

Use of molecular biomarkers to assess exposure to harmful agents at work

142,400 people die each year in the EU from occupational diseases and 8,900 others from accidents. Up to one third of these 142,400 deaths each year are attributed to hazardous substances at work including asbestos (21,000 deaths) and heavy metals.

6% of the working population in the EU is involved in maintenance work. The majority of workers in the service sector are men (about 90% in France and 65% in Spain). The largest age group is that of 30-49 years. Industrial maintenance employees have an 8–10 times *greater* chance of developing an **occupational disease** than the average population.

Maintenance workers often have contact with vapour or gases, particles (dust, smoke), fibres (asbestos, glass fibre) and mists. During typical maintenance, the technicians come in contact with chemical substances including asbestos and heavy metals. This triggers a mechanism that may result in the development of occupational diseases as shown in the following figure.

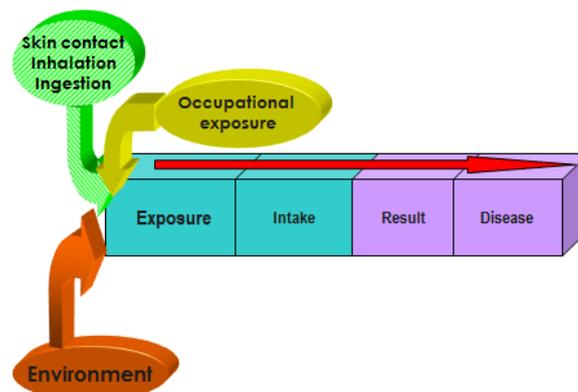
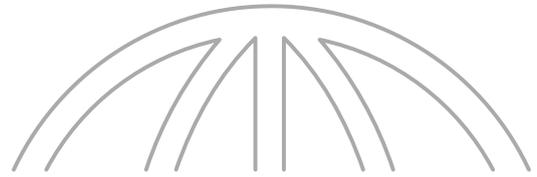


Fig.1: The mechanism of occupational disease exposure for maintenance activities

Most maintenance activities are high-risk by default like metal degreasing, painting, surface preparation, welding, lubrication and use of toxic substances (PCB's, CFC's etc.).



The main goal for the protection of employee health & safety through prevention occupational diseases is: **Early Detection by health monitoring.** There is a scientific way to achieve early detection in medicine which is called a **biomarker**. Biomarkers are molecular tools that can be used to identify changes or effects that occur in the body as a result of exposure to any given toxicant.

Bio-markers should ideally be sensitive enough to identify incidents at a stage when harm to the health is still reversible or to help prevent further deterioration of health. In addition, they should be as invasive as possible and easily measurable. The search for appropriate biomarkers can help to assess the potential carcinogenic risks associated with exposure to harmful cytotoxic agents.

At this stage it is necessary to introduce another definition that of the **micronucleus**, a nuclear material that is distinct from the main nucleus and within the cytoplasm (fig.2). Sparing the deep scientific details, micronuclei may serve as an indication of tissue damage in workers exposed to carcinogens, so they can serve as a biomarker.

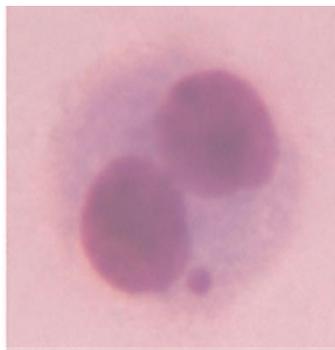


Fig.2: A micronucleus as seen on the microscope, formed during cell division

However, there is a relatively new research method experimentally developed in Greece which can predict predisposition trends for lung cancer development among workers exposed to carcinogenic substances. This technique is called micronucleus biomarker comet assay.

The technique is based on the fact that when carcinogenic cells are subject to a separation technique called “electrophoresis” and then are painted with fluorescent material called “fluorescent staining”, then the damaged cells can be separated. When observed under the microscope, the damaged cells look like a comet; the longer the tail, the more damaged the cell is (figure 4). This method is therefore called “Comet Assay Overview” as shown in figure 3.

