ggbio: visualization toolkits for genomic data

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Contents

1	Getting started	3									
	1.1 Citation	3									
	1.2 Introduction	3									
2	Case study: building your first tracks	4									
	2.1 Add an ideogram track	4									
	2.2 Add a gene model track	5									
	2.2.1 Introduction	5									
	2.2.2 Make gene model from <i>OrganismDb</i> object	5									
	2.2.3 Make gene model from <i>TranscriptDb</i> object	9									
	2.2.4 Make gene model from <i>GRangesList</i> object	10									
	2.3 Add a reference track	13									
	2.3.1 Semantic zoom	13									
	2.4 Add an alignment track	15									
		19									
		19 22									
	2.6 Building your tracks	22									
3	Simple navigation	24									
4	26 26										
	4.1 how to make circular plots	26									
	4.1.1 Introduction	26									
		26									
	4.1.3 Complex arragnment of plots	34									
	4.2 How to make grandlinear plots	35									
	4.2.1 Introduction	36									
	4.2.2 Corrdinate genome	37									
	4.2.3 Convenient plotGrandLinear function	38									
	4.2.4 How to highlight some points?	40									
	4.3 How to make stacked karyogram overview plots	41									
	4.3.1 Introduction	41									
	4.3.2 Create karyogram temlate	41									
	4.3.3 Add data on karyogram layout	43									
	4.3.4 Add more data using layout_karyogram function	46									
		48									
		E۵									
5	Link ranges to your data	50									
6	Miscellaneous 52										
	6.1 Themes	52									
	6.1.1 Plot theme	52									
	6.1.2 Track theme	58									

7 Session Information

60

Chapter 1

Getting started

1.1 Citation

```
citation("ggbio")
##
## To cite package 'ggbio' in publications use:
##
##
     Tengfei Yin, Dianne Cook and Michael Lawrence (2012): ggbio: an R
##
     package for extending the grammar of graphics for genomic data Genome
##
     Biology 13:R77
##
## A BibTeX entry for LaTeX users is
##
##
     @Article{,
##
       title = {ggbio: an R package for extending the grammar of graphics for genomic data},
##
       author = {Tengfei Yin and Dianne Cook and Michael Lawrence},
       journal = {Genome Biology},
##
       volume = \{13\},
##
##
       number = \{8\},
##
       pages = \{R77\},
##
       year = \{2012\},\
       publisher = {BioMed Central Ltd},
##
##
     }
```

1.2 Introduction

ggbio is a Bioconductor package building on top of ggplot2(), leveraging the rich objects defined by Bioconductor and its statistical and computational power, it provides a flexible genomic visualization framework, extends the grammar of graphics into genomic data, try to delivers high quality, highly customizable graphics to the users.

What it features

- autoplot function provides ready-to-use template for Bioconductor objects and different types of data.
- flexible low level components to use grammar of graphics to build you graphics layer by layer.
- layout transformation, so you could generate circular plot, grandlinear plot, stacked overview more easily.
- flexible tracks function to bind any ggplot2(), ggbio based plots.

Chapter 2

Case study: building your first tracks

In this chapter, you will learn

- how to add ideogram track.
- How to add gene model track.
- how to add track for bam files to visualize coverage and mismatch summary.
- how to add track for vcf file to visualize the variants.

2.1 Add an ideogram track

Ideogram provides functionality to construct ideogram, check the manual for more flexible methods. We build genome *hg19*, *hg18*, *mm10*, *mm9* inside, so you don't have download it on the fly. When embed with tracks, ideogram show zoomed region highlights automatically. xlim has special function here, is too changed highlighted zoomed region on the ideogram.

```
library(ggbio)
p.ideo <- Ideogram(genome = "hg19")
p.ideo</pre>
```



```
library(GenomicRanges)
## special highlights instead of zoomin!
p.ideo + xlim(GRanges("chr2", IRanges(1e8, 1e8+1000000)))
```

