POSTERS

P XIV B.1 Molecular dosimetry of cancer risk factors

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Molecular dosimetry addresses quantification of biomarkers in individuals susceptible to develop disease. Carcinogenesis is a multi-step process and host factors as well as exogenous elements predispose an individual to cancer. Life style, dietary factors are risk associated. The Ehrenberg paradigm conceptualizes the steps from exposure to the mutation end point and defines a suite of subordinate dosimetries. The common approach is toward measurement of mutations, cytogenetic changes and carcinogen-macromolecular adducts in the target epithelial cells and surrogate blood cells as well, that predicts the end result polymorphic phenotypes. Single cell gel assay that detects DNA strand breaks and alkali labile damages were shown to be in fair agreement with ploidy status and cytoflowmetric profile. We compared surgical specimens and distal tissues from human gastric tumors and the damage pattern was qualitatively comparable to pathology that varied from normal, chronic gastritis and malignancy. Gastritis showed reasonable increase in strand breaks that suggested putative lesions. Flavonoids, quercetin and luteolin are antimutagenic, are potential chemopreventors and our animal studies were in agreement with the strand break assay. The assay emerges as a powerful tool in terms of prognostic significance and cost effectiveness and we believe this assay at single cell level may be good to biomonitor high risk population.

P XIV B.2 DNA damage in smokers' lymphocytes

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It is well known that tobacco contains potential carcinogens including aromatic hydrocarbons, N-nitrosamines, aromatic amines, aldehydes and halomethanes. Single Cell Gel Electrophoresis (SCGE, comet assay) is a simple and very sensitive method which measures single strand breaks (ssbs, alkaline method), as well as double strand breaks (dsbs, neutral method) at the single cell level. Here the alkaline method was used in a Molecular Epidemiological study to compare lymphocytes of two groups of smokers (first group 20–25, second group 40–60 years old) with each other and with the corresponding control groups. The effect of H_2O_2 on these lymphocytes was also examined. The total number of these participants in the study was 80 individuals. Since other environmental factors can also be involved, a detailed questionnaire was given to the subjects inquiring particulars about their working conditions and dietary habits. The results revealed an increased DNA damage of the smokers' lymphocytes if compared with those of the non smokers. A correlation according to age was also found. Finally H_2O_2 treatment induced higher levels of damage in smokers if compared to non smokers.

Keyword(s): smoking; lymphocytes; comet assay

P XIV B.3 Evaluation of the genotoxic effect of 2-ethoxyethanol

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Glycol ethers such us 2-ethoxyethanol or ethylene glycol monoethyl ether (2-EE) are used extensively as solvents in paints, dyes, lacquers in domestic and industrial products to dissolve the mixtures in order to keep it in solution. Therefore, we can find an important number of exposed people and a growing concern on the effect of this product on the human health. Excessive exposure to 2-EE in rats has been reported to cause testicular toxicity.

The aim of this study was to investigate the effect of this agent on lymphocyte cultures, exposed to different concentrations of 2-EE (5, 10, 30, 40, 60, 80 and 100 mM) using micronucleus and the comet assay. It's very important to conduct this studies in order to assess the risk associated with exposure to genotoxic compounds and its results should aid in the identification of hazards to make programs, at a national level, of control and prevention.

Keyword(s): 2-ethoxyethanol; Mutagenicity

P XIV B.4 Detection of primary DNA damage in an unicellular protozoa *Acanthamoeba castellanii* by a modified test protocol of the comet assay - investigations of humic substances and their interaction with genotoxines

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One of the important tasks of environmental monitoring is the detection of potentially hazardous compounds in water. Many of these compounds in e.g. surface water are described as DNA damaging agents in aquatic animals such as fish and mussel. But there is still a necessity for the development of methods to detect DNA damage in planktonic and benthic organisms of the aquatic foodweb such as algae or protozoa. The comet assay is a powerful tool for measuring primary DNA damage or strand breaks caused by genotoxines in single cells. Thus, the application of the comet assay to the aquatic unicellular protozoa Acanthamoeba castellanii was tested. A test protocol conventionally used for the detection of DNA damage in animal cells as reported by Singh et al. 1988 was modified to optimize the experimental conditions e.g. duration of cell lysis, preincubation and electrophoresis. After optimization of the test protocol well known genotoxines such as 4-nitroquinoline-1-oxide were tested and found to induce genotoxic effects in this aquatic species. Besides the effects caused by genotoxines of anthropogenic sources, it is interesting to know whether natural water compounds such as humic substances influence the genotoxic potential in water samples. Humic substances are known to be hardly degradable, but only few information about their biological function is available. Toxic effects of humic substances on aquatic microorganisms are known from former studies and therefore expected to create genotoxic effects as well in various aquatic species. Because of their molecular structure humic substances may interact with toxicants in such a way, that they are able to mask or to demask the toxicity or genotoxicity of toxicants in water. The aim of the present study was i) the adaption and modification of the comet assay test protocol to cells of Acanthamoeba castellanii, ii) the detection of potential DNA damaging effects of humic substances and iii) the evaluation of the masking influence of humic substances on well known genotoxines.

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Keyword(s): Comet assay; Acanthamoeba castellanii; Humic substances

P XIV B.5 The use of the alkaline comet assay to detect DNA crosslinks: Improved methodology

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We have recently demonstrated that the alkaline comet assay is able to rapidly and easily detect crosslinking agents in human leucocytes (1). To enable this, a defined amount of DNA strand breaks is induced by damaging the DNA in a stan-dardized way. DNA-DNA and/or DNA-protein crosslinks, caused by crosslinking agents, are capable of reducing the migration of these DNA fragments. To improve the sensitivity of the method, the cells were irradiated by gamma-radiation to set the standard damage instead of treating them with methyl methanesulfonate, using isolated lymphocytes and not whole blood. With these modifications we reached a more steady distribution of strand-breaks and made the detection of crosslinks more sensitive.

This methodology was used to investigate the DNA-crosslinking potency of different chemicals in vitro. Isolated human lymphocytes were treated with the agents for two hours, embedded in agarose, and irradiated with