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Comet Assay on Toxicogenetics; Several Studies in Recent Years on Several Genotoxicological Agents

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Abstract

Cancer is one of the main causes of death in the world, and a major issue for human health. Prolonged exposure to a number of chemicals was observed to be one of the primary causes of cancer [1]. The monitoring of the surrounding environment for chemicals and compounds with possible genotoxic activity is of high priority [2]. Thus, the development of instruments for identifying risky chemicals and the understanding of their toxicity mecanism is a major objective for scientific research [3]. A number of assays exist for detection of genotoxicity in a variety of experimental systems, some of them with limited use due to complicated technical setup, the single cell gel electrophoresis assay also defines the Comet assay [3], discovered for the first time in 1984 by two Swedish researchers, Ostling and Johanson [4]. In 1988 Singh et al., introduced the concept of alkaline version [2,4]. It allows investigation of DNA damage in virtually all cell types without the necessity of cell cultures [2]. It is widely used to detect DNA damage [5] as an indicator of exposure to genotoxicogical agents [2,6,7]. The Comet assay is a used method in human, environmental, and ecogenotoxicological studies [2] and it is performed to detect genotoxicity effect of biocides, chemicals prodcuts, agrochemicals, pharmaceuticals and food additives in genotoxicity assaying [8]

The comet assay is widely used to assess DNA damage. Before lysis and leaving nucleoids, cell should embedding in agarose than fixed on microscope slide. After an alkaline electrophoresis, DNA loops containing breaks are relaxed and extend towards the anode, forming a comet-like image viewed by fluorescence microscopy with a suitable stain [9,10]. The different steps of Comet assay are Preparation of microscope slides: The aim of microscope slide preparation is to ensure the uniformity of the gel, assure the stability and the survival for the collection of data, minimise background noise as well as to ensure well visualized of comets [11]. Release of DNA from lysed cells Apply on the slides, a lysis solution that contain Triton X-100 and a high concentration of salt with 2.5 M NaCl [4,12]. Lysis allow the removing membranes, releases the soluble components of the cell, strips histones from DNA, and sheets of compact structures that are nucleoids wherein the DNA is attached at intervals to the nuclear matrix [13].

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