chr2

2.2 Add a gene model track

2.2.1 Introduction

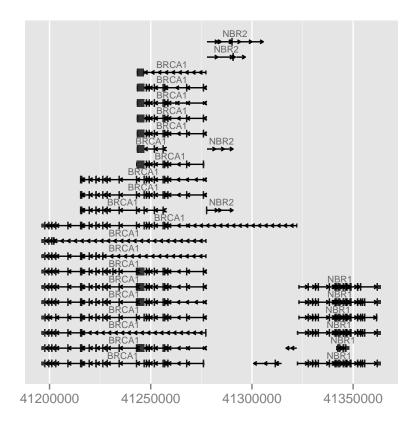
Gene model track is one of the most frequently used track in genome browser, it is composed of genetic features CDS, UTR, introns, exons and non-genetic region. In *ggbio* we support three methods to make gene model track:

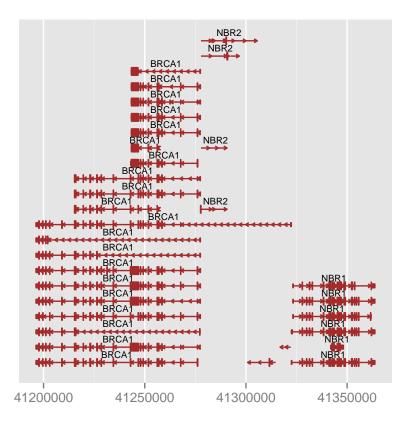
- OrganismDb object: recommended, support gene symbols and other combination of columns as label.
- TranscriptDb object: don't support gene symbol labeling.
- *GRangesList* object: flexible, if you don't have annotation package available for the first two methods, you could prepare a data set parsed from gtf file, you can simply use it and plot it as gene model track.

2.2.2 Make gene model from OrganismDb object

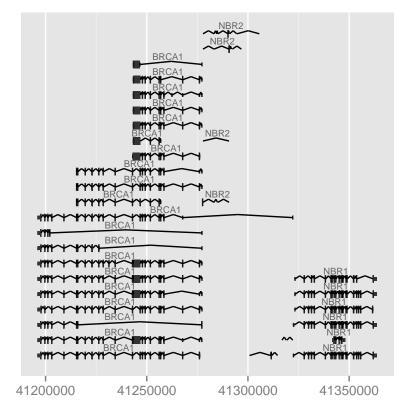
OrganismDb object has a simpler API to retrieve data from different annotation resources, so we could label our transcripts in different ways

```
library(ggbio)
library(Homo.sapiens)
class(Homo.sapiens)
## [1] "OrganismDb"
## attr(,"package")
## [1] "OrganismDbi"
##
data(genesymbol, package = "biovizBase")
wh <- genesymbol[c("BRCA1", "NBR1")]
wh <- range(wh, ignore.strand = TRUE)
p.txdb <- autoplot(Homo.sapiens, which = wh)
p.txdb</pre>
```



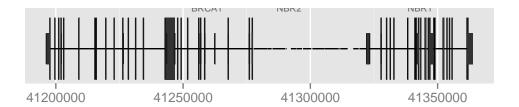


To change the intron geometry, use gap.geom to control it, check out geom_alignment for more control parameters. autoplot(Homo.sapiens, which = wh, gap.geom = "chevron")



To collapse all features, use stat 'reduce'

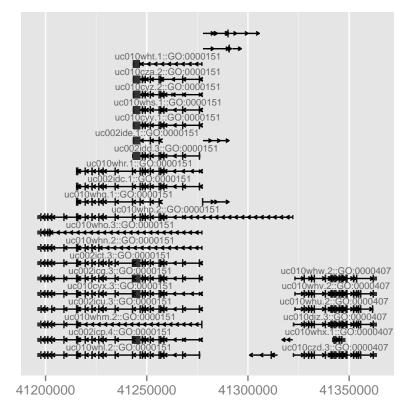
autoplot(Homo.sapiens, which = wh, stat = "reduce")



Label could be turned off by setting it to FALSE, you could also use expression to make a flexible label combination from column names.

