

## Introduction

Between Aix-en-Provence and Marseille, the former coal mining area constitutes a typical peri-urban zone including mining industries and heavy traffic. Fine atmospheric particles (PM<sub>2.5</sub>) reaches high levels in this mining area of Provence.

PM<sub>2.5</sub> can lead to cardiovascular diseases, impairment in lung function and life expectancy decrease. These physiological disorders are due to various mechanisms of action including genotoxicity.

**Aim of the study: Chemical profile and chromosome damaging effects of the PM<sub>2.5</sub> lipophilic phase**

## Materials and methods

**Cells:** Normal Human Lung Fibroblasts (NHLF)

**Sampling:** High flow sampler, for a week in summer and winter

**Chemical characterization:** ICP-MS for metal composition, GC-MS analysis for the 16 EPA-PAHs

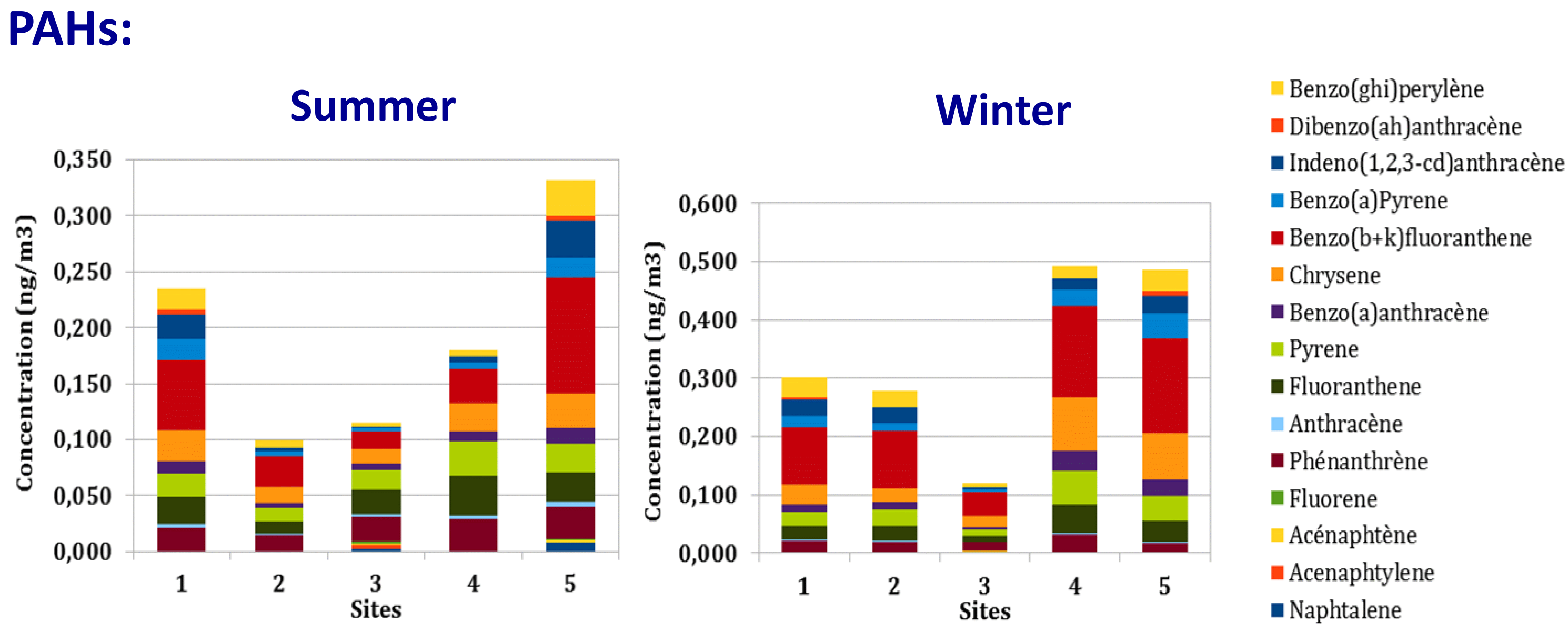
**Extraction:** Lipophilic phase extracted from filters by dichloromethane, then dissolved in DMSO at 5mg/ml

**Genotoxicity test:** Cytokinesis-block micronucleus (CBMN) assay + CREST antibodies (centromeric labeling to discriminate aneugenic and clastogenic events) ± S9mix (metabolic activator)

NHLF were exposed to the lipophilic PM<sub>2.5</sub> fraction for 3 hours and 24 hours respectively with and without S9mix.

