Interphase Chromosome Conformation and Chromatin-chromatin Interactions in Human Epithelial Cells Cultured under Different Gravity Conditions

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Chromatin Folding Models

Random walk/polymer model of chromatin folding on 0.5-5 mbp scales

Non-randomness of chromatin folding on multi-megabase scales

- Being confined in localized chromosome domains
- Limiting the spatial extension of the chromatin
- With regions of different transcriptional activities and gene densities
Distributions of low- and high-LET radiation-induced breaks in chromosome 3 are associated with inter- and intra- chromosomal exchanges in human mammary epithelial cells

Hada et al, Radiation Research, 2011

Chromosome 3 is hybridized with mBAND probes in 5 fluorophore dyes. The chromosome is separated in six colored regions and 5 junction regions.
Chromosome 3

Chromosome 3 spans almost 200 million base pairs, representing about 6.5 percent of the total DNA in cells, and containing about 1,980 genes (Human genome overview page, Build 36.3 http://www.ncbi.nlm.nih.gov/mapview/maps.cgi?taxid=9606&build=previous&chr=3)

<table>
<thead>
<tr>
<th>Topography</th>
<th>Case</th>
<th>Case with Ch 3 Aberration</th>
<th>%</th>
<th>Case Analyzed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bone and Soft Tissues</td>
<td>3406</td>
<td>576</td>
<td>16.9</td>
<td>500</td>
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<tr>
<td>Breast</td>
<td>1128</td>
<td>293</td>
<td>25.9</td>
<td>245</td>
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<tr>
<td>Cardiovascular</td>
<td>48</td>
<td>2</td>
<td>4.2</td>
<td>1</td>
</tr>
<tr>
<td>CNS</td>
<td>2511</td>
<td>208</td>
<td>8.3</td>
<td>175</td>
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<td>Digestive System</td>
<td>2285</td>
<td>543</td>
<td>23.8</td>
<td>492</td>
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<tr>
<td>Endocrine System</td>
<td>605</td>
<td>115</td>
<td>24.8</td>
<td>107</td>
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<td>Female Genital Organs</td>
<td>1552</td>
<td>266</td>
<td>17.1</td>
<td>225</td>
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<tr>
<td>Male Genital Organs</td>
<td>609</td>
<td>94</td>
<td>15.4</td>
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<td>Respiratory System</td>
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<td>365</td>
<td>32.4</td>
<td>319</td>
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<tr>
<td>Skin</td>
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<td>76</td>
<td>22.9</td>
<td>71</td>
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<td>Urinary Tract</td>
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<td>640</td>
<td>29.5</td>
<td>569</td>
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<td><strong>Overall</strong></td>
<td><strong>15769</strong></td>
<td><strong>3178</strong></td>
<td><strong>20.2</strong></td>
<td><strong>2767</strong></td>
</tr>
</tbody>
</table>

Yunis and Hoffman 1989

The hotspots for radiation exposure coincide with some but not all hotspots for solid cancers. The region of 3p21.3 (band 6 within region 2) is a known fragile site and contain several tumor suppression genes/regions.

Distributions of breaks involved in intra-chromosome 3 exchanges in cancer patients
γ-rays of low dose rate (Panel A), neutrons (Panel B), γ-rays of high dose rate (Panel C) or Fe ions (Panel D). Panel E: Most of the intra-chromosomal exchanges occurred between a break in band 5-9 (region 2) and one in band 11-13 (junction 3/4).  

Hada et al. Rad Res., 2011
Distributions of radiation-induced intra‐chromosomal exchanges in human lymphocytes
Proximity within interphase chromosome contributes to the breakpoint distribution in radiation-induced intra-chromosomal exchanges

Zhang et al, LSSR, 2014
Distances between the center of each region and the center of the chromosome domain
Integral distribution of the angles extended from the center of each colored region to the neighboring colored regions
Distances between regions 2 or 4 and other regions

Epithelial cells

Lymphocytes
In Simulated Microgravity:

- Transcriptional changes

Distribution of genes with significantly altered expression on chromosome 3 in lymphoblastoid cells

Mangala et al, 2011

- Chromatin folding

- Adaptation
Distance between the neighboring regions

Distance (Micron)

Measurement Endpoints (in order on Chromosome 3)

1 to 1/2, 1/2 to 2, 2 to 2/3, 2/3 to 3, 3 to 3/4, 3/4 to 4, 4 to 4/5, 4/5 to 5, 5 to 5/6, 5/6 to 6

Static epithelial cells

RPM_Epithelial

Lymphocyte

Distance (Micron)

1 to 2, 2 to 3, 3 to 4, 4 to 5, 5 to 6
Distances between the center of each region and the center of the chromosome domain

![Graph showing distances for RPM_Epithelial, Stat_Epithelial, and Lymphocyte regions.]

- RPM_Epithelial
- Stat_Epithelial
- Lymphocyte
Distances between Region 2 and other regions
Questions to be investigated:

- Changes of distribution of radiation induced chromosome aberration in simulated microgravity culture condition
- Gene-gene interaction and transcriptional regulation
- Other chromosomes
- Normal vs Cancerous cells

In Space: *unknown*
Conclusion

• On a multi-mega base pair scale of the DNA, the arrangement of chromatin is non-random.

• In M10 epithelial cells, both telomere regions tend to be located towards the exterior of the chromosome domain, whereas the rest p-arm of the chromatin region towards the interior. In contrast, most of the q-arm of the chromatin is found in the peripheral of the domain.

• In lymphocytes, the p-arm chromatin regions towards the interior in close proximity with each other, whereas two q-arm regions are nearness in space. It indicates that G0 lymphocytes may lack secondary 3D chromatin folding.

• There chromatin folding patterns are consistent with our previous finding of non-random distribution of intra-chromosomal exchanges.

• In simulated microgravity conditions, the chromosome conformation may be altered and new regions in close proximity, especially to region 2 are suggested.
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