Computing with Sequences and Ranges

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Sequences: representation

DNAStringSet: Vector of sequences, e.g., sequence of each exon in the UCSC knownGene track

```
  A DNAStringSet instance of length 289969
          width seq
      [1]   354 CTTGCCGTCAGCCTTT...TCACAACCTAGGCCA
      [2]   127 GCTCCTGTCTCCCCCC...CCCAGTGTTGCAGAG
      [3]   109 GTGTGTGGTGATGCCA...CCCAGTGTTGCAGAG
      ...   ... ...
 [289968]   109 GTGTGTGGTGATGCCA...CCCAGTGTTGCAGAG
 [289969]   354 CTTGCCGTCAGCCTTT...TGACAACCTAGGCCA
```

- Acts like a *vector*, e.g., `length()`, `[`, `[[`
- Many methods – `methods(class="DNAStringSet")` – e.g., `reverseComplement()`, `letterFrequency()`, ...

Sequences: packages

Biostrings	General purpose biological sequence representation.
BSgenome	Whole-genome representation.
ShortRead	High-throughput sequencing.

Sequences: classes

DNAString	Single DNA sequence, e.g., chromosome
DNAStringSet	Vector of DNA sequences. Actually, `XString`, `XStringSet`: `X` could be DNA, RNA, AA)
BSgenome	Collection of (large) DNA sequences
ShortReadQ	High-throughput reads & their qualities

Sequences: file references

TwoBitFile, FaFile `.2bit` (in *rtracklayer*) or `.fa` (in *Rsamtools*) indexed genome-scale fasta files.

FastqFile , e.g., *FastqStreamer* (in *ShortRead*)

Effectively manage large data

- *Restrict* input to specific genomic locations
- *Iterate* through large files in chunks

Sequences: annotations

*BSgenome.** packages

- E.g., *BSgenome.Hsapiens.UCSC.hg19*
- Packages containing whole-genome sequences for model organisms

AnnotationHub resources

- e.g., Ensembl FASTA files

Ranges: *GRanges* representation

```
> gr = exons(TxDb.Hsapiens.UCSC.hg19.knownGene); gr
GRanges with 289969 ranges and 1 metadata column:
           seqnames              ranges strand |   exon_id
              <Rle>           <IRanges>  <Rle> | <integer>
       [1]     chr1   [11874, 12227]      + |         1
       [2]     chr1   [12595, 12721]      + |         2
       [3]     chr1   [12613, 12721]      + |         3
       ...      ...                 ...    ... ...       ...
  [289967]     chrY [59358329, 59359508]      - |    277748
  [289968]     chrY [59360007, 59360115]      - |    277749
  [289969]     chrY [59360501, 59360854]      - |    277750
  ---
  seqinfo: 93 sequences (1 circular) from hg19 genome
```

GRanges

```
length(gr); gr[1:5]
seqnames(gr)
start(gr)
end(gr)
width(gr)
strand(gr)
```

DataFrame

```
mcols(gr)
gr$exon_id
```

Seqinfo

```
seqlevels(gr)
seqlengths(gr)
genome(gr)
```

- Data: aligned reads, called peaks, SNP locations, CNVs, ...
- Annotation: gene models, variants, regulatory regions, ...

