

Absence of micronucleus formation in CHO-K1 cells cultivated in platelet lysate enriched medium

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Abstract

Human platelet lysate (PL) represents an effective substitute of fetal bovine serum (FBS) for mesenchymal stromal cell (MSC) cultivation. Compared to FBS, PL favors MSC proliferation significantly shortening the population doubling time and avoiding the risks related to the use of animal derivatives. Growth factors contained in the platelets are released upon platelet disruption following freezing/thawing cycles or as we have recently described by using ultrasound. We have investigated whether the increased cell proliferation achieved by using PL could induce mitotic stress and whether the potential formation of free radicals during PL production by ultrasound could cause chromosomal instability in mammalian cells. We have applied an image analysis assisted high content screening (HCS) in vitro micronucleus assay in the Chinese Hamster Ovarian K1 (CHO-K1) rodent mammalian cell line. PL was produced by sonication; for the micronucleus assay, CHO-K1 cells were exposed to increasing concentrations of PL. Cytokinesis was blocked by cytochalasin B, nuclei were stained with bisbenzimidazole and images were acquired and analyzed automatically using an HCS system, both with a 20× and a 10× objective. Our results suggest that growth stimulus induced by the use of PL did not significantly increase micronucleus formation in CHO-K1 cells compared to negative control. Micronucleus testing in conjunction with HCS could represent a valid tool to evaluate the safety of ancillary materials used in the production of cell-based medicinal products.