Use of Genotoxicity Methods for Human Biomonitoring:

Prevention – Diagnosis and Therapy of Cancer

International Workshop, June 6th 2025, Location: Hörsaalzentrum 7L (7th floor), AKH, Vienna

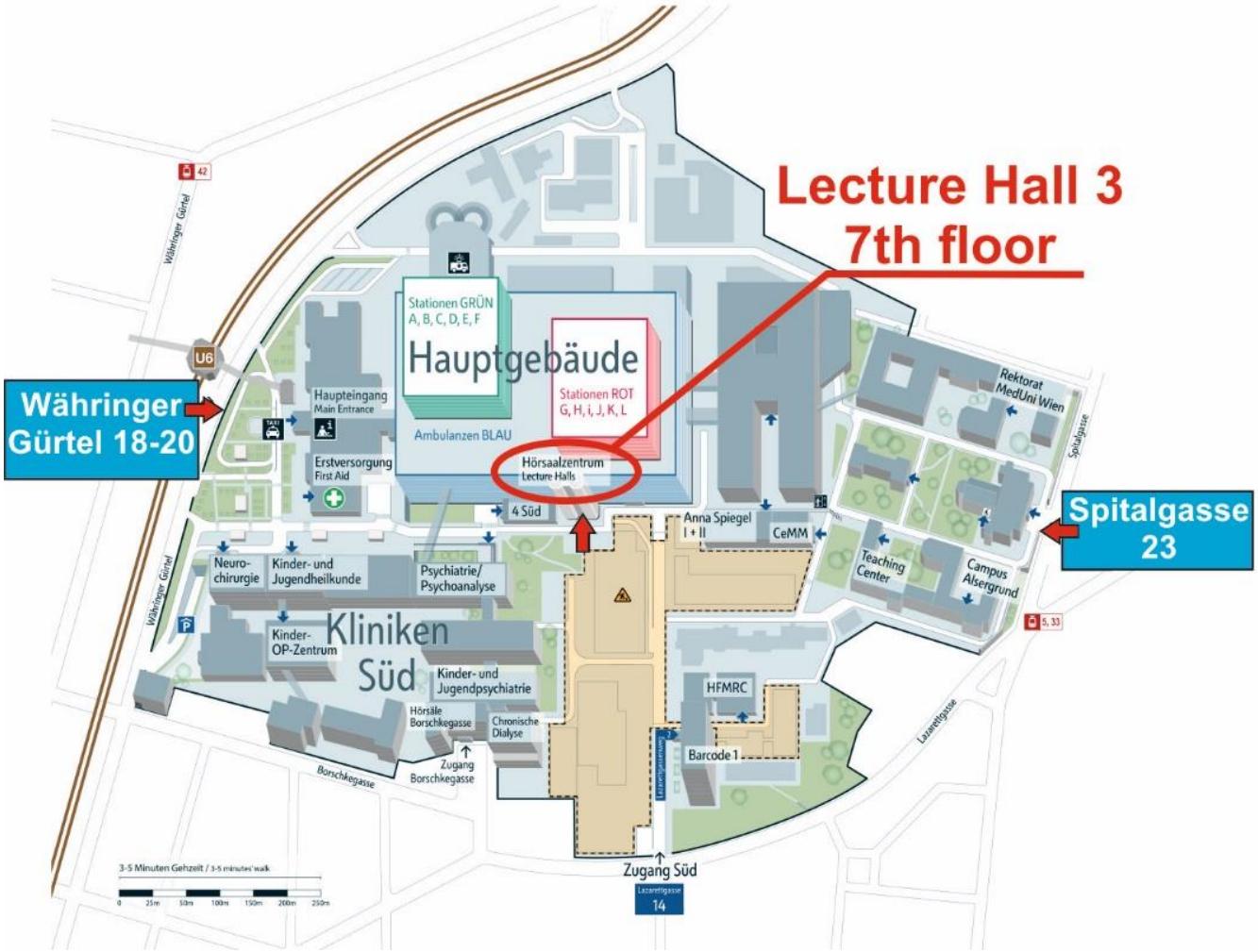
Time	Title	Lecturer	Duration (min)
10.00-10.05	Welcome Address *Medical University of Vienna, Austria*	Maria Sibilia	5
10.05-10.10	Topics of the Workshop *Medical University of Vienna, Austria*	Miroslav Misik	5
10.10-10.25	Importance of DNA Damage Biomarkers in Human Biomonitoring *Genome Health Foundation, Australia*	Michael Fenech	15
10.25-10.45	Biomarkers of Early Effect in Biomonitoring: a Long and Inspiring Journey *IRCCS San Raffaele Roma*	Stefano Bonassi	20
Occupation			
10.45-11.05	Using NanoSeq to Study Mutational Signatures of Carcinogens and Chemotherapeutics *GSK R&D, Hertfordshire, United Kingdom*	Halh Al-Serori	20
11.05-11.25	Use of Micronucleus Experiments with Buccal Cells in Occupational Biomonitoring *ASU Experts Graz, Austria*	Georg Wultsch	20
11.25-11.45	Use of the Comet Assay in Occupational Studies *NILU, Kjeller, Norway*	Maria Dusinska	20
11.45-12.05	Assessment of Occupational Exposures to Combustion-derived Carcinogens by Monitoring Urinary Metabolites of Polycyclic Aromatic Hydrocarbons (PAHs) *Health Canada, Ottawa, Canada*	Paul White	20
12.05-12.30	**Discussion**		
12.30-13.30	**Lunch**		
Nutrition, obesity and diabethes			
13.30-13.50	Dietary Intervention Studies with Humans *Medical University of Vienna, Austria*	Miroslav Misik	20
13.50-14.10	Impact of Overweight and Weight Loss on DNA Damage in Humans and Underlying Mechanisms *University of Wuerzburg, Germany*	Helga Stopper	20
Lifestyle			
14.10-14.30	Genotoxic and Cytotoxic Effects of Electromagnetic Fields *Medical University of Vienna, Austria*	Michael Kundi	20
14.30-14.45	Induction of DNA Damage in Smokers and Chewers *Yerevan, Armenia*	Armen Nersesyan	15
14.45-15.20	**Coffee break**		
Disease and therapy			
15.20-15.35	Periodontitis - DNA Damage and Cancer *Medical University of Vienna, Austria*	Maximilian Scharnagl	15
15.35-15.50	Use of Human Genotoxicity Methods to Assess the Radiosensitivity of Cancer Patients *Institute for Oncology and Radiology of Serbia, Belgrade, Serbia*	Ivana Matic	15

