

Optimizing Premature Chromosome Condensation (PCC) of Human Lymphocytes by Somatic Cell Hybridization to Study Primary DNA Damages

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ABSTRACT Cytogenetic tools are important in understanding the health effects of various chemical and physical agents implicated in human health. Several biodosimetric and cytogenetic techniques help in the assessment of the genotoxic effects of therapeutic drugs and these become essential for human health care management. There exist methods by which genotoxicity can be identified and quantified employing interphase condensed DNA. Premature Chromosome Condensation (PCC) can be achieved by Somatic Cell Fusions and can result in obtaining condensed DNA into “near chromosome like clarity” and the same can be utilized to check for common end-points such as DNA “breaks” and “gaps”, as can be induced by genotoxic drugs. PCC in conjunction with FISH can also be an added advantage for the same. We present here an efficient method to obtain high quality PCC spreads of human lymphocytes, which can be analyzed with relative simplicity for genotoxic monitoring. Mitotic CHO cells were used as the fusion partners followed by incubation and the processing of cells for analysis. Image capturing and analysis was done after fixing and casting cells. The key factors such as the ratio of the fusion partners, fusion and processing protocols are optimized and presented. Although PCC cannot replace conventional lymphocyte culture techniques completely, it can be efficiently utilized for genotoxic monitoring with advantages such as the considerable reduction in time required for processing and analysis of samples. This will also eliminate the chances of genotoxicity underestimation due to the inherent DNA damage repair capacity of human lymphocytes.