Report from

COST-Action No.15129: 
**DiMoPEx (Diagnosis, Monitoring, Prevention of Exposure Related Non-Communicable Diseases)**

**Work Group (WG) 5**

**Genotoxicity and Susceptibility**

**WG Leader:** Dr. Nancy B Hopf

**WG Expert Support:**
- Prof. Dr. Claudia Bolognesi
- Assoc. Prof. Dr. Heidi Schwarzenbach
- Prof. Dr. Fiorella Belpoggi
- Dr. med. Daniele Mandrioli
Aims

➢ identify determinants of prevalence and morbidity of diseases and of developing promising preventive intervention strategies

➢ to establish quantitative relationships between exposures to toxic hazards and the associated risks of the diseases

Generate risk assessment and prevention models to improve health and safety in Europe for the general public, and more specifically for workers, and for consumers
Focus on BIOMARKERS

- **Exposures**
  - environmental
  - occupational
  - Assessment:
    - ambient monitoring
    - human biomonitoring
    - biol. effect monitoring
  - co-exposures
  - life style
  - susceptibility
  - confounding

- **Specific questionnaires**
  - specific parameter
    - nicotin-/alcohol abuse (biomarker, i.e. cotinin)
    - genetic-/ epigenetic biomarker

- **Toxicology**
  - PBK modeling
  - mathematical modeling
  - estimation of variant components
  - bias

- **Correlation**
  - preclinical lesions
  - NCD diagnosis

- **Ethical aspects**
  - risks communication
  - human data
  - biobanks
Goals

➢ Creation of a network of laboratories involved in the application of genotoxicity and epigenetic biomarkers. (CB MN training, FB DimoPEx conf)

➢ Standardization of the procedure for the application of the selected genotoxicity/epigenetic biomarkers

➢ Intercalibration exercises on specific biomarkers applied in multiple labs. (CB MN training)

➢ Identification of specific biomarkers of cancer in the DiMoPEx WG 5 (ongoing…)

➢ Evaluation of potential use of genotoxicity/epigenetic biomarkers in evaluation of risk assessment of chemicals. (NBH and CB meta-analysis)
Creation of a network of laboratories

➢ Training courses

➢ Exchange of protocols/guidelines expertise:
  ✓ Criteria of scoring
  ✓ Automated methods
  ✓ Analysis of the data
  ✓ Interpretation of the results

➢ Implementation of the in vitro/in vivo models
In vitro Evaluation of the Genotoxic Properties of Pencycuron, a Commonly Used Phenylurea Fungicide, by the Cytokinesis-Block Micronucleus (CBMN) Assay

Karoly Nagy, Balazs Adam
University of Debrecen, Faculty of Public Health, Department of Preventive Medicine, Debrecen, Hungary

Claudia Bolognesi
Ospedale Policlinico San Martino IRCCS, Environmental Carcinogenesis Unit, Genoa, Italy

In vitro mammalian cell MN test (OECD guideline 487, 2010) in the basic battery of tests to screen new chemical and physical agents for genotoxicity.
Frequency of MNBN cells/1000 cells induced by exposure to different concentrations of pencycuron in human peripheral lymphocytes and HepG2 cell line

Results: provides some indication on a possible genotoxic effect of pencycuron as observed in vitro in two different human cell types in two laboratories.
In vivo micronucleus assay in bone marrow of Sprague-Dawley rats

Pilot study on glyphosate/Roundup toxicity

Environmental Carcinogenesis Unit, Ospedale Policlinico San Martino, Genova, Italy

Ramazzini Institute, Cesare Maltoni Cancer Research Centre, Bentivoglio, Bologna, Italy

treated from the embryonic life up 120 PND at the US ADI (1.75 mg/kg/day).
MICRONUCLEI TOXICITY COHORT – results so far…

6 week

13 week
Biological monitoring of workers exposed to carcinogens using the buccal micronucleus approach: A systematic review and meta-analysis

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Background

Micronucleus (MN) (extranuclear DNA-containing bodies) frequencies in buccal cells harvested by scraping the inside of the cheeks with a cytobrush proposed to monitor workers exposed to aero-digestive carcinogens for early detection of occupational cancer.

Assessment of the non-invasive MN approach

- What is the extent of MN frequency increase in occupationally exposed over non-exposed populations across studies published in the scientific literature for buccal cells; and

- Which types of occupational exposures give relevant summary MN ratios across studies published in the scientific literature.