Time	Title	Lecturer	Duration (min)
Others			
15.50-16.10	Use of Telomere Lengths in Human Biomonitoring studies *University of Copenhagen, Denmark*	Peter Moller	20
16.10-16.25	Use of Micronucleus Experiments with Exfoliated Cells for the Detection of Bladder and Cervical Cancer Medical University of Vienna, Austria	Siegfried Knasmueller	15
16.25-16.40	Advancing Standardization in Occupational Monitoring Through Automated Microscopy *MetaSystems Hard & Software GmbH, Altlussheim, Germany*	Christian Schunck	15
16.40-18.00	Discussion/Closing Remarks	Siegfried Knasmueller	

For more detailed information please contact directly the organizers (Miroslav Misik miroslav.misik@meduniwien.ac.at)

How to get there

The AKH, located at Währinger Gürtel 18-20, 1090 Vienna, can be easily reached by public transport - from the underground station U6 Michelbeuern-AKH, a pedestrian bridge leads to the main entrance. The trams 5 and 33 (station Lazarettgasse) stop in front of the entrance Spitalgasse 23.



Contents

Using NanoSeq to examine mutational signatures of chemotherapeutics and carcinogens in human tissue-derived organoids

Halh Al-Serori

Oncology Research Unit, GSK R&D, Gunnels Wood Road, Stevenage, Hertfordshire, SG1 2NY

