, 1-, 2-, 3-, and 4-hydroxyphenanthrenes in children living in flats with high PAH-contents of glue for parquet floors – Data of the former American housing estates, Frankfurt am Main, Germany. Umweltmed Forsch Prax 5, 218-226, 2000 (German article, English abstract)

Huel G, Frery N, Takser L, Jouan M, Hellier G, Sahuquillo J, Giordanella JP (2002) Evaluation of blood lead levels in urban French population (1979-1995). Rev Epidemiol Sante Publique 50, 287-295

Hughes, MF. 2006. Biomarkers of exposure: a case study with inorganic arsenic. Environ Health Perspect. 114, 1790-1796

Indulsky J, Jakubowski M, Merecz D (1999) Present environmental lead exposure in Polish children and its possible influence on the CNS functions Eur J Oncol 4, 631-636

Jakubowsky M, Trizcinka-Ochocka M (2005) Biologicalö monitoring of exposure: Trends and key developments. J Occup Health 47, 22-48

Lew et al (2010) Env Sci Technol 44, 3986-3991

LGA (2005) www.landesgesundheitsamt.de

MENDOS (2005) BioMimetic optical sensors for environmental endocrine disruptor screening. Research project funded under the Fifth Framework Programme of the European Commission (contract number: QLRT-2001-02323) www.mendos.org

National Human Adipose Tissue Study

NHEXAS (National Human Exposure Assessment Survey)

Pesch A, Wilhelm M, Rostek U, Schmitz N, Weishoff-Houben M, Ranft U, Idel H (2002) Mercury concentrations in urine, scalp hair, and saliva in children from Germany. J Expo Anal Environ Epidemiol 12, 252-258

PINCHE (2005) Policy interaction network on children's health and environment. Research project funded under the Fifth Framework Programme of the European Commission. www.pinche.nl

Plutocracy (2005) Placental uptake and transfer of environmental chemicals relating to allergy in childhood years. Research project funded under the Fifth Framework Programme of the European Commission. www.bristol.ac.uk/plutocracy/consortium html

Quian GS, Ross RK, Yu MC, Yuan JM, Gao YT, Henderson BE, Wogan GN, Groopman JD (1994) A follow up study of urinary biomarkers of aflatoxin exposure and liver-cancer risk in Shanghai, Peoples Republic of China. Cancer Epidem Biom Prev 3, 3-10

Robison, SH, Barr, DB (2006) Use of biomonitoring data to evaluate methyl eugenol exposure and its relationship to the environmental public health continuum. Environ Health Perspect. 114, 1797-1801

Ross RK, Yuan JM, Yu MC, Wogan GN, Qian GS, Tu JK, Groopman JD, Gao YT, Henderson BE (1992) Urinary aflatoxin biomarkers and risk of hepatocellular carcinoma. Lancet 339, 943-946

Skerfving S, Bencko V, Vahter M, Schutz A, Gerhardsson L: Environmental health in the Baltic region,- Toxic metals. Scand J Work Environ Health, 25, suppl. 3, 40-64, 1999

Smolders R, Koppen G, Schoeters G: Translating biomonitoring data into risk management and policy implementation options for a European Network on Human Biomonitoring. Environ Health Jun 5; 7 Suppl 1:S2

US CDC. 2009. Fourth National Report on Human Exposure to Environmental Chemicals. Atlanta, GA:Centers for Disease Control and Prevention; National Center for Environmental Health; Division of Laboratory Sciences. Available: http://www.cdc.gov/exposurereport/pdf/FourthReport.pdf.

US CDC. 2010. Fourth National Report on Human Exposure to Environmental Chemicals. Updated Tables. Atlanta, GA:Centers for Disease Control and Prevention; National Center for Environmental Health; Division of Laboratory Sciences. Available: http://www.cdc.gov/exposurereport/pdf/Update_Tables.pdf

WVC (2005) Department Welzijn, Volksgezondheit en Cultur. www.wvc.vlaandern.be