NANOPARTICLE CARBON BLACK DRIVEN DNA DAMAGE INDUCES GROWTH ARREST AND AP-1 AND NF-κB DNA BINDING IN LUNG EPITHELIAL A549 CELL LINE

R. M. Mroz[1, 2], R. P.F. Schins[3], H. Li[3], E. M Drost[1], W. MacNee[1], and K. Donaldson[1]
[1]ELEGI/Colt Laboratories, MRC/University of Edinburgh Centre for Inflammation Research, Edinburgh, United Kingdom; [2]Department of Pneumology, Bialystok Medical University, Bialystok, Poland; [3]Institut für Umweltmedizinische Forschung (IUF), Heinrich-Heine-University, Düsseldorf, Germany; robmroz@wp.pl

Background: Reactive oxygen species (ROS) have been implicated in various pulmonary diseases by causing direct injury to lung epithelial cells. Signaling activity of cells through transcription factors such as nuclear factor kappa B (NF-□B) and AP-1 may be correlated with demonstrated ability of PM_{10} and their nanoparticles (NP) to cause oxidative stress-driven DNA damage. To assess whether NP-driven DNA damage induces the expression of proinflammatory transcription factors such as NF-κB and AP-1 A549, lung epithelial cells were treated with Carbon Black (CB), nanoparticulate CB (NPCB), NPCB coated with BaP (NPBP) for various times ranging from 30 min to 24 h.
Material and methods: DNA strand break was determined by the comet assay, and the cell cycle status was analyzed using flow cytometry. Nuclear extracts were used for Western blot analysis of PSer15-p53. Electromobility shift assay was used to detect AP-1 and NF-κB DNA binding.
Results: Tested NP caused single strand breaks and significantly altered cell cycle kinetics. NF-κB and AP-1 DNA binding were increased at early time points (2.3- and 2.6-fold at 1 h, respectively). The effects were also found on Ser15-p53 phosphorylation. *N*-acetylcysteine blocked NP-driven NF-κB and AP-1 DNA binding as well as the P~Ser15-p53 response.
Conclusions: NPCB and NPCB induce DNA damage, activating p53, proteins related to DNA repair and proinflammatory transcription factors. Long-term exposure to NPCB and BaP may lead to chronic inflammation of lung epithelium underlying diseases such as COPD and asthma.