Bioinformatic approaches to regulatory genomics and epigenomics

376-1347-00L | week 04

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Plan for today

- Debriefing on the assignment
- Coverage track generation
- Manipulating and visualizing peaks
- ENCODE & functional elements
- Finding data from the literature

Debriefing on the assignments

- Handing in the exercises etc.:
  - Handing in the exercises: Please name the exercises files just **`assignment.html`**
- File naming

```
download.file("https://www.encodeproject.org/files/ENCFF127RRR/@@download/ENCFF127RRR.fastq.gz", dest="raw/Myc.fastq.gz", mode = "wb")
```

```
align.stats <- Rsubread::align(index="BDGP6_genome/rsubread", type="dna",
                               readfile1="rfastp.trimmed/Myc_R1.fastq.gz",
                               output_file="aligned/Myc.bam",
                               nthreads=6, sortReadsByCoordinates=TRUE)
```

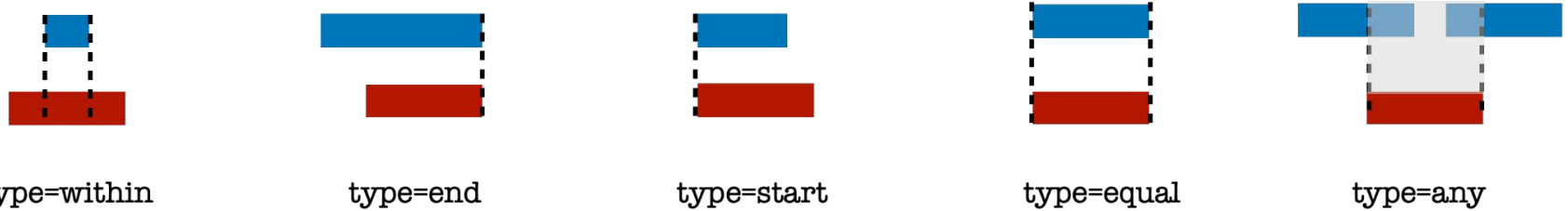
=> however what we downloaded for this exercise is a **CTCF** dataset

Can easily lead to mistakes if you have several files on disk.

Debriefing on the assignments

Finding overlaps

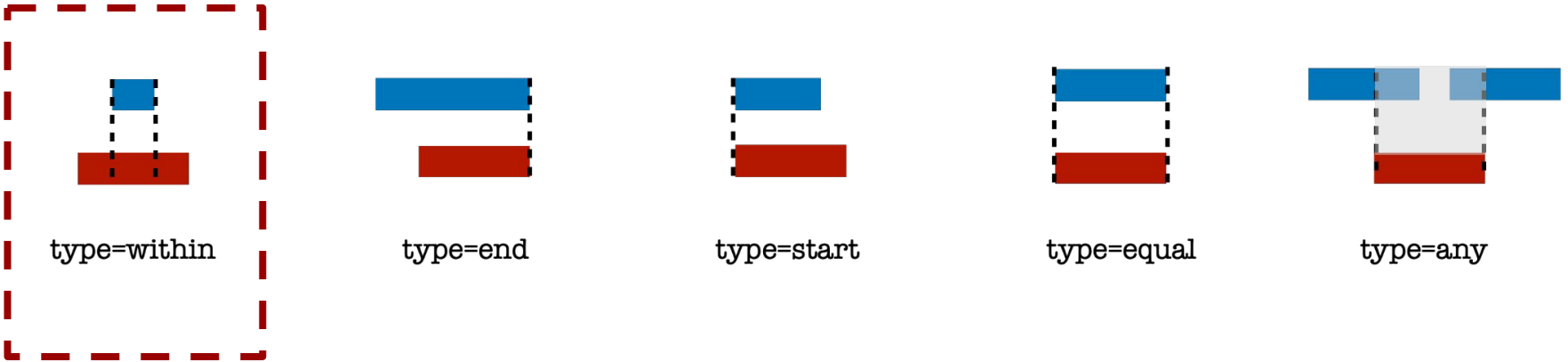
```
findOverlaps(peaks, genes, type="<...>")
```



Debriefing on the assignments

Finding overlaps (e.g for finding a peaks inside a gene)

```
findOverlaps(peaks, genes, type="<...>")
```



