Making and Utilizing TranscriptDb Objects

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1 Introduction

The *GenomicFeatures* package retrieves and manages transcript-related features from the UCSC Genome Bioinformatics¹ and BioMart² data resources. The package is useful for ChIP-chip, ChIP-seq, and RNA-seq analyses.

> library("GenomicFeatures")

2 TranscriptDb Objects

The GenomicFeatures package uses TranscriptDb objects to store transcript metadata. This class maps the 5' and 3' untranslated regions (UTRs), protein coding sequences (CDSs) and exons for a set of mRNA transcripts to their associated genome. TranscriptDb objects have numerous accessors functions to allow such features to be retrieved individually or grouped together in a way that reflects the underlying biology.

All *TranscriptDb* objects are backed by a SQLite database that manages genomic locations and the relationships between pre-processed mRNA transcripts, exons, protein coding sequences, and their related gene identifiers.

3 Retrieving Data from *TranscriptDb* objects

3.1 Loading Transcript Data

There are two ways that users can load pre-existing data to generate a TranscriptDb object. One method is to use the loadDb method to load the object directly from an appropriate sqlite database file.

¹http://genome.ucsc.edu/

²http://www.biomart.org/

Here we are loading a previously created TranscriptDb object based on UCSC known gene data. This database only contains a small subset of the possible annotations for human and is only included to demonstrate and test the functionality of the GenomicFeatures packageas a demonstration.

```
> samplefile <- system.file("extdata", "UCSC_knownGene_sample.sqlite",
                             package="GenomicFeatures")
> txdb <- loadDb(samplefile)</pre>
> txdb
TranscriptDb object:
| Db type: TranscriptDb
| Supporting package: GenomicFeatures
| Data source: UCSC
| Genome: hg18
| Genus and Species: Homo sapiens
| UCSC Table: knownGene
| Resource URL: http://genome.ucsc.edu/
| Type of Gene ID: Entrez Gene ID
| Full dataset: no
| miRBase build ID: NA
| transcript_nrow: 135
| exon_nrow: 544
| cds_nrow: 324
| Db created by: GenomicFeatures package from Bioconductor
| Creation time: 2012-04-13 14:47:54 -0700 (Fri, 13 Apr 2012)
| GenomicFeatures version at creation time: 1.9.4
| RSQLite version at creation time: 0.11.1
| DBSCHEMAVERSION: 1.0
   In this case, the TranscriptDb object has been returned by the loadDb
method.
   More commonly however, we expect that users will just load a Tran-
scriptDb annotation package like this:
> library(TxDb.Hsapiens.UCSC.hg19.knownGene)
> txdb <- TxDb.Hsapiens.UCSC.hg19.knownGene #shorthand (for convenience)
> txdb
TranscriptDb object:
| Db type: TranscriptDb
```

```
| Supporting package: GenomicFeatures
| Data source: UCSC
| Genome: hg19
| Genus and Species: Homo sapiens
| UCSC Table: knownGene
| Resource URL: http://genome.ucsc.edu/
| Type of Gene ID: Entrez Gene ID
| Full dataset: yes
| miRBase build ID: GRCh37
| transcript_nrow: 80922
| exon_nrow: 286852
| cds_nrow: 235842
| Db created by: GenomicFeatures package from Bioconductor
| Creation time: 2012-03-12 21:45:23 -0700 (Mon, 12 Mar 2012)
| GenomicFeatures version at creation time: 1.7.30
| RSQLite version at creation time: 0.11.1
| DBSCHEMAVERSION: 1.0
```

Loading the package like this will also create a TranscriptDb object, and by default that object will have the same name as the package itself.

3.2 Pre-filtering data based on Chromosomes

It is possible to filter the data that is returned from a TranscriptDb object based on it's chromosome. This can be a useful way to limit the things that are returned if you are only interested in studying a handful of chromosomes.

To determine which chromosomes are currently active, use the isActiveSeq method. For example:

```
> head(isActiveSeq(txdb))
chr1 chr2 chr3 chr4 chr5 chr6
TRUE TRUE TRUE TRUE TRUE TRUE
```

Will tell you all the chromosomes that are active for the TxDb.Hsapiens.UCSC.hg19.knownGene *TranscriptDb* object (by default it will be all of them).