Distinct mutational signatures associated with exposure to specific environmental carcinogens (e.g. tobacco smoke) and chemotherapeutics (e.g. temozolomide [TMZ]) can be detected in the DNA of human tumours and normal tissues using next-generation sequencing. In order to better understand the mutations observed in people, we aim to characterise signatures induced by mutagenic exposures experimentally. We are currently using normal human tissue-derived organoids, along with a genome-wide, duplex sequencing technology (NanoSeq), to examine mutations caused by a panel of environmental and chemotherapeutic agents. NanoSeq enables highly sensitive, error-free detection of subclonal mutations, eliminating the laborious and time-consuming step of single cell cloning in experimental mutation assays. Using organoids derived from 4 tissues – stomach, colon, kidney and pancreas – treated with several carcinogens (aristolochic acid I [AAI], benzo(a)pyrene, aflatoxin B1 and 2-amino-1-methyl-6-phenylimidazo[4-5-b]pyridine [PhIP]), we found that all accumulated mutations as detected by NanoSeq. We extracted carcinogen-specific mutational signatures consistent with those previously identified using conventional whole-genome sequencing. Further, using this approach in gastric organoids treated with 30 chemotherapeutic agents, we identified a single base substitution (SBS) signature for TMZ that matches a signature observed in human tumours (COSMIC SBS11), as well as SBS signatures for mitomycin C and nitrogen mustard alkylating agents (e.g. chlorambucil).

Biomarkers of early effect in biomonitoring: A long and inspiring journey

Stefano Bonassi, PhD, ERT

Department of Human Sciences and Quality of Life Promotion, San Raffaele University, Rome, Italy; Unit of Clinical and Molecular Epidemiology, IRCCS San Raffaele Roma, Rome, Italy

Traditional occupational and environmental health risk assessments often rely on external exposure measurements, such as air monitoring, which may not fully capture the complexity of exposure. Human biomonitoring provides a more comprehensive assessment by measuring internal exposures or biological effects resulting from all potential routes of exposure (i.e., inhalation, oral, and dermal). Effect biomarkers, which quantify stressor-induced biological responses linked to disease mechanisms, offer a valuable tool for evaluating risks associated with chemical mixtures. These biomarkers help bridge the gap between exposure and health outcomes by capturing key events in the pathogenesis of cancer and major non communicable diseases, offering a strategic tool for prevention. Integrating effect biomarkers into biomonitoring programs enables the identification of at-risk subpopulations and supports the prioritization of risk management measures. Statistical analyses incorporating effect and exposure biomarkers, alongside demographic variables, can provide insights into chemical exposure trends, thereby improving risk assessment strategies. Advancing the application of effect biomarkers in regulatory risk assessment will facilitate harmonized approaches to occupational and environmental health protection. The presentation will discuss the development of genotoxic biomarkers, starting with their early utilization in exposure monitoring, exemplified by ionizing radiation, and then examine the growing evidence that identifies DNA damage and genomic instability as central mechanisms in the pathogenesis of many diseases, leading to the definition of biomarkers of effect. The next steps involve the validation of these biomarkers as biomarkers of risk, with possible use in clinical practice or in occupational safety.

Use of the Comet Assay in Occupational Studies

Maria Dusinska[1], Andrew Collins[2,3]

[1] Departmnet of Environmental Chemistry and Health, The Climate and Environmental Research Institute-NILU, 2007 Kjeller, Norway
[2] NorGenoTech, Oslo Cancer Cluster, Radiumhospitalet, 0379 Oslo, Norway
[3] Department of Nutrition, University of Oslo, 0316 Oslo, Norway

The comet assay is widely used in human biomonitoring as a sensitive method for detecting genetic damage, usually in peripheral blood mononuclear cells, but also in whole blood and buccal epithelial cells. The assay can be applied to frozen cells – meaning that samples do not need to be analysed immediately on collection. Incorporating a digestion with lesion-specific enzymes allows detection of oxidised and alkylated bases as well as DNA breaks. In combination with other biomarkers, comprehensive monitoring of acute and chronic exposure to genotoxins is possible. The capacity of cells to repair DNA damage can also be measured. The assay has been applied across a range of occupational settings, for example in workers exposed to pesticides, lead, formaldehyde, benzene, styrene, microwave radiation, and tobacco-related compounds; a consistent association is seen between specific occupational exposures and increased DNA damage, validating the assay's role in biomonitoring programs.

We assessed DNA damage and repair in 239 workers exposed to asbestos, stone wool, or glass fibres, along with 148 controls, using the comet assay. In 61 asbestos-exposed workers, compared with 21 controls, significantly higher levels of oxidised and alkylated DNA bases were found;DNA damage correlated with years of exposure - indicating persistent genotoxic risk. In 98 stone wool-exposed workers and 43 controls, exposed non-smokers showed more DNA strand breaks, but no specific base damage or change in DNA repair capacity, suggesting limited genotoxic impact. Among 80 glass fibre workers, even low-level exposure was linked to increased strand breaks and oxidation damage compared to 36 unexposed. DNA damage was influenced by antioxidant enzyme activity, while repair capacity showed an inverse relationship with damage. To summarise, mineral fibre exposure, even at low levels, can lead to measurable DNA damage; the comet assay can play an important role in identifying and mitigating genotoxic risks in exposed populations.