Debriefing on the assignments

also note `findOverlaps(a,b)` is not necessarily the same as `findOverlaps(b,a)`.

```
  a chr1     27-40     *
  b chr1     26-40     *
```

```
> findOverlaps(a,b, type="within")
Hits object with 1 hit and 0 metadata columns:
      queryHits subjectHits
      <integer>   <integer>
  [1]         1           1
  -------
  queryLength: 1 / subjectLength: 1
> findOverlaps(b,a, type="within")
Hits object with 0 hits and 0 metadata columns:
   queryHits subjectHits
   <integer>   <integer>
  -------
  queryLength: 1 / subjectLength: 1
```

Debriefing on the assignments

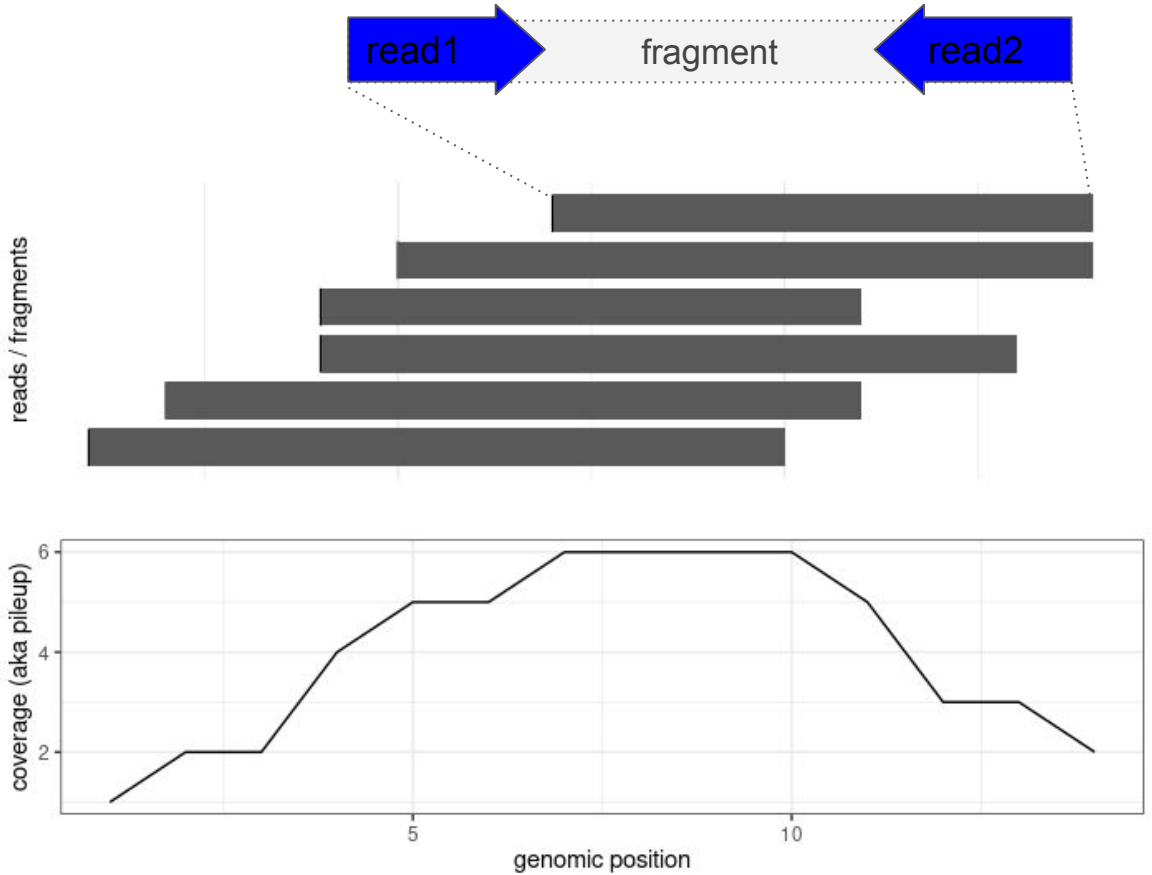
Warning: Each of the 2 combined objects has sequence levels not in the other:

- in 'x': Unmapped_Scaffold_4_D1555_D1692, Unmapped_Scaffold_60_D1601, …

This means that the two objects don’t have exactly the same chromosomes (i.e. “seqLevels”). This can be because:

- You are using objects (e.g. an EnsDb and a genome) that don’t match, or
- Your genome contains un-assembled scaffolds which are absent from the other object (e.g. gene annotation)