If you then wanted to only set Chromosome 1 to be active you could do it like this:

```
> isActiveSeq(txdb)[seqlevels(txdb)] <- FALSE
> isActiveSeq(txdb) <- c("chr1"=TRUE)</pre>
```

So if you ran this, then from this point on in your R session only chromosome 1 would be consulted when you call the various retrieval methods..

Exercise 1

Use isActiveSeq to set only chromsome 15 to be active. BTW, the rest of this vignette will assume you have succeeded at this.

Solution:

- > isActiveSeq(txdb)[seqlevels(txdb)] <- FALSE
 > isActiveSeq(txdb) <- c("chr15"=TRUE)</pre>
- 3.3 Retrieving data using the select method

> keys <- c("100033416", "100033417", "100033420")

"TXCHROM"

The TranscriptDb objects inherit from AnnotationDb objects (just as the ChipDb and OrgDb objects do). One of the implications of this relationship is that these object ought to be used in similar ways to each other. Therefore we have written supporting cols, keytypes, keys and select methods for TranscriptDb objects.

These methods can be a useful way of extracting data from a TranscriptDb object. And they are used in the same way that they would be used to extract information about a ChipDb or an OrgDb object. Here is a simple example of how to find the UCSC transcript names that match with a set of gene IDs.

```
> cols(txdb)
                   "CDSNAME"
 [1] "CDSID"
                                 "CDSCHROM"
                                               "CDSSTRAND"
                                                             "CDSSTART"
 [6] "CDSEND"
                   "EXONID"
                                 "EXONNAME"
                                               "EXONCHROM"
                                                             "EXONSTRAND"
                                 "GENEID"
[11] "EXONSTART"
                   "EXONEND"
                                               "TXID"
                                                             "EXONRANK"
```

"TXSTRAND"

> keytypes(txdb)

[16] "TXNAME"

[1] "GENEID" "TXID" "TXNAME" "EXONID" "EXONNAME" "CDSID" "CDSNAME"

"TXSTART"

"TXEND"

> select(txdb, keys = keys, cols="TXNAME", keytype="GENEID")

```
GENEID TXNAME
1 100033416 uc001yxl.4
2 100033417 uc001yxo.3
3 100033417 uc001yxq.3
4 100033420 uc001yxr.3
```

Exercise 2

For the genes in the example above, find the chromosome and strand information that will go with each of the transcript names.

Solution:

> cols(txdb)

```
[1] "CDSID"
                   "CDSNAME"
                                                             "CDSSTART"
                                 "CDSCHROM"
                                               "CDSSTRAND"
 [6] "CDSEND"
                   "EXONID"
                                 "EXONNAME"
                                               "EXONCHROM"
                                                             "EXONSTRAND"
[11] "EXONSTART"
                   "EXONEND"
                                 "GENEID"
                                               "TXID"
                                                             "EXONRANK"
[16] "TXNAME"
                                 "TXSTRAND"
                                               "TXSTART"
                                                             "TXEND"
                   "TXCHROM"
```

```
> cols <- c("TXNAME", "TXSTRAND", "TXCHROM")
> select(txdb, keys=keys, cols=cols, keytype="GENEID")
```

```
GENEID TXNAME TXCHROM TXSTRAND
1 100033416 uc001yx1.4 chr15 +
2 100033417 uc001yx0.3 chr15 +
3 100033417 uc001yxq.3 chr15 +
4 100033420 uc001yxr.3 chr15 +
```

3.4 Methods for returning *GRanges* objects

Retrieving data with select is useful, but sometimes it is more convenient to extract the result as GRanges objects. This is often the case when you are doing counting or specialized overlap operations downstream. For these use cases there is another family of methods available.

Perhaps the most common operations for a TranscriptDb object is to retrieve the genomic coordinates or ranges for exons, transcripts or coding sequences. The functions transcripts, exons, and cds return the coordinate information as a GRanges object.