Importance of DNA damage biomarkers in human biomonitoring

Prof. Michael Fenech

University of South Australia, Adelaide, SA 5000, Australia
Genome Health Foundation, North Brighton, SA 5048, Australia

Damage to DNA is the most fundamental pathology which increases the risk of several human developmental and degenerative diseases. This knowledge has been accumulated over the past few decades using a wide range of DNA damage biomarkers both at the cytogenetic level (e.g. chromosome aberrations, micronuclei) and at the molecular level (e.g. DNA breaks, DNA deletions, point mutations, telomere loss). Many of these biomarkers have been shown to be robust indicators of DNA damage induced by important preventable conditions such as (i) deficiency in micronutrients required as cofactors for synthesis of nucleotides (e.g. folate, vitamin B-12) and for DNA replication and DNA repair (e.g. zinc, magnesium) and (ii) exposure to high levels of endogenous (e.g. methylglyoxal) or environmental genotoxins (e.g. ionising radiation). The importance of DNA damage biomarkers is that their use *in vitro* and *in vivo* makes it possible to determine which dietary and lifestyle factors need to be optimised and which environmental factors should be minimised to increase genome integrity in human populations. Importantly, we also need to use these biomarkers to understand better the interactive effects of chronic sub-optimal nutrition and simultaneous exposure to various genotoxins and build robust *in vitro* models for this purpose. Today's symposium provides an exciting program of presentations that illustrate the great potential of DNA damage biomarkers in identifying and preventing loss of genome integrity.

Use of micronucleus experiments with exfoliated cells for the detection of bladder and cervical cancer

Siegfried Knasmueller[1], *Michael Kundi*[1], *Miroslav Mišík*[1], *Franziska Ferk*[1], *Armen Nersesyan*[2]

[1]Medical University of Vienna, Vienna, Austria
[2]ASU-experts GesbR., Graz, Austria

Micronucleus (MN) experiments with exfoliated urothelial-derived (UDC) and cervical cells (CC) may be useful tools for the prediction and detection of cancer in these organs. UDC can be collected from urine by centrifugation, while CC can be scored in smears sampled in routine health checks. Increased frequencies of MN were found in UDC in populations consuming contaminated (e.g. As) drinking water, in occupationally exposed workers and also in individuals with bladder infections. These exposures are associated with increased cancer risks. Some investigations indicate that this approach can be also used the recurrence of bladder cancer after therapeutic measures. MN in CC were scored in few studies with smokers while a larger number of studies have investigated MN in patients with preneoplastic lesions and cancer. The results of a meta-analysis showed that the MN rates increased with the degree of neoplastic transformation. The highest frequencies were found in patients that were diagnosed cancer. Taken together, these results indicate that the MN test which can be performed in combination with routine screening (Pap-test) may be useful for the detection/prevention of cervical cancer.

Genotoxic and cytotoxic effects of electromagnetic fields

Michael Kundi

Department of Environmental Health, Center for Public Health, Medical University Vienna, Austria

Power frequency magnetic fields (PF-MF) resulting from generation, distribution and utilization of electrical current as well as radiofrequency electromagnetic fields (RF-EMF) used for telecommunication, broadcasting, radar tracking etc. have both been classified as possible human carcinogens by IARC. This classification was predominantly driven by epidemiological evidence. However, since the 1950s some in vivo and in vitro studies have indicated a genotoxic and carcinogenic potential of these agents. Some conflicting findings could be explained but the bottom line was often: results are inconsistent. During the 1970s to the 1990s a view became prevalent that these fields cause acute effects only and at high levels. This view was soon broadly adopted and became the fundament of exposure guidelines almost worldwide. For both types of fields many animal carcinogenicity studies have been conducted that gave ambiguous results partly attributable to methodological differences but also to a fundamental problem that has rarely been recognized. Briefly, none of the 'known' carcinogens would be positive in animal experiments if conducted in analogy to most of those performed in these fields. If we focus on in vitro genotoxicity studies, there are also ambiguities in the results that are difficult to explain since at least some of those were well conducted. Recently, we have completed a series of experiments that could shed light on the reasons for diverging findings. We have shown that both types of fields are able to induce nucleotide excision repair. A consequence is that exposure on the edge of the equilibrium between DNA damage and repair must result in huge ambiguity. Furthermore, we have shown that after in vivo human experimental exposure to a RF-EMF during one week no genotoxic but clear cytotoxic effects occur. Since chronic exposure to a cytotoxic agent could also increase cancer risk, this possibly offers a new mechanistic.