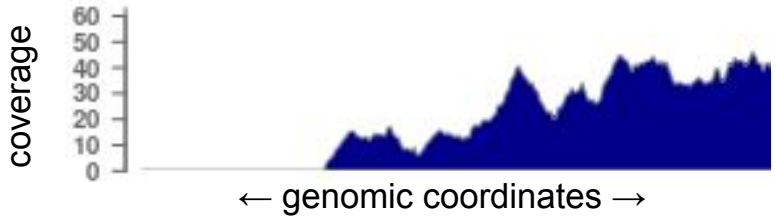
Recap of fragment summarization



Visualizations available in *epiwraps*

[Documentation]

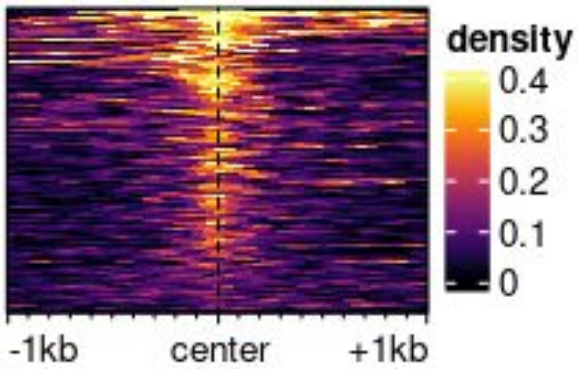
- Signal across one genomic region: `plotSignalTracks`



- `Input: bam/bigwig/bed/GRanges`

(Based on the *Gviz* R package)

- Signal across several genomic regions: `signal2Matrix` → `plotEnrichedHeatmaps`

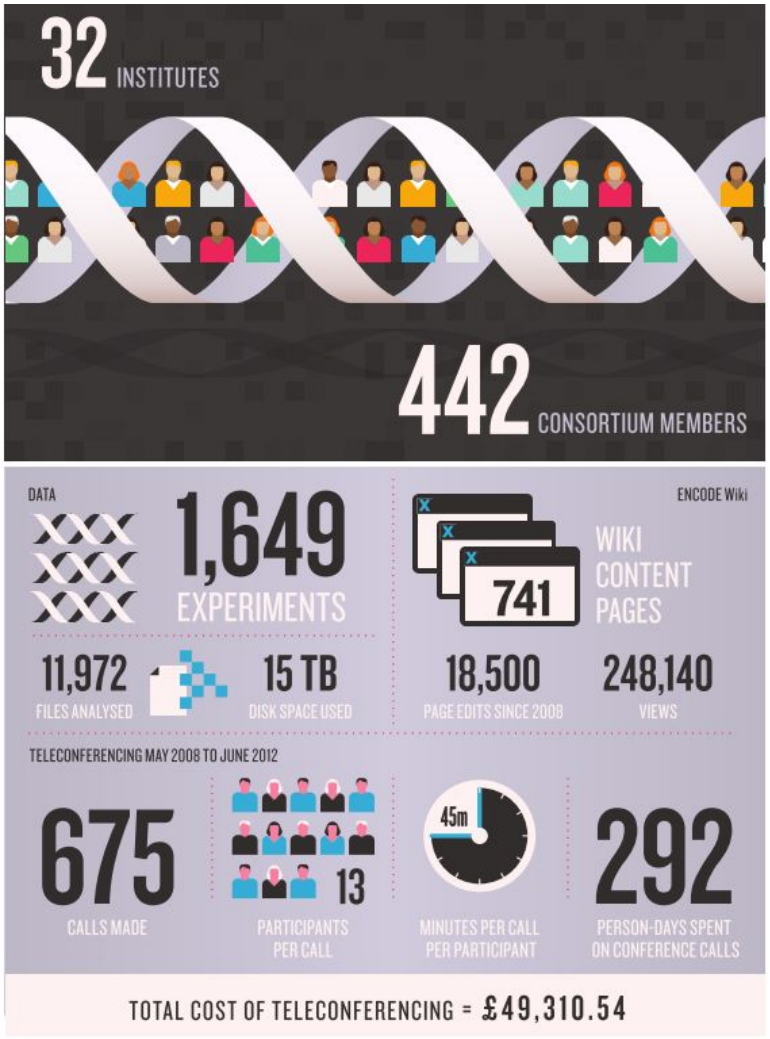


(Mainly based on the EnrichedHeatmap R package, itself based on ComplexHeatmap)

Extension of single-end reads in coverage track generation

Coverage without read extension

Coverage with read extension



The ENCyclopedia Of DNA Elements

~30 publications in September 2012

$288 million USD

… then an ENCODE2, 3, now working towards the 5…

An integrated encyclopedia of DNA elements in the human genome

The ENCODE Project Consortium

Nature **489**, 57–74 (2012) | Cite this article

Bits of Mystery DNA, Far From 'Junk,' Play Crucial Role

The New York Times

by Gina Kolata

"At least 80 percent of this DNA is *active* and *needed*."