As an example, all transcripts present in a TranscriptDb object can be obtained as follows:

```
> GR <- transcripts(txdb)
> GR[1:3]
```

GRanges with 3 ranges and 2 elementMetadata cols:

```
segnames
                            ranges strand |
                                                 tx_id
                                                           tx_name
       <Rle>
                         <IRanges> <Rle> | <integer> <character>
[1]
       chr15 [20362688, 20364420]
                                        + |
                                                 55646
                                                        uc001yte.1
[2]
       chr15 [20487997, 20496811]
                                        + |
                                                 55648
                                                       uc001ytf.1
[3]
       chr15 [20723929, 20727150]
                                        + |
                                                 55654 uc001ytj.3
seqlengths:
     chr15
 102531392
```

The transcripts function returns a *GRanges* class object. You can learn a lot more about the manipulation of these objects by reading the *GenomicRanges* introductory vignette. The show method for a *GRanges* object will display the ranges, seqnames (a chromosome or a contig), and strand on the left side and then present related metadata on the right side. At the bottom, the seqlengths display all the possible seqnames along with the length of each sequence.

In addition, the **transcripts** function can also be used to retrieve a subset of the transcripts available such as those on the +-strand of chromosome 1.

```
> GR <- transcripts(txdb, vals <- list(tx_chrom = "chr15", tx_strand = "+"))
> length(GR)

[1] 1718
> unique(strand(GR))

[1] +
Levels: + - *
```

The exons and cds functions can also be used in a similar fashion to retrive genomic coordinates for exons and coding sequences.

Exercise 3

Use exons to retrieve all the exons from chromosome 15. How does the length of this compare to the value returned by transcripts?

Solution:

> length(GR)

[1] 1718

```
> EX <- exons(txdb)
> EX[1:4]
GRanges with 4 ranges and 1 elementMetadata col:
      segnames
                              ranges strand |
                                                 exon_id
         <Rle>
                                       <Rle> | <integer>
                           <IRanges>
         chr15 [20362688, 20362858]
  [1]
                                           + |
                                                   201302
  [2]
         chr15 [20362943, 20363123]
                                           + |
                                                  201303
         chr15 [20364397, 20364420]
  [3]
                                           + |
                                                  201304
  [4]
         chr15 [20487997, 20488227]
                                           + |
                                                  201306
  seqlengths:
       chr15
   102531392
> length(EX)
[1] 10658
```

3.5 Working with Grouped Features

Often one is interested in how particular genomic features relate to each other, and not just their location. For example, it might be of interest to group transcripts by gene or to group exons by transcript. Such groupings are supported by the transcriptsBy, exonsBy, and cdsBy functions.

The following call can be used to group transcripts by genes:

```
> GRList <- transcriptsBy(txdb, by = "gene")
> length(GRList)
[1] 782
> names(GRList)[10:13]
[1] "100033424" "100033425" "100033426" "100033427"
```

```
> GRList[11:12]
GRangesList of length 2:
$100033425
GRanges with 1 range and 2 elementMetadata cols:
      segnames
                              ranges strand |
                                                  tx_id
                                                             tx_name
         <Rle>
                           <IRanges>
                                      <Rle> | <integer> <character>
  [1]
         chr15 [25324204, 25324297]
                                          + |
                                                  55814 uc001yxw.3
$100033426
GRanges with 1 range and 2 elementMetadata cols:
      seqnames
                              ranges strand | tx_id
                                                        tx_name
  [1]
         chr15 [25325288, 25325381]
                                          + | 55815 uc001yxx.3
seqlengths:
     chr15
 102531392
```

The transcriptsBy function returns a *GRangesList* class object. As with *GRanges* objects, you can learn more about these objects by reading the *GenomicRanges* introductory vignette. The show method for a *GRangesList* object will display as a list of *GRanges* objects. And, at the bottom the seqlengths will be displayed once for the entire list.

For each of these three functions, there is a limited set of options that can be passed into the by argument to allow grouping. For the transcriptsBy function, you can group by gene, exon or cds, whereas for the exonsBy and cdsBy functions can only be grouped by transcript (tx) or gene.