columns(Homo.sapiens)						
##	[1]	"GOID"	"TERM"	"ONTOLOGY"	"DEFINITION"	"ENTREZID"
##	[6]	"PFAM"	"IPI"	"PROSITE"	"ACCNUM"	"ALIAS"
##	[11]	"CHR"	"CHRLOC"	"CHRLOCEND"	"ENZYME"	"MAP"
##	[16]	"PATH"	"PMID"	"REFSEQ"	"SYMBOL"	"UNIGENE"
##	[21]	"ENSEMBL"	"ENSEMBLPROT"	"ENSEMBLTRANS"	"GENENAME"	"UNIPROT"
##	[26]	"GO"	"EVIDENCE"	"GOALL"	"EVIDENCEALL"	"ONTOLOGYALL"
##	[31]	"OMIM"	"UCSCKG"	"CDSID"	"CDSNAME"	"CDSCHROM"
##	[36]	"CDSSTRAND"	"CDSSTART"	"CDSEND"	"EXONID"	"EXONNAME"
##	[41]	"EXONCHROM"	"EXONSTRAND"	"EXONSTART"	"EXONEND"	"GENEID"

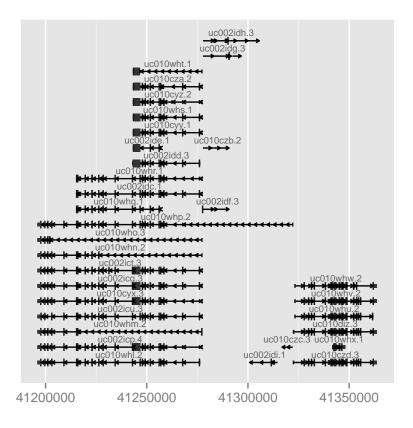
[46] "TXID" "EXONRANK" "TXNAME" "TXCHROM" "TXSTRAND"
[51] "TXSTART" "TXEND"
autoplot(Homo.sapiens, which = wh, columns = c("TXNAME", "GO"), names.expr = "TXNAME::GO")



2.2.3 Make gene model from TranscriptDb object

TranscriptDb doesn't contain any gene symbol information, so we use tx_id as default for label.

library(TxDb.Hsapiens.UCSC.hg19.knownGene)
txdb <- TxDb.Hsapiens.UCSC.hg19.knownGene
autoplot(txdb, which = wh)</pre>



2.2.4 Make gene model from GRangesList object

Sometimes your gene model is not available as none of *OrganismDb* or *TranscriptDb* object, it's may be stored in a table, you could simple parse it into a *GRangeList* object.

- each group indicate one transcripts
- names of group are shown as labels
- this object must has a column contains following key word: cds, exon, intron, and it's not case sensitive. use type to map this column. By default, we will try to parse 'type' column.

