ESTIMATING RADIAL POSITION AND CHROMATIN COMPACTION FROM GAM DATA

This activity will use the data reported in the following article:

Beagrie RA, Scialdone A, Schueler M, Kraemer DC, Chotalia M, Xie SQ, Barbieri

M, de Santiago I, Lavitas LM, Branco MR, Fraser J, Dostie J, Game L, Dillon N, Edwards PA, Nicodemi M, Pombo A.

Complex multi-enhancer contacts captured by genome architecture mapping.

*Nature*. 2017 Mar 23;**543(7646)**:519-524.

<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5366070/#!po=5.27638>

The data may be downloaded here:

<https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE64881>

scroll to the bottom of the page and download the following file:

GSE64881\_segmentation\_at\_30000bp.passqc.multibam.txt.gz

Evaluate data quality:

Identify windows that have an unusually high detection frequency (the detection frequency of a genomic window is the number of NPs that detect the window). One way to do this is to sort the windows by detection frequency and then to look for outliers at the top of the sorted list. Alternately, this can be done by generating a scatter plot and visually determining what threshold constitutes an unusually high detection frequency.

Estimate radial position of each NP:

The GAM paper describes how to estimate radial position of an NP (see below for the relevant excerpt from the paper). Use these insights to estimate radial position for each NP on a scale of 1-5:

1 – strongly apical

2 – somewhat apical

3 – neither apical nor equatorial

4 – somewhat equatorial

5 – strongly apical

### **Estimation of chromosome radial position from GAM data**

Thanks to the random orientation of sectioning with respect to the nucleus, the DNA content of NPs originated from different latitudes of the nucleus can be used to estimate radial distributions of genomic regions. For example, NPs cut through nuclei close to their periphery contain, by definition, a smaller proportion of the nuclear volume (or DNA content) than equatorial NPs ([Extended Data Fig. 9a](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5366070/figure/F14/?report=objectonly)). Therefore, we predicted that the percentage of the genome covered by each NP could be used as a proxy for its latitude relative to the most equatorial NPs.

Estimate compaction of each genomic window:

The GAM paper describes how to estimate the degree of compaction of a genomic window (see below for the relevant excerpt from the paper). Use these insights to assign each window a compaction rating between 1-10 (10 is most condensed; 1 is least condensed).

**Estimation of locus volume from GAM data**

We reasoned that de-condensed genomic loci should occupy larger volumes (or adopt more elongated conformations) than more condensed loci. De-condensed loci would therefore be intersected more frequently (and be detected more frequently in randomly-oriented nuclear profiles) than smaller or more spherical loci ([Extended Data Fig. 9c](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5366070/figure/F14/?report=objectonly)).