3D genomic architecture and transcriptional regulation

Understanding the conformations at single cell and single gene level

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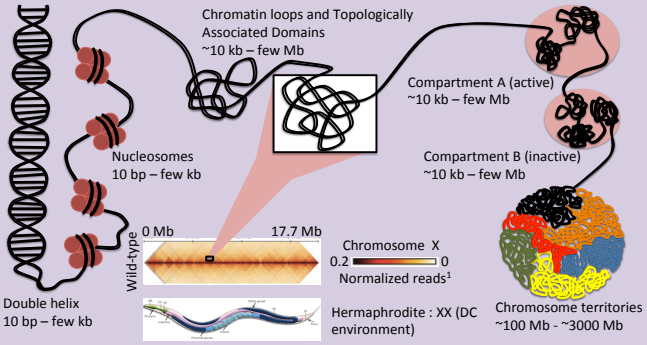
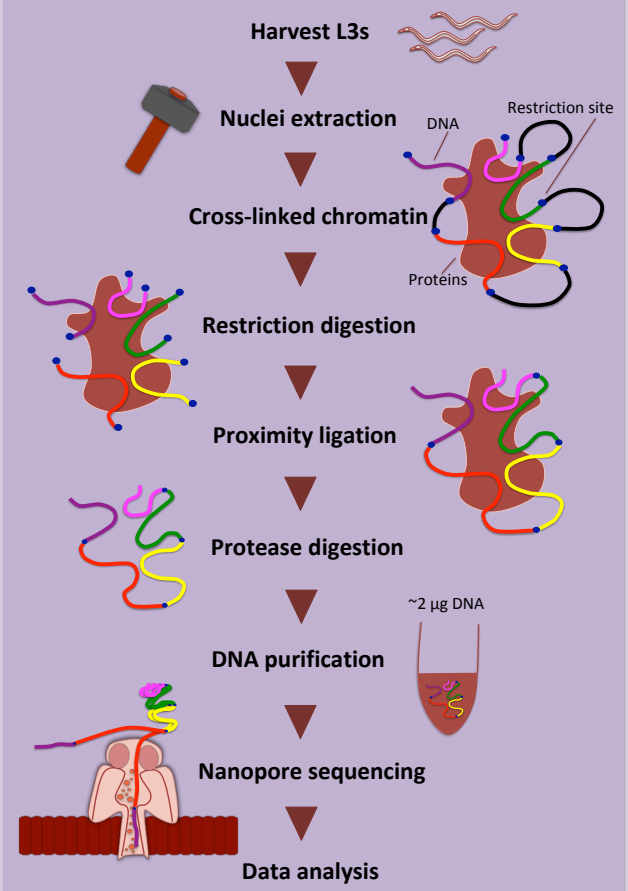
INTRODUCTION

- Dosage Compensation (DC) in *Caenorhabditis elegans*, is achieved by half-repression of X-linked genes on both X chromosomes in the hermaphrodites (XX) as compared to the males (X). This reduction of gene expression is associated with the formation of enhanced Topologically Associated Domains (eTADs)[1]. TADs are defined as highly self-contacting portions of the genome, delimited by boundaries enriched with structural maintenance complexes (SMC) [2-4].
- TADs show variability in internal contact frequency[5]; some TADs display high contact frequencies while in others these contacts are sparse. The level of TAD internal contacts can be used as a measure of chromatin compaction, which in turn can be correlated with changes in gene transcription.
- eTADs in *C. elegans* result from the combined action of the Dosage Compensation Complex (DCC), an SMC complex, and highly oriented sequence motifs called *rex* sites. However, the question remains on how loading of the DCC induces changes of the chromosome structure and the correlation with gene expression regulation. **Therefore, the formation of the eTAD exclusively on the X chromosome in the hermaphrodites and its dependence on DC provides the perfect system to decipher if and how these domains regulate transcription.**



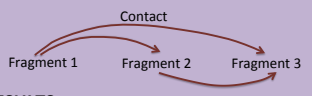
How to capture single gene conformation in single cell to understand gene expression regulation?

