

***In vitro* characterization of genotoxic damage induced by various PM sources on the bronchial epithelial cell line BEAS-2B**

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Particulate matter (PM) is a complex air pollutant, comprising both particles and gases, whose presence negatively affects individuals' life quality and constitutes a major risk factor for health worldwide. It is composed by a particles' population characterized by a variety of physiochemical characteristics (e.g. size, composition, aerodynamic behaviour) and sources.

The wide spectrum of adverse health effects occurring after PM exposure is also reflected at cellular level in the activation of many different toxicity mechanisms. The variety of the latter can suggest that each source may be responsible for specific kinds of cellular damage. To validate this hypothesis, 5 different sources of PM were evaluated in the present study: a positive control DEP NIES no.8 (D), coke dust (C), pellet ashes (PA), incinerator ashes (IA), and brake dust (BD); these sources were previously characterized and are all comprised in the PM₁₀ class.

To deeply investigate the genotoxic potential of the different PMs, an *in vitro* investigation was performed using the human bronchial epithelial cell line BEAS-2B. Cells were treated for 24 hours with increasing concentrations of PM (25, 50, 100, and 150 µg/mL). Previous analyses confirmed the absence of cytotoxicity at all concentrations tested. The modified alkaline Comet assay was used in combination with three endonuclease enzymes (ENDOIII, FPG, and ENDOV) to recognize oxidative or direct damage on the DNA strand. PM treatments induced an increase of DNA damage at all concentrations tested, compared to controls. The characterization of the genotoxic damage revealed that all the sources of PM are particularly active in inducing oxidation of the DNA bases. Analysing each source allowed to highlight some peculiarities as the oxidation of purines (adenine and guanine) identified by FPG treatments, that results to be particularly increased in samples treated with D, C, and BD; while ENDOIII highlighted the presence of oxidised pyrimidines in samples treated with PA and IA. Moreover, the involvement of direct damage, in the form of cyclobutane pyrimidine dimers, at the higher dose of treatment with D was recognized by ENDOV.

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