P56

Semi-Quantitation of Polycyclic Aromatic Hydrocarbon (PAH)-DNA Adducts in Human Placenta by Immunohistochemistry (IHC) and the Automated Cellular Imaging System (ACIS): Effect of Smoking. Sirajuddin, P[1], Pratt, MM[1], Sram, RJ[2], Manchester, DK[3], Poirier, MC[1]. [1]Carcinogen-DNA Interaclions Section, LCCTP, NCI, Bethesda, MD, United States. [2]Institute of Experimental Medicine AS CR, Prague, Czech Republic, Prague, Czech Republic, Czech Republic. [3]University of Colorado School of Medicine, Denver, Colorado, United States.

Epidemiologic studies have shown that parental exposure to carcinogens in tobacco smoke is implicated in the development of childhood cancers and leukemia. The placenta is known to metabolize drugs and other compounds, including PAHs, that subsequently enter fetal circulation. PAH-DNA adducts have been detected in DNA extracted from human placenta, however, tissue distribution of these adducts and their proximity to fetal circulation has not been reported. It is therefore of interest to examine localization of PAH-DNA adducts in intact human placentas. Previously, antiserum elicited against DNA modified with anti (+/-)r-7,t-8-dihydroxy-t-9,10-epoxy-benzo[a]pyrene (BPDE) was validated for semi-quantitative IHC of human esophagus using the Automated Cellular Imaging System (ACIS). Here we report results for PAH-DNA adduct visualization and semi-quantitation in human placenta. Cultured human keratinocytes, exposed for 1 hr to 0, 0.053, 0.153, or 0.331 µM BPDE, were paraffin embedded, stained, and measured by ACIS to provide a standard curve. These cells showed parallel, dose-dependent increases in BPDE-DNA adducts determined by IHC/ACIS and by quantitative BPDE-DNA chemiluminescence immunoassay (CIA). Fourteen full-term placenta samples were stained for PAH-DNA adducts and showed positive nuclear staining localized in the metabolically-active syncytiotrophoblast (ST) and cytotrophoblast (CT) cells that line the chorionic villi and form the barrier between the maternal and fetal circulation. Serum specificity was confirmed when nuclear staining was significantly reduced in parallel sections stained with anti-BPDE-DNA that had previously been absorbed with the BPDE-DNA immunogen. Comparison of placenta ACIS values with the keratinocyte standard curve revealed PAH-DNA adduct values of 173 ± $35/10^8$ nucleotides (mean ± SE) for 7 smokers and 58 ± $15/10^8$ nucleotides for 7 non-smokers ($p<0.02$, Mann-Whitney and two tail t test). This study shows significantly higher PAH-DNA adduct levels in smokers compared to non-smokers, and that adducts were highly concentrated in the ST and CT cells that metabolize PAHs. Comparing placental PAH-DNA adduct formation with smoking status and childhood health may yield insights into the etiology of childhood cancers.

P57

International Study of Somatic Cell Translocation Frequencies in Control Populations. Tucker, JD[1], Kleinerman, R[2], Ha, M[2], Bhatti, P[2], Hauptmann, M[2], Sigurdson, A[2], Sram, RJ[3], Beskid, O[3], Tawn, EJ[4], Whitehouse, C[4], Lindholm, C[5], Kodama, Y[6], Nakamura, N[6], Vorobstova, I[7], Oestreicher, U[8], Stephan, G[8], Yong, L[9], Bauchinger, M[10], Chung, H-W[11], Darroudi, F[12], Roy, L[13], Barquinero, J[14], Livingston, G[15], Schmid, E[16], Blakey, D[17], Voisin, P[18], Littlefield, G[19], Edwards, A[20]. [1]Wayne State University, Detroit, MI, United States. [2]National Cancer Institute, NIH, DHHS, Rockville, MD, United States. [3]Institute of Experimental Medicine and Health Institute of Central Bohemia, Prague, Czech Republic. [4]Westlakes Research Institute, Cumbria, United Kingdom. [5]Radiation and Nuclear Safety Authority (STUK), Helsinki, Finland. [6]Radiation Effects Research Foundation, Hiroshima, Japan. [7]Central Research Institute of Roentgenology and Radiology, St. Petersburg, Russia. [8]Bundesamt für Strahlenschutz (BfS), Obeschleissheim, Germany. [9]National Institute for Occupational Safety and Health, Cincinnati, OH, United States. [10]GSF-National Research Centre for Environment and Health, Neuherberg, Germany. [11]Seoul National University, Seoul, Korea, South. [12]Leiden University Medical Centre (LUMC), Leiden, Netherlands. [13]Institut de Radioprotection et Sûreté Nucléaire (IRSN), Fontenay-aux-Roses, France. [14]Universitat Automoma de Barcelona (UAB), Bellaterra, Spain. [15]Centers for Disease Control, Atlanta, GA, United States. [16]LMU Munchen, Neuherberg, Germany. [17]Health Canada, Ottawa, ON, Canada. [18]Institut de Radioprotection et Sûreté Nucléaire (IRSN), Fontenay-aux-Roses, France. [19]Oak Ridge Associated Universities, Oak Ridge, TN, United States. [20]Health Protection Agency, Didcot, United Kingdom.

Biological monitoring of radiation doses in humans can contribute important estimates of exposure, especially when physical measurements are unavailable. Translocations have been the most widely applied biomarker of past radiation exposure in epidemiologic studies because of well-characterized dose-response curves and the persistence of translocations detectable many years later. Establishing baseline levels of translocations will contribute to the usefulness of this endpoint in cases of accidental exposure. It is well accepted that the frequency of chromosome aberrations increases with radiation exposure and age, but the effects of gender, ethnicity and lifestyle (e.g. smoking) on background translocation yields is not known with certainty. Pooled analyses of translocation data in unexposed individuals have been conducted, but the largest study included only 385 subjects. Background aberration frequencies were overwhelmingly influenced by age, but the effect of age as modified by gender and cigarette smoking remains unclear. Here we expanded the number of subjects with the goal of establishing control levels of translocation frequencies by age, gender, ethnicity and smoking status. Fifteen laboratories in North America, Europe and Asia contributed data on 1,822 unexposed individuals, with a minimum of 200 cell equivalents (CE) per subject. Ages ranged from newborn (cord blood) to 85 years. The study population was 37% female, 40% reported ever smoking, and 77% were Caucasian, 13% Black, 8% Asian, and 2% were other ethnicity. Age was the strongest predictor of translocation frequency ($p<0.001$). The mean number of translocations was 0.03/100 CE (95% CI=0.02-0.04) for newborns and 1.7/100 CE (95% CI=1.4-2.0) for subjects 75 years and older. Translocation frequencies per 100 CEs were similar for men and women up to age 50, when they diverged, with women having higher frequencies than men. Smokers had higher translocation frequencies than non-smokers ($p < 0.001$) after adjustment for gender, ethnicity, and laboratory. We noted significant variation by laboratory in all analyses. We were unable to separate the effect of ethnicity on translocations from inter-laboratory variation. More work is needed to understand the different age responses in different populations.