Ranges: *GRangesList* representation

```
> grl = exonsBy(TxDb.Hsapiens.UCSC.hg19.knownGene, "tx", use.names=TRUE); grl
GRangesList of length 82960:
$uc001aaa.3
GRanges with 3 ranges and 3 metadata columns:
      seqnames          ranges strand |   exon_id   exon_name exon_rank
         <Rle>       <IRanges>  <Rle> | <integer> <character> <integer>
  [1]     chr1 [11874, 12227]      + |         1        <NA>         1
  [2]     chr1 [12613, 12721]      + |         3        <NA>         2
  [3]     chr1 [13221, 14409]      + |         5        <NA>         3

$uc010nxq.1
GRanges with 3 ranges and 3 metadata columns:
      seqnames          ranges strand | exon_id exon_name exon_rank
  [1]     chr1 [11874, 12227]      + |       1      <NA>         1
  [2]     chr1 [12595, 12721]      + |       2      <NA>         2
  [3]     chr1 [13403, 14409]      + |       6      <NA>         3

$uc010nxr.1
GRanges with 3 ranges and 3 metadata columns:
      seqnames          ranges strand | exon_id exon_name exon_rank
  [1]     chr1 [11874, 12227]      + |       1      <NA>         1
  [2]     chr1 [12646, 12697]      + |       4      <NA>         2
  [3]     chr1 [13221, 14409]      + |       5      <NA>         3

...
<82957 more elements>
---
 seqinfo: 93 sequences (1 circular) from hg19 genome
```

```
GRangesList
  (list of GRanges)
  length(grl)
  grl[1:3]
  shift(grl, 1)
  range(grl)
```

```
GRanges
  grl[[2]]
  grl[["uc010nxq.1"]]
```

```
Two kinds of fun!
  introns =
    psetdiff(range(grl), grl)

  grr = unlist(grl)
  ## transform grr, then...
  grl = relist(grr, grl)
```

'flesh' 'skeleton'

Ranges: packages

GenomicRanges Essential representation and operations

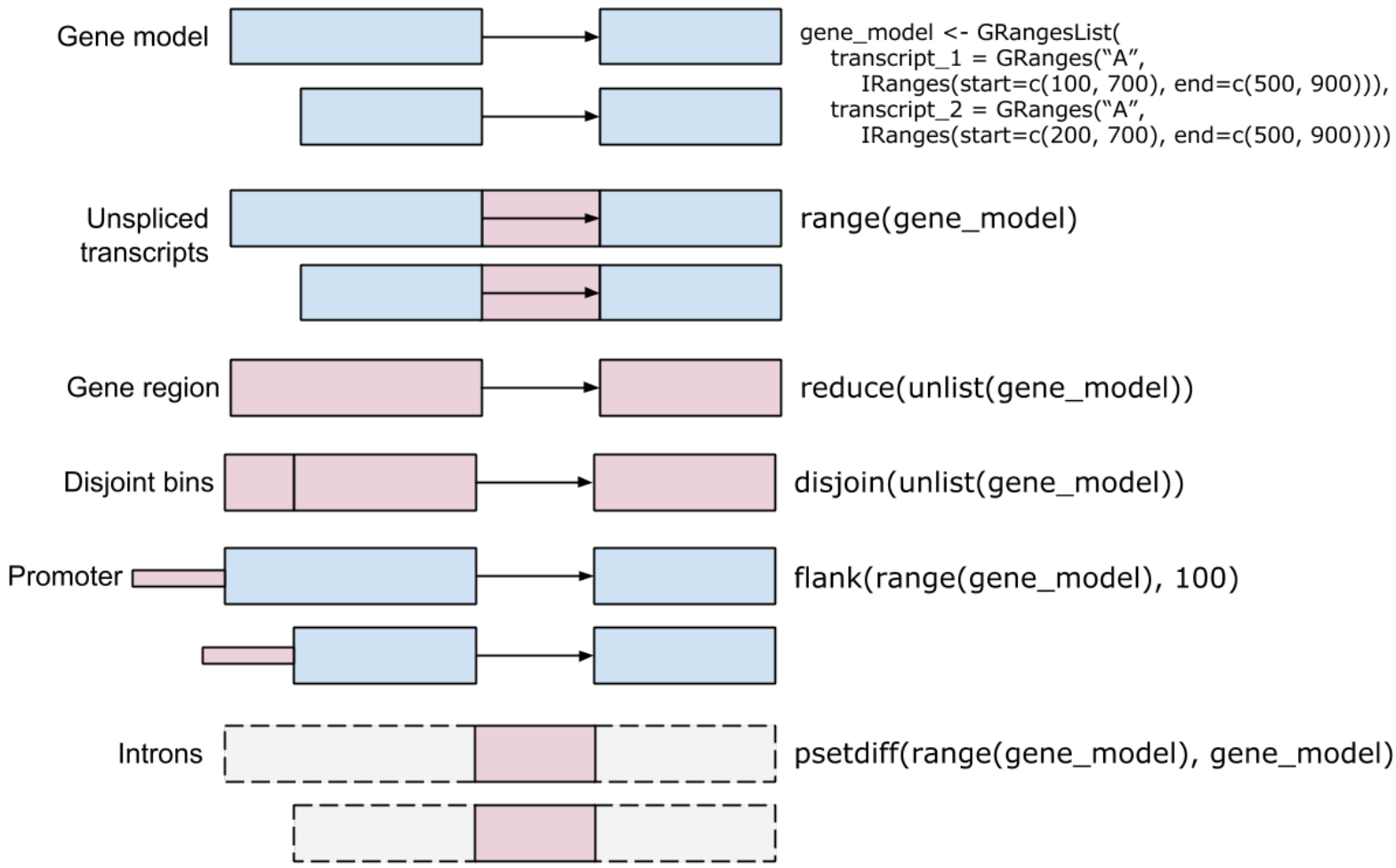
GenomicAlignments Aligned reads as genomic ranges

