Chromatin Folding, Fragile Sites, and Chromosome Aberrations Induced by Low- and High-LET Radiation

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Abstract

We previously demonstrated non-random distributions of breaks involved in chromosome aberrations induced by low- and high-LET radiation. To investigate the factors contributing to the break point distribution in radiation-induced chromosome aberrations, human epithelial cells were fixed in G1 phase. Interphase chromosomes were hybridized with a multicolor banding (mBAND) probe for chromosome 3 which distinguishes six regions of the chromosome in separate colors. After the images were captured with a laser scanning confocal microscope, the three-dimensional structure of interphase chromosome 3 was reconstructed at multi-mega base pair scale. Specific locations of the chromosome, in interphase, were also analyzed with bacterial artificial chromosome (BAC) probes.

Both mBAND and BAC studies revealed non-random folding of chromatin in interphase, and suggested association of interphase chromatin folding to the radiation-induced chromosome aberration hotspots. We further investigated the distribution of genic content, as well as the distribution of breaks found in tumor cells. Comparisons of these distributions to the radiation hotspots showed that some of the radiation hotspots coincide with the frequent breaks found in solid tumors and with the fragile sites for other environmental toxins. Our results suggest that multiple factors, including the chromatin structure and the gene distribution, can contribute to radiation-induced chromosome aberrations.

Materials and Methods

Cells
Human epithelial cells (CH184B5F5/M10) were cultured in chamber slides. Upon confluency, the cells in were fixed with methanol/acetic acid.

Interphase chromosome painting with mBAND probes
The interphase cells were hybridized using the XCyto3 mBAND kit from MetaSystems. The 3-D images of chromosomes 3 were captured with a laser scanning confocal microscope, and the three dimensional structure of interphase chromosome 3 with six colored regions was reconstructed using 3D analysis software Imaris®. The distances between different regions were measured as well.

Results

The Distance between Different Regions of Chromosome 3 and the Center of the Chromosome 3D Domain

The center of Region B is closest to the center of the domain of chromosome 3, Region A (telomere on the p-arm), region C (close to centromere), and region E (telomere on the q-arm) are farther away from the chromosome domain center than either Region B or region E.

Distribution of Angles Extended to the Neighboring Regions

Regions A, B, and C appeared to be folded more compactly with sharp turns compared to region D, and F, which wrapping around at the peripheral area of the chromosome domain. It has been confirmed by analyzing the locations of four BAC probes labeled 100K fragments, showing that the sequence is not random positioned, but wrapping around the chromosome peripherally.

Conclusions

- The regions towards the telomeres are likely to occupy the peripheral area of the chromosome domain. These heterochromatin regions located in the peripheral of the chromosome domain may provide the structural support, define the chromosome territory, and maintain the chromosome integrity.
- The present results showed that Band 6-8 tends to locate near the interior of the chromosome domain, and are localized closely to Band 12-13 and Band 19-22. This finding is consistent with the frequency of intra-chromosome exchanges between breaks in these regions.
- The non-random breakpoint distribution in chromosome 3 after radiation exposure may be associated with the folding of chromatin in interphase.
- Other factors, including the location of the fragile sites and transcription activities, may also contribute to the distribution of radiation-induced inter- and intra-chromatome exchange hotspots.
- The distribution of breaks participated in inter- and intra-chromosome exchanges found in chromosome 3 of solid tumors is in partial agreement with radiation induced chromosome exchange hotspots.

Introduction

Location of breaks participated in inter- and intrachromosome exchanges after gamma exposure

Distribution of Distance between Regions 8, 12, and 20, which Involve in Intra-Chromosome Ablations

The distance between regions 12 and 20 was much longer than the distance from B to 12, and from B to 20, indicating that region 12 and 20 may exhibit a lower frequency of intra-chromosome exchanges with each other.

Chromosome 3 and Cancer

Chromosome 3 contains multiple oncogenes and tumor suppressor genes. Chromosome aberrations in the p-arm are known to be associated with various cancer types.