correlated very well with Cr in RBC, 8-OHdG, and comet scores, suggesting their potential usefulness as new Cr(VI) biomarkers if further validation, purification, identification, and assay development are conducted. (supported by grants: RD830682, ES00260, and ES10344)

**#2208 Methylation and mRNA expression of the $p15^{INK4b}$, $p16^{INK4a}$, *BRCA1 and hMLH1* genes in ovarian cancer.** Zhensheng Liu, Li-E Wang, Luo Wang, Karen H. Lu, Melissa Bondy, Gordon B. Mills, Qingyi Wei. *The University of Texas M.D. Anderson Cancer Center, Houston, TX.*

Aberrant methylation of CpG islands can cause inactivation of tumor suppressors, contributing to the development of cancer. Although $p15^{INK4b}$, $p16^{INK4a}$, *BRCA1* and *hMLH1* hypermethylation has been reported in a number of primary tumors and cancer cell lines, no reported studies have simultaneously investigated the correlation between methylation and mRNA expression of these genes in epithelial ovarian cancers and normal ovary. We used the methylation-specific polymerase chain reaction (MSP) and real-time reverse transcription-PCR to determine methylation status and the relative mRNA expression of the genes, respectively, in 52 ovarian cancer tissues and 48 normal ovarian tissues. We found that, methylation of $p15^{INK4b}$, $p16^{INK4a}$, *BRCA1* and *hMLH1* genes was detected in 31% (16/52), 25% (13/52), 52% (27/52) and 19% (10/52), respectively, of ovarian cancers, as compared with 6% (93/48) ($P = 0.002$), 38% (18/48) ($P = 0.178$), 69% (33/48) ($P = 0.086$), and 21% (10/48) ($P = 0.841$), respectively. These results suggested that only $p15^{INK4b}$ was methylated at a higher rate in ovarian cancer. The relative expression levels of $p15^{INK4b}$, *BRCA1* and *hMLh1* were significantly lower in ovarian cancers (13%, 10% and 11%, respectively) than in normal ovary (40% [$P = 0.007$], 64% [$P = 0.007$] and 35%[$P = 0.003$], respectively, but not for $p16^{INK4a}$ ($P = 0.789$). However, low expression of $p15^{INK4b}$, $p16^{INK4a}$, *BRCA1* and *hMLH1* were associated with significantly increased risk of ovarian cancer (OR = 9.14; 95% CI = 2.62-31.9 for $p15^{INK4b}$, OR = 3.02; 95% CI = 1.15-7.94 for $p16^{INK4a}$, OR = 10.5; 95% CI = 2.96-37.5 for *BRCA1*, OR = 17.0; 95% CI = 4.11-69.9 for *hMLH1*). Surprisingly, methylation status and expression levels do not appear to be linked, as there was no correlation between the two events. We conclude that low expression of $p15^{INK4b}$, $p16^{INK4a}$, *BRCA1* and *hMLH1* may contribute to the development of ovarian cancer [Supported by CA083639, ES11740, CA100264, ES11047 and CA16672].

**#2209 Development of a biomarker decision support system.** Russell E. Savage, Jr., Andrew Maier, Lynne Haber, Eric Hack, W. Gregory Lotz, Paul Schulte, Bruce Fowler. *National Institute for Occupational Safety & Health, Cincinnati, OH, Toxicology Excellence for Risk Assessment (TERA), Cincinnati, OH, Agency for Toxic Substances and Disease Registry, Atlanta, GA.*

For over two decades, scientists have been touting the importance, and ultimate application of biomarkers in reducing disease and protecting individuals from the harmful effects of exposure to occupational and/or environmental chemicals. However, few scientists apply stringent or recommended criteria to biological end points before proclaiming them biomarkers. While established guidelines for biomarker validation exist, methods for their implementation and case studies testing the methods are rare. This pilot study seeks to develop and demonstrate the use of a framework for integrating complex and multifaceted data, validating biomarkers, and incorporation of the biomarkers into an occupational risk assessment. The objectives are 1) to identify an occupationally relevant case-study chemical; 2) to develop a structure for a biomarker database that can be used to organize the diverse types of data; 3) to use Bayesian analysis and regression techniques to test and validate (or discount) biomarkers along the entire exposure-disease continuum and 4) use the biomarker database information to develop a risk assessment and compare that with a risk assessment developed using historical, traditional methodologies. A survey was developed and disseminated to 59 occupational safety and health professionals to help identify an occupationally relevant, data rich, case study compound. The Biomarker framework includes multiple steps. The data are binned into one of the exposure-disease categories (e.g., exposure, internal dose, effective dose, early effects, mild/moderate effects, or severe effects). The data are ranked based on relevance, study quality, and continuity (e.g., exposure and gene expression and disease data), and the highest ranking data are reformatted and entered into a database. A Bayesian belief network is used to validate the biomarkers by analyzing the strength of the dependencies between exposure, the potential biomarkers, and disease. Regression analysis is used to generate dose-biomarker relationships that are examined to confirm or reject biomarkers. The framework lays out an approach to consider a variety of biomarkers from the exposure-disease continuum for the enhancement of occupational risk assessment.