Let's make a sample GRangesList object which contains all information, and fake some labels.

```
library(biovizBase)
gr.txdb <- crunch(txdb, which = wh)
## change column to 'model'
colnames(values(gr.txdb))[4] <- "model"</pre>
grl <- split(gr.txdb, gr.txdb$tx_id)</pre>
## fake some randome names
names(grl) <- sample(LETTERS, size = length(grl), replace = TRUE)</pre>
grl
## GRangesList of length 32:
## $F
## GRanges with 7 ranges and 4 metadata columns:
                              ranges strand |
##
         seqnames
                                                  tx_id
                                                           tx_name gene_id
            <Rle>
                 <IRanges> <Rle> | <factor>
##
                                                           <factor> <factor>
         chr17 [41277600, 41277787]
                                       + |
                                                  61241 uc002idf.3
                                                                       10230
##
     [1]
```

```
##
      [2]
              chr17 [41283225, 41283287] + |
                                                             61241 uc002idf.3 10230
##
      [3]
              chr17 [41284973, 41285154]
                                                     + |
                                                             61241 uc002idf.3 10230
      [4]
              chr17 [41290674, 41292342]
                                                    + |
                                                             61241 uc002idf.3
##
                                                                                     10230
      [5] chr17 [41277788, 41283224]
##
                                                  * |
                                                             61241 uc002idf.3 10230
      [6] chr17 [41283288, 41284972]
                                                  * |
##
                                                             61241 uc002idf.3 10230
              chr17 [41285155, 41290673] * |
                                                             61241 uc002idf.3 10230
##
      [7]
##
              model
##
          <factor>
##
      [1]
              exon
      [2]
##
               exon
      [3]
##
               exon
##
      [4] exon
      [5]
##
                gap
##
      [6]
              gap
##
      [7]
                gap
##
## $H
## GRanges with 3 ranges and 4 metadata columns:
                                    ranges strand | tx_id tx_name gene_id model
##
      seqnames
      [1] chr17 [41277600, 41277787] + | 61242 uc010czb.2 10230 exon
##
      [2] chr17 [41290674, 41292342]
                                                  + | 61242 uc010czb.2 10230 exon
##
             chr17 [41277788, 41290673] * | 61242 uc010czb.2 10230
##
      [3]
                                                                                        gap
##
## $L
   GRanges with 9 ranges and 4 metadata columns:
##
##
       seqnames
                                    ranges strand | tx_id
                                                                  tx_name gene_id model
##
      [1] chr17 [41277600, 41277787] + | 61243 uc002idg.3 10230 exon

      [2]
      chr17 [41283225, 41283287]
      + | 61243 uc002idg.3
      10230 exon

      [3]
      chr17 [41290674, 41290939]
      + | 61243 uc002idg.3
      10230 exon

      [4]
      chr17 [41291833, 41292300]
      + | 61243 uc002idg.3
      10230 exon

##
##
##
##
      [5] chr17 [41296745, 41297125]
                                                  + | 61243 uc002idg.3
                                                                                10230 exon
##
     [6] chr17 [41277788, 41283224]
                                                  * | 61243 uc002idg.3
                                                                                10230
                                                                                        gap

      [7]
      chr17 [41283288, 41290673]
      * | 61243 uc002idg.3

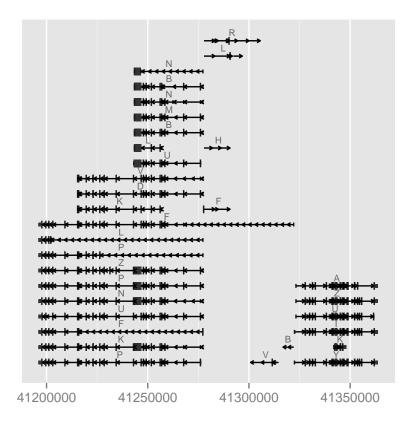
      [8]
      chr17 [41290940, 41291832]
      * | 61243 uc002idg.3

      [9]
      chr17 [41292301, 41296744]
      * | 61243 uc002idg.3

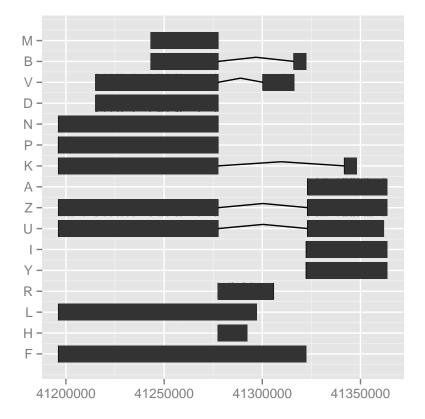
##
                                                                                10230
                                                                                          gap
##
                                                                                10230
                                                                                          gap
                                                                                10230 gap
##
##
## ...
## <29 more elements>
## ---
## seqlengths:
##
      chr17
## 81195210
```

We get our example data ready, it meets all requirements, to make it a gene model track it's pretty simple to use autoplot, but don't forget mapping because we changed our column names, assume you store you model key words in column 'model'.

autoplot(grl, aes(type = model))



ggplot() + geom_alignment(grl, type = "model")



2.3 Add a reference track

To add a reference track, we need to load a *BSgenome* object from the annotation package. You can choose to plot the sequence as *text*, *rect*, *segment*.