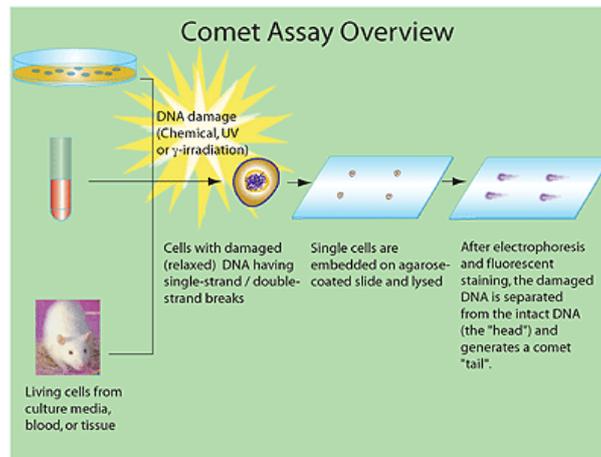
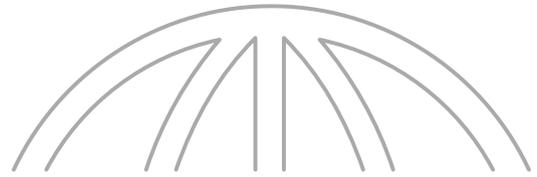


Fig.3: The Comet Assay Overview technique

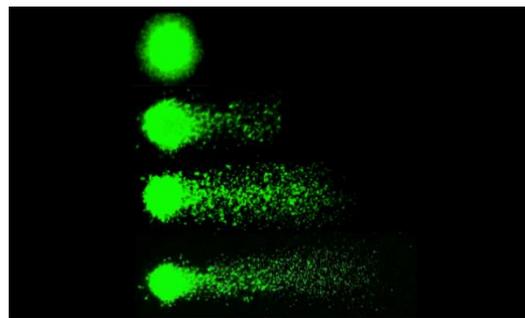
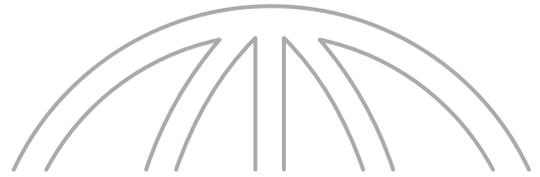


Fig.4: Various cells under Comet Assay Overview after the electrophoresis

The above micronucleus assay in cells of the oral epithelium is an innovative technique that could predict genotoxicity, which is promising for the study of carcinogenic epithelium. It can be a very useful biomarker and potentially provides a unique model for research of mutations allows the genetic alterations. The method is a quick and minimally invasive technique which is able to identify the genetic defect in the DNA, chromosomal instability and cell death.

This method is being tested for maintenance workers in a metal industry in Greece aiming to investigate possible alterations in the genetic material of maintenance workers in a mining and processing of nickel. The IARC (International Agency for Research on Cancer) classified soluble forms of nickel in group 1, and metallic nickel and nickel carbonyl as potentially carcinogenic to man in category 2B.

The study focuses on assessing the percentage of induced micronuclei as biomarkers, as well as the observation of changes in various cell groups in the epithelium of the oral mucosa. To compare the induced alterations in the genetic material of non-smoking employees compared to smokers a sample of 61 workers was used. The individuals were separated into two groups: a) employees in the production procedure of Ni and b) white collar workers (office services - no contact with the dust of the production process). Each of these groups was then divided into smokers and non-smokers.



The sampling of the cells from oral epithelium took place after the end of the shift, by means of a manual toothbrush which has rotated ten times with light pressure on the inside of the cheek. Samples of the oral epithelium were collected in sterile tubes containing a suitable Buffer and properly transferred to further processing according to the protocol for micronuclei (Thomas et al. 2009; Tolbert et al. 1992).

Since the study is under way, results are pending but the objective is to determine whether the exposure limits (TWA, OEL, STEL and CL) must be revised and also assist the Occupational Physician prescribe medical examinations to early-detect carcinogenicity trends among high-risk employees.

The EFNMS's Health, Safety & Environmental Committee (EHSEC) has addressed an invitation to all National Maintenance Society Members to contribute by providing specimens for laboratory testing in order to contribute to this effort.