Use of human genotoxicity methods to assess the radiosensitivity of cancer patients

Ivana Z. Matić[1], Tatjana Stanojković[1], Nina Petrović[2,1], Marina Nikitović[1,3], Siegfried Knasmueller[4], Miroslav Mišík[4]

[1] Institute for Oncology and Radiology of Serbia, Belgrade, Serbia
[2] "VINČA" Institute of Nuclear Sciences-National Institute of the Republic of Serbia, University of Belgrade, Belgrade, Serbia
[3] Faculty of Medicine, University of Belgrade, Belgrade, Serbia
[4] Center for Cancer Research, Medical University of Vienna, Austria

A variety of different methods have been used to investigate the radiosensitivity of cancer patients treated with radiotherapy in order to predict the risk for developing adverse effects, improve the therapeutic success, and patient's quality-of-life. The most investigations were realised with peripheral blood cells from head and neck cancer, breast cancer, and prostate cancer patients. The irradiation response of patient-derived lymphocytes has been associated with acute and late normal tissue reactions to radiotherapy. The approaches which were used include measurements of cytotoxic effects (including apoptosis and cell cycle delay), identification and analyses of specific genes and use of methods which reflect genotoxicity and DNA repair. The most frequently employed approaches that detect damage of the genotoxic material which is induced by ionizing radiation (double strand breaks and oxidative damage) are chromosomal aberration (CA) analyses of metaphase cells, micronucleus (MN) experiments detecting structural and numerical CA, γH2AX assays which can be used to measure double strand breaks, and single cell gel electrophoresis (SCGE) assays reflecting single and double DNA strand breaks. Individual differences in the repair capacity were mainly studied in SCGE experiments in which the time kinetics of the disappearance of "comets" was measured. Overall the results of these studies are highly controversial, possibly due to inconsistent experimental designs and methodological shortcomings (including small study groups). However, the possible correlations between repair kinetics of radiation-induced DNA damage in cancer patient lymphocytes after irradiation and the severity of radiotherapy adverse effects should be further explored in larger cohorts. In the last years standardised/validated protocols have been pushed for MN, SCGE, γH2AX, and DNA repair (BER and NER) experiments with humans that will be used in an ongoing Horizon Europe Twinning project (RadExIORSBoost 101158832) to identify biological parameters which reflect the individual normal tissue radiosensitivity of patients with prostate cancer undergoing radiotherapy.

Acknowledgments: "This Project is funded by the European Union, under Horizon Europe programme Grant agreement number 101158832. Views and opinions expressed are however those of the author(s) only and do not necessarily reflect those of the European Union or European Research Executive Agency (REA). Neither the European Union nor the granting authority can be held responsible for them."

Dietary intervention studies with humans

Miroslav Mišík[1], Armen Nersesyan[2], Michael Kundi[3], Franziska Ferk[1], Siegfried Knasmueller[1]

[1]Center for Cancer Research, Medical University of Vienna
[2]ASU-Eperts group GesbR., Graz, Austria
[3]Center for Public Health, Department of Environmental Health, Medical University of Vienna