2.3.1 Semantic zoom

Here we introduce semantic zoom in *ggbio*, for some plots like reference sequence, we use pre-defined zoom level threshold to automatically assign geom to the track, unless the geom is explicitly specified. In the example below, when your region is too wide we show text 'zoom in to see text', when you zoom into different level, it shows you different details. zoom is a function we will introduce more in chapter 3 when we introduce more about navigation.

You can pass a zoom in factor into zoom function, if it's over 1 it's zooming out, if it's smaller than 1 it's zooming in.

```
library(BSgenome.Hsapiens.UCSC.hg19)
bg <- BSgenome.Hsapiens.UCSC.hg19
p.bg <- autoplot(bg, which = wh)
## no geom
p.bg</pre>
```



To override a zemantic zoom threshold, you simply provide a geom explicitly.

41279990

library(BSgenome.Hsapiens.UCSC.hg19)
bg <- BSgenome.Hsapiens.UCSC.hg19
force to use geom 'segment' at this level
autoplot(bg, which = resize(wh, width = width(wh)/2000), geom = "segment")</pre>

41280010

41280030

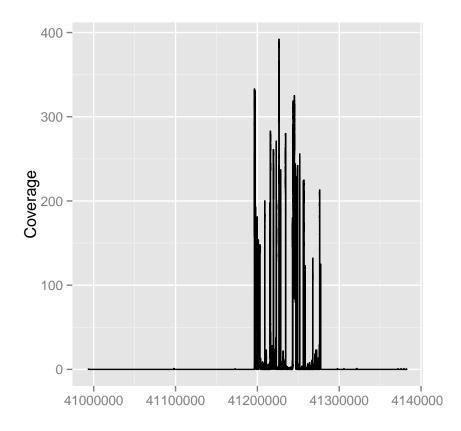
2.4 Add an alignment track

ggbio supports visuaization of alignemnts file stored in bam, autoplot method accepts

- bam file path (indexed)
- BamFile object
- GappedAlignemnt object

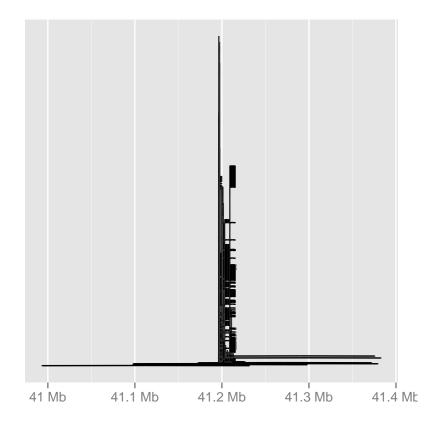
It's simple to just pass a file path to autoplot function, you can stream a chunk of region by providing 'which' parameter. Otherwise please use method 'estiamte' to show overall estiamted coverage.

```
fl.bam <- system.file("extdata", "wg-brca1.sorted.bam", package = "biovizBase")
wh <- keepSeqlevels(wh, "chr17")
autoplot(fl.bam, which = wh)</pre>
```



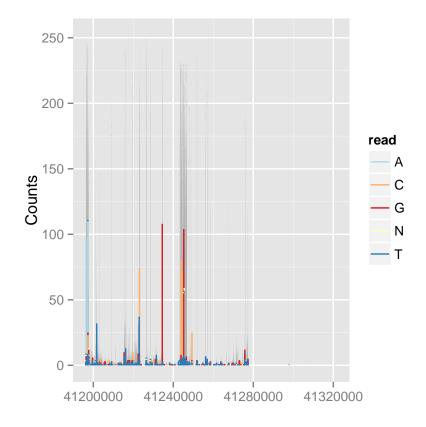
geom 'gapped pair' will show you alignments.

fl.bam <- system.file("extdata", "wg-brca1.sorted.bam", package = "biovizBase")
wh <- keepSeqlevels(wh, "chr17")
autoplot(fl.bam, which = resize(wh, width = width(wh)/10), geom = "gapped.pair")</pre>