So as a further example, to extract all the exons for each transcript you can call:

```
> GRList <- exonsBy(txdb, by = "tx")
> length(GRList)

[1] 3281
> names(GRList)[10:13]

[1] "55653" "55654" "55655" "55656"
> GRList[[12]]
```

GRanges with 9 ranges and 3 elementMetadata cols:

	seqnames		ranges	strand	exon_id	exon_name	exon_rank
	<rle></rle>		<pre><iranges></iranges></pre>	<rle></rle>	<integer></integer>	<character></character>	<integer></integer>
[1]	chr15	[20746826,	20747114]	_	201358	<na></na>	1
[2]	chr15	[20745051,	20745179]	_	201357	<na></na>	2
[3]	chr15	[20743914,	20743964]	_	201356	<na></na>	3
[4]	chr15	[20743732,	20743825]	_	201355	<na></na>	4
[5]	chr15	[20743530,	20743636]	_	201354	<na></na>	5
[6]	chr15	[20742094,	20742152]	_	201353	<na></na>	6
[7]	chr15	[20741622,	20741763]	_	201352	<na></na>	7
[8]	chr15	[20739497,	20740969]	_	201351	<na></na>	8
[9]	chr15	[20737094,	20738840]	_	201350	<na></na>	9

seqlengths:

chr15

102531392

As you can see, the *GRangesList* objects returned from each function contain locations and identifiers grouped into a list like object according to the type of feature specified in the by argument. The object returned can then be used by functions like findOverlaps to contextualize alignments from high-throughput sequencing.

The identifiers used to label the *GRanges* objects depend upon the data source used to create the *TranscriptDb* object. So the list identifiers will not always be Entrez Gene IDs, as they were in the first example. Furthermore, some data sources do not provide a unique identifier for all features. In this situation, the group label will be a synthetic ID created by *GenomicFeatures* to keep the relations between features consistent in the database this was the case in the 2nd example. Even though the results will sometimes have to come back to you as synthetic IDs, you can still always retrieve the original IDs.

Exercise 4

Starting with the tx_ids that are the names of the GRList object we just made, use select to retrieve that matching transcript names. Remember that the list used a by argument = "tx", so the list is grouped by transcript IDs.

Solution:

```
> GRList <- exonsBy(txdb, by = "tx")
```

```
> tx_ids <- names(GRList)
> head(select(txdb, keys=tx_ids, cols="TXNAME", keytype="TXID"))

TXID     TXNAME
1 55644 uc021sev.1
2 55645 uc010tyw.1
3 55646 uc001yte.1
4 55647 uc021sew.1
5 55648 uc001ytf.1
6 55649 uc001ytg.3
```

Finally, the order of the results in a *GRangesList* object can vary with the way in which things were grouped. In most cases the grouped elements of the *GRangesList* object will be listed in the order that they occurred along the chromosome. However, when exons or CDS are grouped by transcript, they will instead be grouped according to their position along the transcript itself. This is important because alternative splicing can mean that the order along the transcript can be different from that along the chromosome.

3.6 Predefined grouping functions

The intronsByTranscript, fiveUTRsByTranscript and threeUTRsByTranscript are convenience functions that provide behavior equivalent to the grouping functions, but in prespecified form. These functions return a *GRangesList* object grouped by transcript for introns, 5' UTR's, and 3' UTR's, respectively. Below are examples of how you can call these methods.

```
> length(intronsByTranscript(txdb))
[1] 3281
> length(fiveUTRsByTranscript(txdb))
[1] 1770
> length(threeUTRsByTranscript(txdb))
[1] 1748
```

3.7 Getting the actual sequence data

The *GenomicFeatures* package also provides provides functions for converting from ranges to actual sequence (when paired with an appropriate *BSgenome* package).

```
> library(BSgenome.Hsapiens.UCSC.hg19)
> tx_seqs1 <- extractTranscriptsFromGenome(Hsapiens,
+ TxDb.Hsapiens.UCSC.hg19.knownGene)</pre>
```

And, once these sequences have been extracted, you can translate them into proteins with translate:

> translate(tx_seqs1)

```
A AAStringSet instance of length 3281
      width seq
   [1]
          10 SY*SKWTWHP
   [2]
           9 SAGSPV*SA
   [3]
         125 EDQDDEARVQYEGFRPGMYVRVEIENVPCEFV...EDHNGRQRLLKYTPQHMHCGAAFWA*FSDSCH
   [4]
           9 CFWLTSSGC
   [5]
         288 RIAS*GRAEFSSAQTSEIQRRRSSVLLSTDPG...TFFHSVIFLFFESVFYSVYFNYGNNCFFTVTD
   [6]
       1467 G*ASGSWEAASARRRGGAAGPCASGLRPGRKL...CSHGQQAL*IQSMVQDMHDKITLLK*MQQEKW
   [7]
        1394 A*DLGGSWRKDAL*IFVFGCPGSPRLQMVENR...EE*TQCVRRRIQMQQPQISSGCLLGFCNRFPR
   [8]
        1426 G*ASGSWEAASARRRGGAAGPCASGLRPGRKL...KYAWTCFQRAKRPPEVLSLLRYDDANKSVTSE
  [9]
        1022 KSLTYFLLTISPVALQAS*IELMLLFPFLFQL...GEWPSPFPVPW*RVVQQLLPWFPPQSPR*GGA
[3273]
         641 TIFQHTVQWD*VDSHCCAAITTTHFQHPSITP...YIADLGPGIAPSAPGTIPELPTFHTEVAEPLK
[3274]
         851 TIFQHTVQWD*VDSHCCAAITTTHFQHPSITP...LKRLPEKTAHQSIKN*AETNSVLLIKDL*LCR
[3275]
         151 AP*LL*GCSTPWQVRPMPCPSSSQTCGERRPS...PL*ASVFSFVKLW**W*LPGRIIVNIKLIKT*
[3276]
         440 EKLTFLCA*APSRSPRTMQKRDLGVFPATSAL...LKRLPEKTAHQSIKN*AETNSVLLIKDL*LCR
[3277]
         431 EKLTFLCA*APSRSPRTMQKRDLGVFPATSAL...S*EASRENGTPINKELSRNQQCAFNKGSLAVQ
         283 TYKMGY*HHPHRPHHHPQLLRCWPVHPHSHPQ...LRGFQRKRHTNQ*RTEQKPTVCF**RISSCAG
[3278]
[3279]
         550 LAVSLFFDLFFLFMCICCLLAQTSRVLSTGPL...LGTNRGRRQSLTPRRLHPAQLEILY*KHTVGF
[3280]
         496 LAVSLFFDLFFLFMCICCLLAQTSRVLSTGPL...RDQQGQEAVTDPETFASCTARDPLLKAHCWFL
```

531 LAVSLFFDLFFLFMCICCLLAQTSRVLSTGPL...LGTNRGRRQSLTPRRLHPAQLEILY*KHTVGF

Exercise 5

[3281]

But of course this is not a meaningful translation, because the call to extractTranscriptsFromGenome will have extracted all the transcribed regions of the genome regardless of whether or not they are translated. look at the manual page for extractTranscriptsFromGenome and see how you can use cdsBy to only translate only the coding regions.