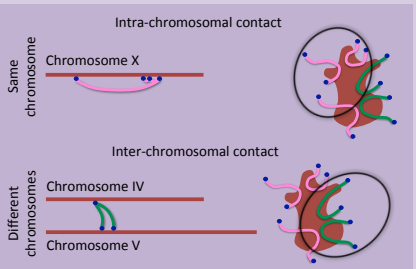
Nano-HiC: HiC coupled with nanopore to capture gene conformations in single cell



DATA PROCESSING

	Dataset A	Dataset B
Data generated	1.8 GB	4.9 GB
Reads	970,509	2,507,330
Reads > 10 kb	1026	1851
Contacts	6,196,396	11,167,948





RESULTS

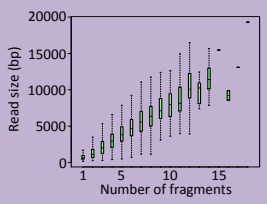


Figure 1: Read size per fragment. There are a significant number of long reads sequenced, comprising multiple fragments.

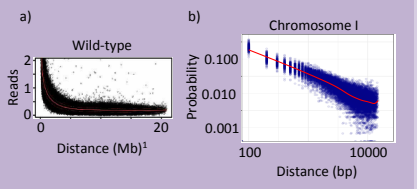


Figure 2: a) Decay plot shows that the interaction frequency between fragments decrease as genomic distance increase[1]. b) Similar observations detected for Nano-HiC datasets, Chromosome I decay plot, bin size: 100kb.

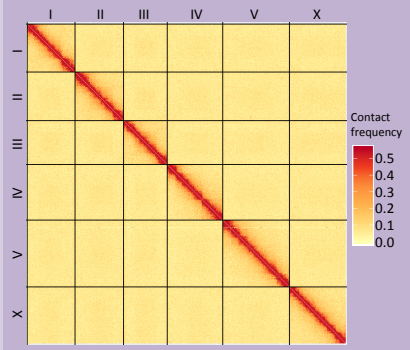


Figure 3: Genome-wide interaction map, normalized reads, bin size: 100kb. Frequency of interaction of the entire hermaphrodite genome.

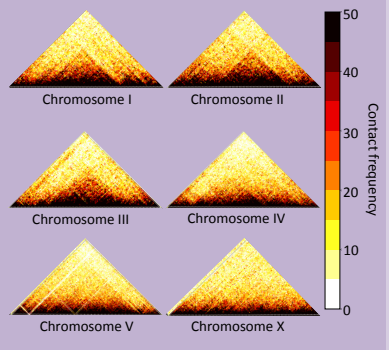


Figure 4: Chromosome interaction map, bin size: 50kb. Frequency of self-interaction map of the individual chromosomes in the hermaphrodite genome, plotted from a dataset of ~6GB. Distinct domains can be observed.

REFERENCES

1. Crane, E. *et al.* Condensin-driven remodelling of X chromosome topology during dosage compensation. *Nature* **523**, (2015)
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3. Nora, E. P. *et al.* Spatial partitioning of the regulatory landscape of the X-inactivation centre. *Nature* **485**, (2012).
4. Dekker, J. & Heard, E. Structural and functional diversity of Topologically Associating Domains. *FEBS Lett* **589**, (2015).
5. Dixon, J. R. *et al.* Chromatin architecture reorganization during stem cell differentiation. *Nature* **518**, (2015).

CONCLUSIONS

- HiC data generated reflects on the chromatin structures in the DC environment (wild-type). Work on the pipeline for data analysis is ongoing, with emphasis on the multi-contact fragments from single reads.
- In order to identify the chromatin structures in a non-DC environment, the HiC technique has to be conducted in transgenic strains, developed in our laboratory, where the DCC is removed from the chromatin. A comparative study should then provide enough high quality data to understand how TADs impact transcriptional regulation.