DNA damage is a key factor in the development and origin of cancer, neurodegenerative disorders, infertility, and accelerated aging. Optimizing nutrient intake offers a promising strategy to mitigate DNA damage and enhance genome integrity. Over the past 25 years, more than one hundred human intervention studies concerning nutrition and its protective effects against genotoxic damage have been conducted. Most of these studies utilize the single-cell gel electrophoresis (comet assay), which quantifies DNA migration in an electric field. This assay has emerged as a vital tool and has been employed in approximately 90 human intervention trials. Other assays, such as the micronucleus test in lymphocytes or buccal cells, are also used, though less frequently. Overall, the protective effects were observed in half of the studies, with pronounced benefits from plant foods (spinach, kiwi, onions), coffee, green tea, honey, olive oil, and vegetable-rich diets. Small doses of specific phenolics (e.g., gallic acid, xanthohumol) reduced oxidative DNA damage. Randomized controlled trials and longitudinal studies have demonstrated that supplementation with micronutrients such as folic acid, vitamin C, zinc (Zn), and selenium (Se) reduces biomarkers of DNA damage, including chromosomal aberrations, oxidized bases, and telomere attrition. Despite promising results, methodological shortcomings in many studies—such as inadequate controls, uncalibrated repair enzymes, and poor statistical rigor—highlight the need for standardized protocols. Future research should prioritize high-quality human trials to refine dietary recommendations for DNA damage prevention. Collectively, these findings underscore the potential of targeted nutrient intake to improve genome maintenance and reduce disease risk.

Use of telomere length in human biomonitoring studies

Peter Møller

University of Copenhagen, Department of Public Health, Section of Environmental Health, Copenhagen, Denmark

The chromosome ends (i.e. telomeres) consists of a repetitive non-coding nucleotide sequence (TTAGGG in humans), which forms a loop, supported by shelterin proteins. This serves to avoid it from being detected as a double strand break. The telomeric sequence is added to DNA by telomerase, which is active in proliferating cells and silenced in somatic cells. While telomeric DNA functions to avoid loss of vital information during replication of DNA (i.e. end-replication problem), replication of cells with low telomerase activity will lead to a gradual loss of telomere length. In vitro studies have shown that cells with too short telomeres go into replicative senescence (or apoptosis). Alternatively, cells may go through a crisis and become immortalized, typically with reactivated telomerase activity. Studies from numerous studies have shown an age-associated shortening in telomere length in human leukocytes. Observations from cross-sectional studies indicate a large inter-individual variation in telomere length, which depends on genetics, age, sex, life-style factors, diet and external exposures. A number of studies have assessed the role of external exposures on telomere length in human leukocytes, although without showing consistent results on the same type of agents such as air pollution, heavy metals and persistent organic pollutants. Lastly, studies on associations between leukocyte telomere length and diseases suggest that some diseases (e.g. cardiovascular diseases) is associated with short telomeres, whereas at least cancers are associated with long telomeres. In summary, research is still needed to link environmental/occupations exposures, telomere length, and risk of disease and mortality in humans.

Induction of DNA damage in smokers and chewers

Armen Nersesyan[1], *Michael Kundi*[2]. *Miroslav Mišík*[2], *Franziska Ferk*[2], *Siegfried Knasmueller*[2]

[1]ASU-experts GesbR., Graz, Austria
[2]Center for cancer Research, Medical University of Vienna, Vienna, Austria

Numerous studies show that cytogenetic experiments can be used to assess the contribution of chromosomal damage caused by tobacco smoking and to identify factors which have an impact on the individual risks. Most micronucleus (MN) experiments with exfoliated cells showed that cigarette smoke causes chromosomal damage and acute cytotoxicity, but clear effects were only seen in heavy smokers. We showed that the introduction of inadequate (non-DNA-specific stains) leads to an overestimation of the effects, and that the MN rates increase with the amount of tar, while the nicotine content of the cigarettes was inversely related to chromosomal damage. Another relevant factor that increases the effects is the duration of cigarette consumption (pack-years). We have showed that smoking has severe effects on the cervical cells of women regardless of their hormonal status. Studies with water pipe smokers yielded generally clear positive results. In individuals consuming electronic cigarettes, evidence of the induction of chromosomal damage and cytotoxic effects was found in three studies. Also betel chewing leads to MN induction and to acute cytotoxic effects, regardless if the chewers consumed the nuts and leaves with or without tobacco. No data are currently available regarding the use of nicotine patches. We also conducted studies, with preparations of plant materials, except tobacco, and found higher MN rates in khat chewers in Ethiopia, while no adverse effects were detected in a study, which was conducted among coca chewers in Peru.