Solution:

```
> cds_seqs <- extractTranscriptsFromGenome(Hsapiens, cdsBy(txdb, by="tx"))
> translate(cds_seqs)
 A AAStringSet instance of length 1819
      width seq
         102 MYVRVEIENVPCEFVQNIDPHYPIILGGLGNS...IPLYYIEDHNGRQRLLKYTPQHMHCGAAFWA*
  [1]
   [2]
       1159 MHAFCVGQYLEPDQEGVTIPDLGSLSSPLIDT...RRTTKLEQILLFIRRMNSVCEKENTNATASN*
        1135 MHAFCVGQYLEPDQEGVTIPDLGSLSSPLIDT...PSPVKAIFDKQELEPFRLFLPVTLSSSLGSV*
  [3]
        844 MHAFCVGQYLEPDQEGVTIPDLGSLSSPLIDT...LFDFLGSLLTMCSSDVPLLRGGWPSPFPVPW*
   [4]
   [5]
        343 MFPPEHPVEEVGRLLLCCLLKHEDLGHVALSL...YAWAVQNIRNVLVDASAKFKELEMSVGANAH*
        751 MLMWPQPHLPTHPHLPTHPHLPTHPHLPTHPH...QEQEEKMRRQEEKMWEQEVRLRQQEEKMQEH*
   [6]
   [7]
        435 MEWKLEQSMREQALLKAQLTQLKESLKEVQLE...PIVQDHQEHPGLGSNCCVPFFCWAWPPRRRR*
         101 MSCCLSSRVHITRPVLEQFLSFAKYLDGLSHG...FIGTATIYTTICRIGTVIKDNAHLKILLLGY*
   [8]
  [9]
         545 MVAEVCSMPAASAVKKPFDLRSKMGKWCHHRF...ENKILEEIESVKEKLLKTIQLNEEALTKTSI*
[1811]
         803 MAAEALAAEAVASRLERQEEDIRWLWSEVERL...NKIHGEILVTSAIDKLKNLRKTRTLNAEEAF*
[1812]
         183 MLQLAGVSNSTCGGVRNDSKAEENGSHSFMHS...TRCCFSELLSNLHSLGNKKTLVEASAEQADQG
[1813]
         313 MDEANHSVVSEFVFLGLSDSRKIQLLLFLFFS...LNPVIYTFRNKEMMVAMRRRCSQFVNYSKIF*
[1814]
         313 MNGMNHSVVSEFVFMGLTNSREIQLLLFVFSL...LNPVIYTFRNKDMKVAMRRLCSRLAHFTKIL*
[1815]
        274 MANLIVVVTVTSDPYLHSSLYILLANLSVIDL...LNPVIYTFRNREMKIAIRRVFGQFMGFRKTT*
         306 MVTEFIFLGLSDSQELQTFLFMLFFVFYGGIV...LNPIIYTLRNKDMKTAIRRLRKWDAHSSVKF*
[1816]
         136 MTPVRMQHSLAGQTYAVPLIQPDLRREEAVQQ...SIFTGAQDPGLQRRPRHRIQSKHRPLDERALQ
[1817]
         265 MLGAETEEKLFDAPLSISKREQLEQQVPENYF...GGAFARVSDSIPPLPPPQQPQAEEDEDDWES*
[1818]
         134 MSESINFSHNLGQLLSPPRCVVMPGMPFPSIR...CHGPGLQGSCYKGETQESVESRVLPGPRHRH*
Г18197
```

4 Creating New TranscriptDb Objects or Packages

The GenomicFeatures package provides functions to create TranscriptDb objects based on data downloaded from UCSC Genome Bioinformatics or BioMart. The following subsections demonstrate the use of these functions. There is also support for creating TranscriptDb objects from custom data sources using makeTranscriptDb; see the help page for this function for details.

4.1 Using makeTranscriptDbFromUCSC

The function makeTranscriptDbFromUCSC downloads UCSC Genome Bioinformatics transcript tables (e.g. "knownGene", "refGene", "ensGene") for a genome build (e.g. "mm9", "hg19"). Use the supportedUCSCtables utility function to get the list of supported tables.

> supportedUCSCtables()[1:4,]

		${\tt track}$	subtrack
knownGene	UCSC	Genes	<na></na>
knownGeneOld3	Old UCSC	Genes	<na></na>
wgEncodeGencodeManualV3	Gencode	Genes	Genecode Manual
${\tt wgEncodeGencodeAutoV3}$	Gencode	Genes	Genecode Auto

> mm9KG <- makeTranscriptDbFromUCSC(genome = "mm9", tablename = "knownGene")

The function makeTranscriptDbFromUCSC also takes an important argument called circ_seqs to label which chromosomes are circular. The argument is a character vector of strings that correspond to the circular chromosomes (as labeled by the source). To discover what the source calls their chromosomes, use the getChromInfoFromUCSC function to list them. By default, there is a supplied character vector that will attempt to label all the mitochondrial chromosomes as circular by matching to them. This is the DEFAULT_CIRC_SEQS vector. It contains strings that usually correspond to mitochondrial chromosomes. Once the database has been generated with the circular chromosomes tagged in this way, all subsequent analysis of these chromosomes will be able to consider their circularity for analysis. So it is important for the user to make sure that they pass in the correct strings to the circ_seqs argument to ensure that the correct sequences are tagged as circular by the database.

