A method for assessment of the genotoxicity of mainstream cigarette-smoke by use of the bacterial reverse-mutation assay and an aerosol-based exposure system

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Abstract

To date there are no widely accepted methods for the toxicological testing of complex gaseous mixtures and aerosols, such as cigarette smoke, although some modifications to the standard regulatory methods have been developed and used. Historically, routine testing of cigarettes has primarily focused on the particulate fraction of cigarette smoke. However, this fraction may not accurately reflect the full toxicity and mutagenicity of the smoke aerosol as a whole, which contains semi-
volatiles and short-lived products of combustion. In this study we have used a modified version of the bacterial reverse-mutation (Ames) assay for the testing of mainstream smoke generated from 3R4F reference cigarettes with a Vitrocell® VC 10 exposure system. This method has been evaluated in four strains of Salmonella typhimurium (TA98, TA100, YG1024 and YG1042) and one strain of Escherichia coli (WP2 uvrA pKM101) in the absence and presence of a metabolic activation system. Following exposure at four concentrations of diluted mainstream cigarette-smoke, concentration-related and reproducible increases in the number of revertants were observed in all four Salmonella strains. E. coli strain WP2 uvrA pKM101 was unresponsive at the four concentrations tested. To quantify the exposure dose and to enable biological response to be plotted as a function of deposited mass, quartz-crystal microbalances were included in situ in the smoke-exposure set-up. This methodology was further assessed by comparing the responses of strain YG1042 to mainstream cigarette-smoke on a second VC 10 Smoking Robot. In summary, the Ames assay can be successfully modified to assess the toxicological impact of mainstream cigarette-smoke.