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THE COMBINATION OF MICRONUCLEUS ASSAY AND FISH TECHNIQUE FOR THE EVALUATION OF GENOTOXICITY OF 1, 2, 4-BENZENETRIOL

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The cytokinesis-block micronucleus assay have been emerged as one of the preferred method for assessing chromosome damage. Micronucleus are small, extranuclear bodies that are formed in mitosis from acentric chromosomal fragments or chromosome that are not included in each daughter nuclei. Thus, micronucleus contain either chromosomal fragments or whole chromosomes. The cytokinesis-block micronucleus assay together with FISH technique using specific centromere probes for chromosome 7 and 8 were employed in mitogen-stimulated human lymphocyte pretreated with Benzen metabolite, 1, 2, 4-benzenetriol.

Treatment of human lymphocyte resulted in the induction of micronucleus in a dose-dependent manner. The frequency of MN in untreated control lymphocyte was 4.5 per 1000 binucleated cells and it increased to 8, 9.5, 14, 26.5 and 35.5 per 1000 binuclated cells at concentration of 10 μM, 25 μM, 50 μM and 100 μM respectively. Frequencies of aneusomy 7 and 8 in binucleated cell also increased with dose. Aneusomy 8 was more frequent than aneusomy 7, suggesting that chromosome 8 is more sensitive for aneusomy induction by BT. The frequency of MN with centromere positive signals for chromosome 7 and 8 increased with dose of BT. The frequency of MN with centromere positive signals was higher for chromosome 8 than for chromosome 7, also suggesting the greater sensitivity of chromosome 8.

The results suggested that combined application of CBMN assay with FISH technique using chromosome specific centromere probe allows to detect aneuploid in human lymphocyte and to identify the mechanistic origin of micronucleus induced by clastogen or aneugen.