> head(getChromInfoFromUCSC("hg19"))

chrom length
1 chr1 249250621
2 chr2 243199373
3 chr3 198022430
4 chr4 191154276
5 chr5 180915260
6 chr6 171115067

4.2 Using makeTranscriptDbFromBiomart

Retrieve data from BioMart by specifying the mart and the data set to the makeTranscriptDbFromBiomart function (not all BioMart data sets are currently supported):

```
> mmusculusEnsembl <-
+ makeTranscriptDbFromBiomart(biomart = "ensembl",
+ dataset = "mmusculus_gene_ensembl")</pre>
```

As with the makeTranscriptDbFromUCSC function, the makeTranscriptDbFromBiomart function also has a circ_seqs argument that will default to using the contents of the DEFAULT_CIRC_SEQS vector. And just like those UCSC sources, there is also a helper function called getChromInfoFromBiomart that can show what the different chromosomes are called for a given source.

Using the makeTranscriptDbFromBiomart makeTranscriptDbFromUCSC functions can take a while and may also require some bandwidth as these methods have to download and then assemble a database from their respective sources. It is not expected that most users will want to do this step every time. Instead, we suggest that you save your annotation objects and label them with an appropriate time stamp so as to facilitate reproducible research.

4.3 Using makeTranscriptDbFromGFF

You can also extract transcript information from either GFF3 or GTF files by using the makeTranscriptDbFromGFF function. Usage is similar to makeTranscriptDbFromBiomart and makeTranscriptDbFromUCSC.

4.4 Saving and Loading a *TranscriptDb* Object

Once a TranscriptDb object has been created, it can be saved to avoid the time and bandwidth costs of recreating it and to make it possible to reproduce results with identical genomic feature data at a later date. Since TranscriptDb objects are backed by a SQLite database, the save format is a SQLite database file (which could be accessed from programs other than Rif desired). Note that it is not possible to serialize a TranscriptDb object using R's save function.

```
> saveDb(mm9KG, file="fileName.sqlite")
```

And as was mentioned earlier, a saved TranscriptDb object can be initialized from a sqlite file by simply using loadDb.

> mm9KG <- loadDb("fileName.sqlite")</pre>

4.5 Using makeTxDbPackageFromUCSC and makeTxDbPackageFrom-Biomart

It is often much more convenient to just make an annotation package out of your annotations. If you are finding that this is the case, then you should consider the convenience functions: makeTxDbPackageFromUCSC and makeTxDbPackageFromBiomart. These functions are similar to makeTranscriptDbFromUCSC and makeTranscriptDbFromBiomart except that they will take the extra step of actually wrapping the database up into an annotation package for you. This package can then be installed and used as of the standard TranscriptDb packages found on in the Bioconductor repository.

5 Session Information

R version 2.15.1 (2012-06-22)

Platform: x86_64-unknown-linux-gnu (64-bit)

locale:

```
[1] LC_CTYPE=en_US.UTF-8 LC_NUMERIC=C
[3] LC_TIME=en_US.UTF-8 LC_COLLATE=C
```

[5] LC_MONETARY=en_US.UTF-8 LC_MESSAGES=en_US.UTF-8

[7] LC_PAPER=C LC_NAME=C
[9] LC_ADDRESS=C LC_TELEPHONE=C
11] LC_MEASIDEMENT=ON_US_UTE=S_LC_IDENTIFICATION

[11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C

attached base packages:

[1] stats graphics grDevices utils datasets methods base

other attached packages:

- [1] BSgenome. Hsapiens. UCSC.hg19_1.3.17
- [2] BSgenome_1.25.3
- [3] Biostrings_2.25.8
- [4] TxDb.Hsapiens.UCSC.hg19.knownGene_2.7.1
- [5] GenomicFeatures_1.9.28
- [6] AnnotationDbi_1.19.28

- [7] Biobase_2.17.6
- [8] GenomicRanges_1.9.36
- [9] IRanges_1.15.22
- [10] BiocGenerics_0.3.0

loaded via a namespace (and not attached):

[1]	DBI_0.2-5	RCurl_1.91-1	RSQLite_0.11.1
[4]	Rsamtools_1.9.23	XML_3.9-4	biomaRt_2.13.1
[7]	bitops_1.0-4.1	rtracklayer_1.17.13	stats4_2.15.1
[10]	tools_2.15.1	zlibbioc_1.3.0	