Periodontitis - DNA Damage and Cancer

Maximillian Scharnagl[1], *Miroslav Mišík*[1], *Siegfried Knasmueller*[1], *Armen Nersesyan*[2]

[1]Medical University of Vienna, Vienna, Austria
[2]ASU-Eperts group GesbR., Graz, Austria

Periodontitis (PD) is a widespread, chronical, multifactorial and inflammatory disease of the oral cavity. About half of the population in Europe are affected and a severe stadium was diagnosed in approximately 7% (worldwide). Several studies indicate that PD patients have an (up to 5-fold) increased risk for oral cancer. Furthermore, it was reported that also the prevalence of other forms of cancer (colorectal cancer, prostate cancer, liver cancer, etc.) is significantly higher. Cytogenetic studies and micronucleus (MN) experiments with exfoliated buccal and gingival cells which reflect chromosomal damage (structural and numerical aberrations) indicate that these effects may be due to damage of the genetic material which plays a key role in neoplastic transformation. However, the overall results which have been published so far are controversial. Clear positive findings were reported 8 out of 14 studies. In some investigations additional nuclear anomalies were evaluated which reflect acute cytotoxic effects and an increase of these parameter was found in most investigations. It was postulated that the DNA damaging and acute toxic effect are due to release of bacterial toxins that cause inflammations and as a consequence release of reactive oxygen radicals. Notably higher levels of inflammatory cytokines and lipid peroxidation products were found in PD patients. Some studies indicate that MN rates in individuals with PD are associated with alcohol consumption and diabetes, which are known risk factors for the disease. The discrepancies of the results of MN studies may be due to methodological shortcomings, i.e. to the use of inadequate stains, and invalid diagnosis (lack of data concerning the severity of the disease). We are currently realizing comprehensive investigations to find out which stages of the disease lead to chromosomal damage and acute cytotoxicity and also if therapeutic measures can reduce the toxic effects.

Advancing Standardization in Occupational Monitoring Through Automated Microscopy

Christian Schunck[1], *Siegfried Knasmueller*[2], *Armen Nerseyan*[3], *Tilman Johannes*[1]

[1]MetaSystems Hard & Software GmbH, Robert-Bosch-Str. 6, 68804 Altlussheim, Germany
[2]Center for Cancer Research, Medical University of Vienna, Austria
[3]ASU-experts GesbR., Graz, Austria

The continuous evaluation of workplace risks to employees' genetic material is a fundamental aspect of occupational medicine. Genetic toxicology employs various assays that quantify biological markers, such as chromosomal aberrations and micronuclei, primarily through microscopy. Although numerous studies have validated the reliability of these assays, and several are incorporated into OECD guidelines, standardization remains a challenge due to the predominance of manual evaluation. Automated microscopy presents a promising approach to improving the standardization of test evaluation in genetic toxicology. While the concept itself is not new, and automated systems for evaluating established genetic toxicology assays are already utilized in fields such as preclinical pharmaceutical testing, their application in occupational exposure monitoring has yet to be established. Recent advancements, including the integration of artificial intelligence through Deep Neural Networks (DNN), offer new perspectives on the potential of automated microscopy in this domain. Using the Metafer slide scanning platform software from MetaSystems as an example, we will provide an overview of the feasibility of these technologies in occupational monitoring. Where possible, preliminary results from ongoing studies employing the cytokinesis-block micronucleus assay will be presented. We propose that the automation of microscopy-based evaluations, guided by well-defined assessment parameters, can substantially contribute to the standardization of genetic toxicology assays recommended for occupational monitoring. This hypothesis is supported by evidence from related fields facing similar standardization challenges, such as preclinical pharmaceutical testing and radiation protection, where automation has already demonstrated significant benefits, as extensively documented in the literature.

Impact of Overweight and Weight Loss on DNA Damage in Humans and Underlying Mechanisms

Helga Stopper

Department of Pharmacology and Toxicology, University of Würzburg, Würzburg, Germany

Overweight including obesity is a growing global health concern and a well-established risk factor for various cancers and other diseases. While cancer development is a long-term process, taking years to decades to manifest after the beginning of exposures or circumstances with increased cancer risk, DNA damage alterations can be detected without delay, providing a valuable biomarker for assessing cancer risk.

Several established methods in human biomonitoring, including micronucleus formation, comet assay, and γH2AX detection, allow for the assessment of DNA damage at the cellular level in human samples. The most often used cells are human peripheral blood lymphocytes.

