P108 Centromere staining by telomere FISH in *in vitro* micronucleus assay using CHL/IU cells

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The *in vitro* micronucleus (MN) assay in conjunction with centromere staining is utilized for discrimination between clastogenic and aneugenic effects in human cells. However, appropriate probes or antibodies for this purpose are hardly known for CHL/IU cells derived from Chinese hamster. In this study, a FISH staining method for telomeric repeats, which were reported to be present in pericentromeric regions of most of chromosomes of Chinese hamster, was evaluated to detect centromere in *in vitro* MN assay using CHL/IU cells.

Cells treated with each 5 clastogen and 7 aneugen were labeled with the FITC-conjugated peptide nucleic acid probe for telomeric sequence (DAKO) by FISH. The frequency of MN was scored by counting the number of cells with MN per 1000 interphase cells, and the frequency of MN with FISH signals (CEN+MN) were examined.

All compounds increased the frequency of MN. All aneugens, but not clastogens, significantly increased the frequency of CEN+MN (>60%) in comparison with the vehicle control (43.4%). The results suggest that centromere staining by telomere FISH is useful to discriminate clastogenic and aneugenic effects in *in vitro* micronucleus test using CHL/IU cells.

CHL/IU細胞を用いたin vitro小核試験における動原体染色:telomere FISH法 有質千浪,山崎秀子,倉部道恵,藤村久子,鳥海亙:田辺三菱製薬株式会社安全性研究所

P109 The comparison of the positive control in the chromosome aberration test in the CHL/IU cell

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Historical data on the negative control and the positive control are necessary as a substance which the matter whether that examination could be done precisely in the top to do a chromosome aberration test is judged. We report it here because we compared it about 3 positive control substance to set up the positive control of the metabolic activation method newly this time. It was cultivated by using the CHL/IU cell with a CS minimum essential medium culture fluid 10%. S9 used the thing of Oriental Yeast. N-nitrosodimethylamine (DMN), Cyclophosphamide (CPA) and Benzo(a) pyrene (B(a)P) were used as the positive control substance. DMN in the positive control was made with normal saline solution before use, and used for the experiment. It froze in -80°C, and dissolved at the time of the use, and used for the experiment after B(a)P dissolved in DMSO after CPA dissolved in the normal saline solution. At present, change data on every test are being collected.

CHL/IU細胞の染色体異常試験における陽性対照の比較

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