## EPIDEMIOLOGY 5: Biomarkers of Human Exposure to Tobacco

**#2210 A case-control study of passive smoking and bladder cancer risk among lifelong nonsmokers in Los Angeles.** Xuejuan Jiang, Jian-Min Yuan, Paul L. Skipper, Steven R. Tannenbaum, Ronald K. Ross, Mimi C. Yu. *University of Southern California, Los Angeles, CA and Massachusetts Institute of Technology, Cambridge, MA.*

Background: Cigarette smoking is a major risk factor for bladder cancer as well as a prominent point source of 4-aminobiphenyl (4-ABP), a recognized human bladder carcinogen. 4-ABP hemoglobin adducts (4-ABP-Hb adducts) are established biomarkers of 4-ABP exposure to the bladder. The role of passive smoking in the etiology of bladder cancer is largely unknown. Methods: A population-based case-control study of bladder cancer was conducted in Los Angeles County from 1987-1999. Passive smoking status was ascertained for 154 lifelong nonsmoking cases and 313 lifelong nonsmoking controls through in-person interviews. 4-ABP-Hb adducts were quantitatively measured by GC-MS analysis. Results: Relative to subjects without passive smoke exposure, those with current exposure were more likely to exhibit high (i.e., above median level, or 19.70 pg/g Hb) than low (below median level) level of 4-ABP-Hb adducts (odds ratio - OR, 2.03; 95% confidence interval - 95% CI, 0.86-4.76). This association was stronger for exposure from spouse/partner (OR, 2.65; 95% CI, 1.03-6.78) than that from coworkers (OR, 1.93; 95% CI, 0.81-4.61). Overall, passive smokers were 39% more likely to develop bladder cancer than non-passive smokers (OR, 1.39; 95% CI: 0.74-2.61). Elevated and comparable risks were noted whether exposure occurred during childhood (OR, 1.32; 95% CI, 0.69-2.51) or adulthood (OR, 1.29; 95% CI, 0.68-2.45), and whether exposure occurred at home (OR, 1.22; 95% CI, 0.63-2.37) or at work (OR, 1.35; 95% CI, 0.70-2.58) during adulthood. There were increasing risks with increasing numbers of years of passive smoke exposure from spouse/partner or coworkers. Conclusion: Passive smoking may be a risk factor for bladder cancer in lifelong nonsmokers.

**#2211 DNA repair polymorphisms, second hand smoke and lung cancer risk.** David P. Miller, Sohee Park, Elena Gitin, Wei Zhou, Geoffrey Liu, John C. Wain, Thomas J. Lynch, David C. Christiani. *Harvard School of Public Health, Boston, MA and Massachusetts General Hospital, Boston, MA.*

Background: Excision repair cross complementation group 1 (ERCC1) is a highly conserved protein and the lead enzyme in the nucleotide excision repair process. Previous studies have considered the interaction between this polymorphism and active smoking. Second hand smoke (SHS) is composed of emissions from cigarette smoke and contains a higher concentration of tobacco carcinogens than active smoke. SHS has been associated with lung cancer, though its measurement has been a challenging task for many epidemiologic studies. We examined the association between SHS and lung cancer stratified by the *ERCC1 C8092A* polymorphism. Methods: This is a part of an ongoing case control study evaluating the molecular epidemiology of lung cancer, which began in 1992 at the Massachusetts General Hospital. We analyzed all cases (n=1061) and controls (n=1156), in addition to cases and controls with primary smoking packyears fewer than or equal to 15, which represented a subsample less affected by active smoking for evaluation of SHS relationships. We evaluated SHS exposure in different settings (leisure, work and at home) for different time periods of exposure. We captured length and location of exposure and timing of exposure with respect to a study participant's age (0 - 25, 25 - 50, >50 years of age). The *ERCC1* genotypes were detected using the polymerase chain reaction with fluorescent allele-specific oligonucleotide probes (TaqMan method). Multiple logistic regression was used to study the association between SHS and lung cancer, adjusting for age, gender, active smoking variables. Results: Second hand smoke exposure during leisure activities was associated with increased lung cancer risk. In all cases and controls, the adjusted odds ratio (AOR) for the *A/A+A/C* strata was 1.2 (95%CI: 0.5 - 2.7) compared to *C/C* (AOR = 2.7; 95%CI = 1.6-6.4). In the subset of cases and controls with packyears ≤ 15, the *A/A+A/C* AOR was 3.6 (95%CI 0.9 - 15.5), and the AOR for the C/C strata was 5.2 (95% CI: 1.3 - 20.8). For cases and controls with packyears ≤ 15, given the same duration of exposure, if the exposure took place in the earlier part of life (0 to 25 years of age) compared to an exposure occurring after age 25, this would result in 2.2 fold increase in risk of lung cancer for the A/A + A/C strata compared to only a 3% increase in risk for the C/C strata. Conclusions: The presence of the *ERCC1 C8092A* polymorphism appear to greatly modify the association between SHS and lung cancer risk, particularly when the exposure takes place early in life. Supported by NIH grants CA 74386 and ES00002 and the Flight Attendant Medical Research Institute Award.

**#2212 The glutathione S-Transferase mu polymorphism is associated with an increased tobacco-associated risk of oral and pharyngeal carcinomas.** Edward S. Peters, Mei Liu, Michael McClean, Karl Kelsey, Ellen A. Eisen. *Louisiana School of Public Health, New Orleans, LA, Harvard School of Public Health, Boston, MA, Boston University School of Public Health, Boston, MA.*