Research, including our own findings, consistently shows increased DNA damage in individuals with overweight or obesity compared to those with a healthy weight. This DNA damage is likely driven by oxidative stress, chronic inflammation, hormonal imbalances, the dysregulation of pro-inflammatory cytokines and compromised DNA repair mechanisms. Importantly, for obesity, weight loss has been found to be associated with a significant reduction in DNA damage, which may contribute to a lower cancer risk. Understanding the molecular mechanisms involved could enhance obesity-related cancer risk characterization and prevention strategies.

Assessment of Occupational Exposures to Combustion-derived Carcinogens by Monitoring Urinary Metabolites of Polycyclic Aromatic Hydrocarbons (PAHs)

Paul White

Environmental Health Science and Research Bureau, Environmental and Radiation Health Sciences Directorate, Health Canada, Ottawa, Ontario, Canada

Polycyclic aromatic hydrocarbons (PAHs) are organic compounds characterized by two or more fused aromatic rings. They are formed by incomplete combustion of organic matter and ubiquitous environmental contaminants. They are commonly detected in urban air, vehicle exhaust, grilled and roasted foods, tobacco smoke, power plant emissions, forest fire emissions, and a variety of occupational settings. Exposures to PAHs can occur via inhalation, dermal contact, or incidental ingestion, with food being a major source of PAH exposure for the general population. Following exposure and absorption, PAHs are initially metabolised by cytochrome P450 isozymes, with primary oxidation by-products (i.e., epoxides and phenols) being further metabolized to quinones and diols. Secondary metabolites are subsequently conjugated to endogenous substances such as glucuronide and glutathione prior to excretion via the urine or faeces. Importantly, the oxidized metabolites of some PAHs (e.g., benzo[*a*]pyrene) are DNA-reactive, contributing to the formation of bulky adducts and subsequent genotoxic effects (e.g., chromosomal damage and mutations). Additionally, numerous PAHs are known or suspected carcinogens. Biomonitoring to assess PAH exposure and internal dose can be accomplished by monitoring the concentration of secondary metabolites in bile and urine; with the latter being used to assess human exposure. Indeed, monitoring of urinary PAH metabolites is commonly used to assess human exposures to PAHs in occupational settings. For example, monitoring of urinary 1-hydroxypyrene has been used to assess PAH exposures in people engaged in metal refining and founding, roofing and paving, coke production, wood preservation, coal-tar distillation, and firefighting. Exposures during firefighting are of particular interest in light of firefighters' increased cancer risk relative to the general population. Indeed, numerous studies have used urinary hydroxypyrene biomonitoring to assess PAH exposures of municipal and wildland firefighters. Of particular interest is the efficacy of interventions (e.g., dermal decontamination and respiratory protection) to reduce firefighters' PAH exposure levels.

Use of Micronucleus Experiments with Buccal Cells in Occupational Biomonitoring

Georg Wultsch[1,3], *Michael Kundi*[2], *Miroslav Mišík*[2], *Siegfried Knasmueller*[2], *Armen Nersesyan*[3]

[1]KNAPP AG, Graz, Austria
[2]Medical University of Vienna, Vienna, Austria
[3]ASU-Eperts group GesbR., Graz, Austria

MN experiments with buccal cells are increasingly used in occupational studies. They have the advantage that the collection of the cells is not invasive and that no cultivation is required. In total, 288 studies have been published with different groups of workers in the last four decades. Most studies were realized with agricultural workers, miners, medical staff, painters, petrol station attendants and metal production workers. The most pronounced effects were observed with metal production workers followed by miners and painters. The analysis of the published articles shows that some of them have methodological shortcomings, in particular lack of the control of confounding factors (nutrition), use of inadequate stains, scoring problems and lack of chemical exposure measurements. The publication of guidelines, picture galleries and inter-laboratory validation experiments has led to a substantial improvement of the quality of the studies in the last years. We realized in Austria a number of human studies with different occupational groups. The findings show that the results depend on the specific occupational settings. No positive results were obtained with electroplaters, workers that are exposed to animal manure and welders, while positive findings were obtained with carpenters. Furthermore, we observed for the first time a pronounced effect in road markers that are inhalatively exposed to silica dust and different reactive chemicals. Micronucleus reflect structural and chromosomal aberrations, which play a key role in the etiology of human cancer. At present the surveillance of workers is based solely on chemical analytical exposure measurements, which do not reflect synergistic effects, which can be detected in experiments.