A systematic literature review was performed, and the MN frequency ratios for buccal cells were calculated for each occupational study.
**silica** : FR 9.02 95%CI 6.83-11.93

**formaldehyde** : FR 4.81 95%CI 2.19-10.60

**hairdressers** : FR 7.06 95%CI 3.96-12.57
Results

The meta-analysis of the published literature (n=96 studies) showed large random variability between studies within each category ($I^2 > 90\%$),

- except where exposures were to a single carcinogenic substance (e.g. formaldehyde, silica) or
- similar jobs were performed (e.g. antineoplastic dispensing, gasoline attendants).

The unexplained between-study variability can be related to exposures to mixtures of carcinogens with unknown mechanisms.

Conclusion

**Better exposure assessments** are needed to understand the *relationship between occupational exposures and MN frequency responses in buccal cells*.

Our analysis indicates that the buccal cell MN assay is a promising tool for surveying workers exposed to genotoxic agents.
Report from
DiMoPEX Conference Working Group5 Meeting

“Biomarker of effect and latest developments in carcinogenicity testing”

Meeting Organizer: Nancy Hopf,

28-30 October 2019

Venue: Cesare Maltoni Cancer Research Center
Ramazzini Institute, Bentivoglio, Italy
via Saliceto 3, Bentivoglio (BO), 40010
https://www.ramazzini.org/en/how-to-get-there/

Local organizer:
Fiorella Belpoggi, Daniele Mandrioli, Fabiana Manservisi
WG 5 meeting outcome

• Submit a Commentary article from the Workshop
• Present the summary of the discussion on Cancer Biomarkers
  – current knowledge on the biology of the biomarker,
  – description of the recommended protocol for the biomarker,
  – possible applications of the assay, and
  – conclusions on knowledge gaps and future developments.
• Structure the commentary as a question and answer for each biomarker to facilitate the reading of these diverse cancer biomarkers.
The following 13 questions were used:

• What is the mechanism (AOPs or MOAs) of this cancer biomarker?
• Is “the biomarker” associated with development of cancer?
• How is “the biomarker” determined?
• How can “the biomarker” be used as surveying exposures to carcinogens?
• How do lifestyle factors affect “the biomarker”?
• Is age related to your biomarker?
• What are possible applications of “the biomarker”?
• How can “the biomarker” be applied as a clinical biomarker?
• How can “the biomarker” be used as a population screening tool?
• What are the limiting factors for applying “the biomarker”?
• What susceptibilities do we need to be aware of when using your biomarker?
• What research is needed to apply “the biomarker” as an individual screening tool?
• What are the ethical issues implement “the biomarker”?
Micronuclei frequency (MN) in blood [JP Teixera with P Kossomenos, C Bolognesi, S Bonassi]
Micronuclei frequency in Buccal cells [NB Hopf with C Bolognesi, A Paschalidou, and Bonassi]
Comet Assay [M Ghosh with K Nagy, J Puiso]
Aneuploidy (CA) [author missing]
Histone phosphorylation [author missing]
Telomere length [author missing]
DNA adducts [author missing]
DNA repair capacity [L Roisman]
Ortho-Tyrosine from EBC [H Moldovan and R Duca with A Aykut]
Breath analysis for lung cancer [P Scheepers]
Oxidative stress biomarkers and carcinogens [H Moldovan and V Valdiglesias]
Epigenetic biomarkers of cancer / GSTP1 methylation [R Duca]
mRNA exposome [H Schwartzenbach]
Fungal exposure and cancer [T Damialis, L Castelyn]
Immunomarkers and cancer [Stephanie Gilles, Laila Roisman]
Bioassays and cancer [Fiorella Belpoggio, Fabiana Manservisi]
Occupational physicians role in interpreting biomarkers of exposure and effect [Doina Giurgiu, Karel Vandamme, Oxzlem Gokzel, Diana Adliene, and Stelian Ioan Morariu]
Calculation of global burden of cancer [Daria Sgargi, Balasz Adam]
MN for buccal cells Q&A written and is to review

Co-other authors will have to write their paragraphs

 Invite WG5 members (not attending the meeting) to contribute as co-authors for their specific cancer biomarker

Submit manuscript

WG 5 summary

• Training school to standardize laboratory protocols
• Collaboration across countries and institutes possible from the networking meetings
• Review of effect biomarkers for carcinogenic exposures