The evolutionary arguments for junk:

- 1% protein-coding
- ~4 to 10% evolutionarily conserved
- >50% transposable elements
- Onions have a 5 times bigger genome

The very angry response:

- Graur et al., GBE 2013

NEWS&ANALYSIS

GENOMICS

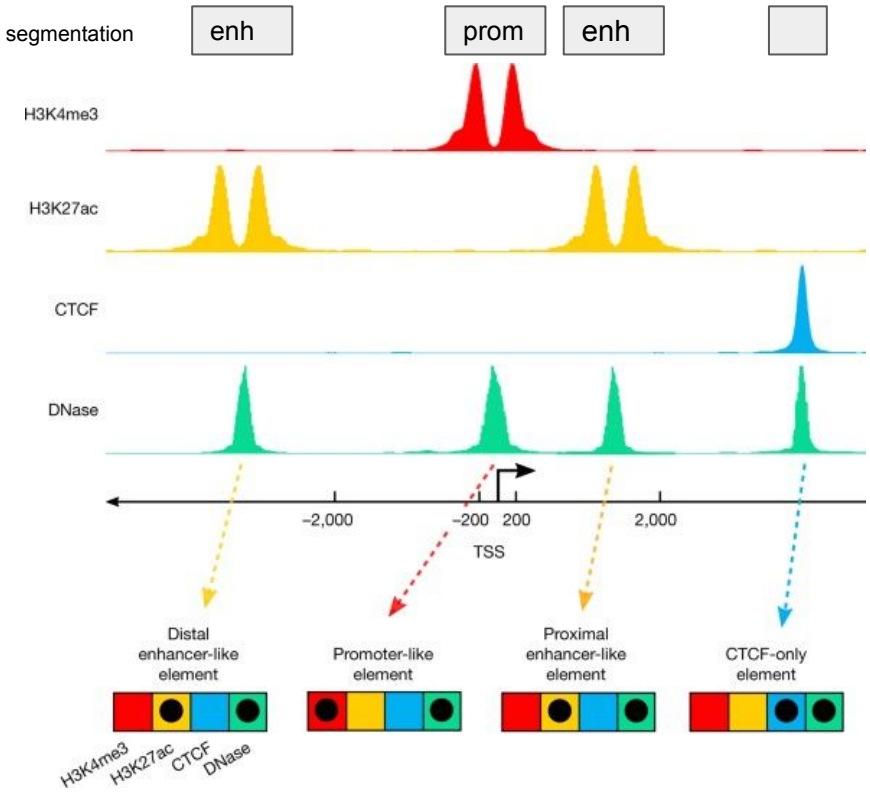
ENCODE Project Writes Eulogy For Junk DNA

–ELIZABETH PENNISI

SCIENCE VOL 337 7 SEPTEMBER 2012

(For more on the topic, see Germain, Ratti and Boem 2014; Ratti and Germain 2022)

A signature-based encyclopedia of DNA elements



ENCODE's "signature strategy":

- Different types of functional genetic elements are associated with different chemical signatures
- We can identify functional elements by identifying these signatures genome-wide

Generic repositories for NGS data

https://www.ebi.ac.uk/biostudies/arrayexpress

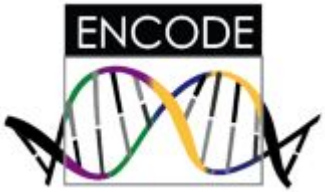
https://www.ncbi.nlm.nih.gov/geo/

https://www.ebi.ac.uk/ena/

https://www.ncbi.nlm.nih.gov/sra

International Nucleotide Sequence Database Collaboration

Quality-controlled and uniformly processed human and mouse NGS datasets



www.roadmapepigenomics.org

www.encodeproject.org

(hematopoietic system)

Assignment

- Find and download from ENCODE the **peaks** (i.e. bed-like format) for the following histone modifications in mouse embryonic stem cells (mESC) from ENCODE:
  - p300, H3K4me3, H3K4me1, H3K27ac, and H3K27me3
  - (when there are replicates, we recommend using the bed file denoted as "`conservative IDR thresholded peaks`")
- Of the p300 peaks, what proportion overlap each of the marks?
- Don't forget to upload your assignment as "`assignment.html`" !