## Results

**Metals:** Summer DMSO fractions contained 3 metals: Cu > Zn > As [3416 – 5303 ppb]; Zn proportion was higher at Bouc-Bel-Air site. Winter DMSO fractions contained the same 3 metals [6131 – 20201 ppb], and lower amounts of Cr, Ni, V, Al; Fe was found on Bouc-Bel-Air and Gardanne sites, and Ti on all sites except Bouc-Bel-Air.



**CBMN + CREST:**

Season	S9mix	Bouc Bel Air		Gardanne		Meyreuil		Fuveau		Trets	
		C+MN	C-MN	C+MN	C-MN	C+MN	C-MN	C+MN	C-MN	C+MN	C-MN
Summer	Without	+	-	-	-	-	+	-	-	+	-
	With	-	-	-	-	-	-	-	+	-	+
Winter	Without	+	-	-	-	+	+	-	-	-	+
	With	-	-	-	+	-	+	-	+	-	+

C+MN: Micronucleus with centromeric labeling, loss of chromosome = aneugenic event

C-MN: Micronucleus without centromeric labeling, loss of chromosome fragment = clastogenic event

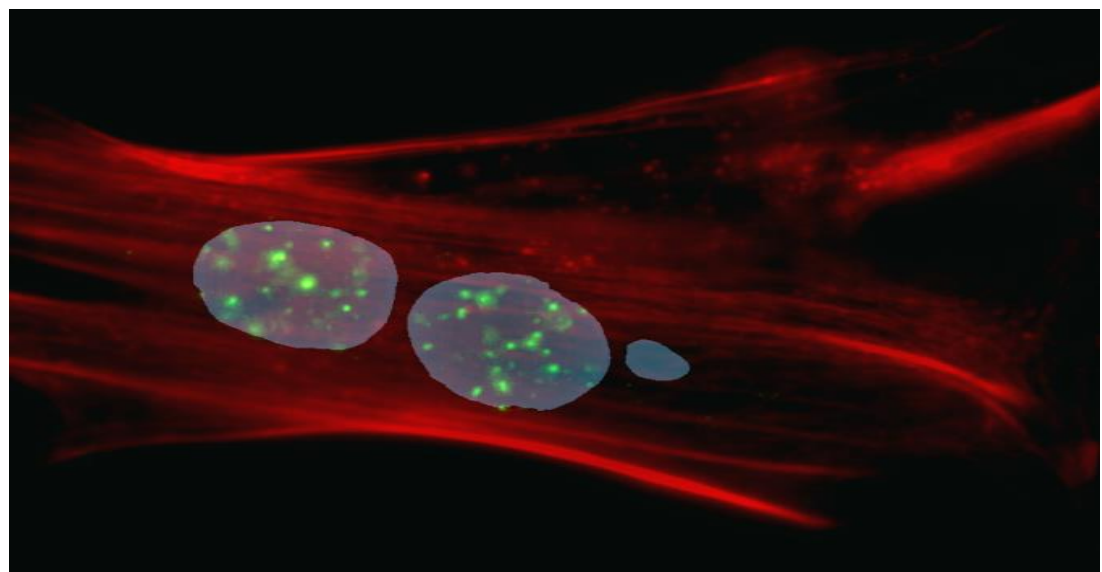
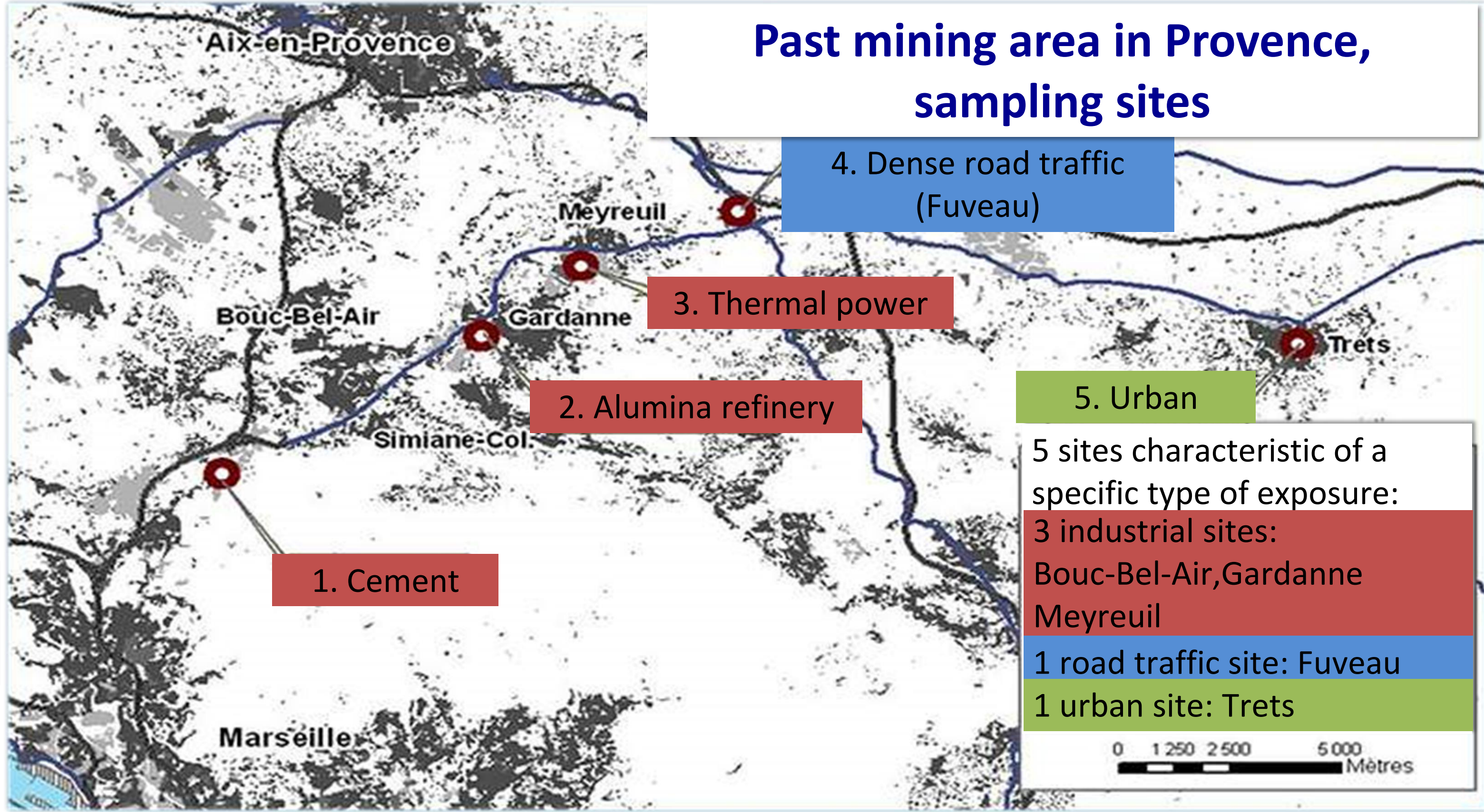
## Conclusion

**Without S9mix:** Increases in C-MN were noted in Meyreuil in summer and winter, and in Trets in winter only. Augmentations in C+MN were revealed in Bouc-Bel-Air in both seasons, in Meyreuil in winter, and in Trets in summer.

**With S9mix:** Increases in C-MN were shown in Fuveau and Trets in both seasons, and in Gardanne and Meyreuil only in winter.

In winter, chromosomal breakages frequently found in presence of metabolic activation may be due to high levels of atmospheric PAHs concentrations. Also, chromosomal breakages and/or losses were observed in the absence of metabolic activation for three sites out of five, regardless of the season. PM<sub>2.5</sub> found in the mining area of Provence, a peri-urban zone which is characterized by heavy traffic and various mining industries, have shown direct and indirect chromosome damaging properties.