GenomicFeatures Annotations as genomic ranges

rtracklayer Annotation (e.g., BED, GTF) input

A little more advanced usage: *IRanges* (); *S4Vectors* (underling conceptual ideas)

Ranges: operations



- Many more, e.g., `methods(class="GRanges")`

Ranges: `findOverlaps()`

- Overlaps between `query` and `subject` genomic ranges
- Different types of overlap, e.g., 'any', 'within', ...

```
> q <- GRanges("chr1", IRanges(10, 20))
> s <- GRanges("chr1", IRanges(5, width=c(3, 6, 9)))
> findOverlaps(q, s)
Hits object with 2 hits and 0 metadata columns:
      queryHits subjectHits
      <integer>   <integer>
  [1]         1           2
  [2]         1           3
  -------
  queryLength: 1
  subjectLength: 3
```

- *Hits* object describing many-to-many relationship between overlapping ranges.

Ranges: working with files

import (*rtracklayer*) for BED, GTF, and other common web file import functions. *BEDFile*, *GTFFile*, etc.

readGAlignments / readGAlignmentsList (*GenomicAlignments*) for aligned reads in BAM files

BamFile (*Rsamtools*) for lower-level access to BAM files, e.g., restriction and iteration

Ranges: annotation

*TxDb.** packages

- E.g., *TxDb.Hsapiens.UCSC.hg19.knownGene*
- Genomic ranges for exons, transcripts, coding sequences, and how these are ordered into gene models, e.g., exons grouped by transcript

AnnotationHub resources

- Ensembl gene models
- Roadmap Epigenomics regulatory marks
- Many other range-based resources

What's to love about a data.frame?

- Coordinated data management
- Familiarity
- Interoperability

And yet...

- Not all columns are equal – seqlevels, start, width, strand are required, other columns are optional.
- Columns have special meaning – e.g., widths of genomic ranges are $>= 0$
- Not every column is a base *R* vector – *DNAStringSet*
- Data from different tables are often related – information about genomes

Principled versus precocious?

- The tidyverse as principled – a few well-designed orthogonal functions operating exclusively (?) on data.frame derivatives
- *R* and *Bioconductor* as precocious – a large number of classes and specialized functions
- Some of precociousness is lack of principle, but some reflects high-level summary of complex work flows

Other resources

- Workflows & package vignettes
- *GenomicRanges* and other 'cheat sheets'
- Course material
- Support site tutorials

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