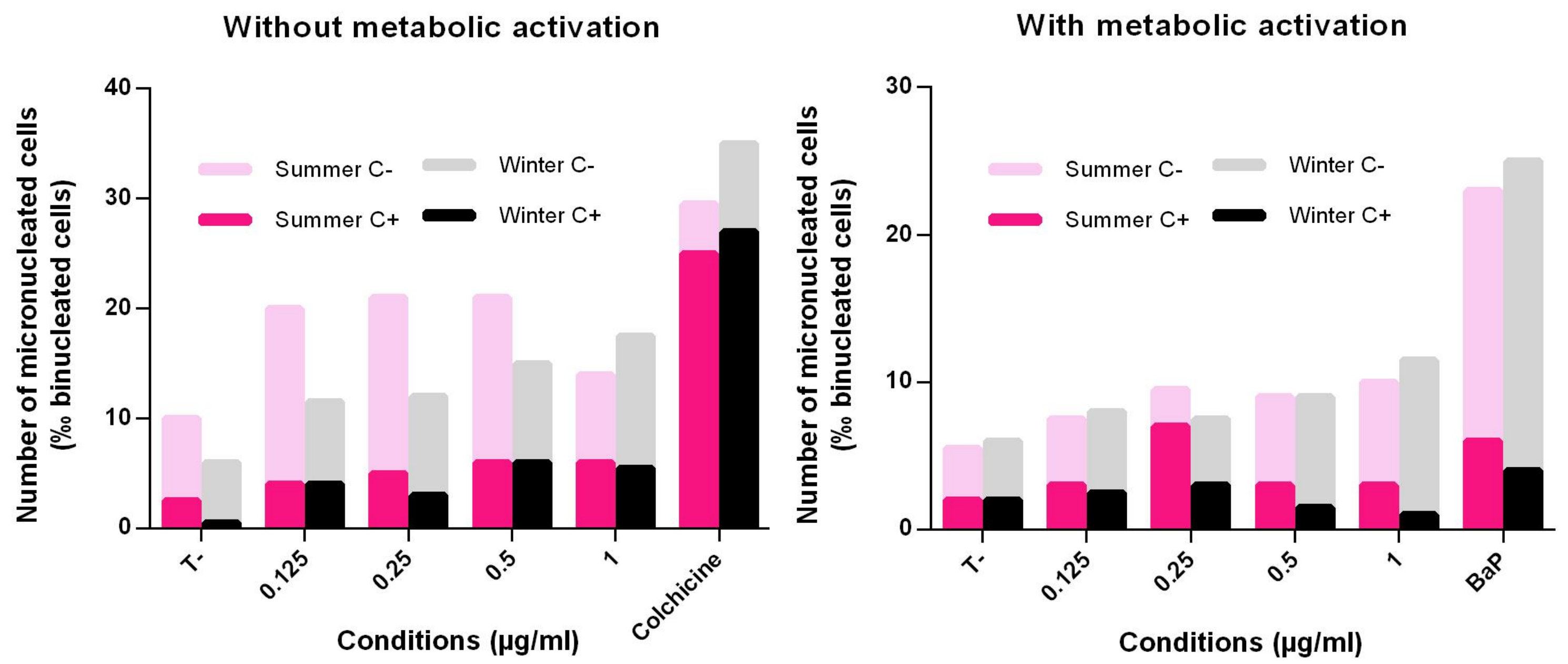
**PM<sub>2.5</sub> genotoxic characteristics and atmospheric levels may be useful in preventing long-term effects related to human exposure.**



Binucleated micronucleated NHLF. DNA is stained using DAPI, cytoplasm with phalloidin-TRITC, and centromere with CREST.

PAHs concentrations detected at the sampling sites vary from [0.099 - 0.492 ng/m<sup>3</sup>].

For each site, higher levels of PAHs were observed in winter, especially for Gardanne, Fuveau and Trets.



E.g.: Micronucleus frequencies observed in Meyreuil site (thermal power plant), in summer and winter, with and without metabolic activation