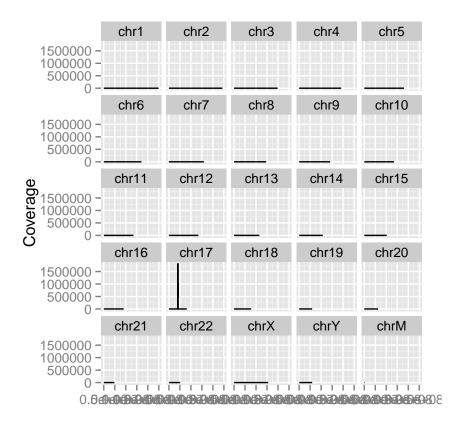
To show mismatch proportion, you have to provide reference sequence, the mismatched proportion is color coded in the bar chart.

```
library(BSgenome.Hsapiens.UCSC.hg19)
bg <- BSgenome.Hsapiens.UCSC.hg19
p.mis <- autoplot(fl.bam, bsgenome = bg, which = wh, stat = "mismatch")
p.mis</pre>
```

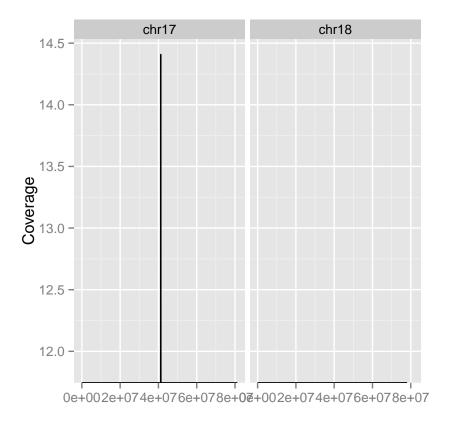


To view overall estimated coverage distribution, please use method 'estiamte'. 'which' parameter also accept characters. And there is a hidden value called '..coverage..' to let you do simple transformation in aes().

autoplot(fl.bam, method = "estimate")



autoplot(fl.bam, method = "estimate", which = paste0("chr", 17:18), aes(y = log(..coverage..)))



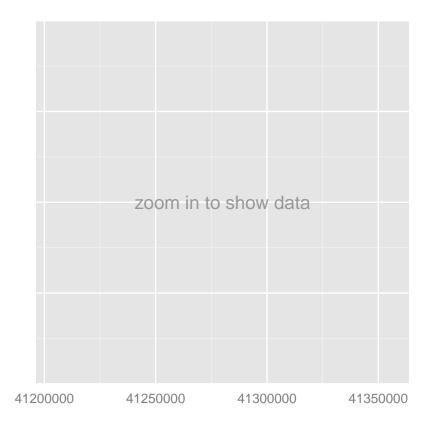
2.5 Add a variants track

This track is supported by semantic zoom.

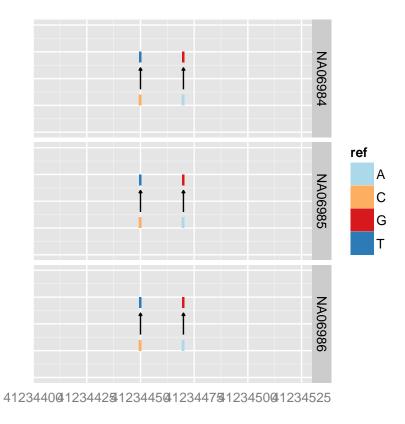
To view your variants file, you could

- Import it using package VariantAnntoation as VCF object, then use autoplot
- Convert it into *VRanges* object and use autoplot.
- Simply provide vcf file path in autoplot().

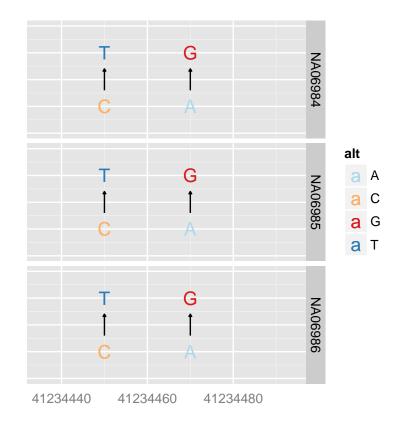
```
library(VariantAnnotation)
fl.vcf <- system.file("extdata", "17-1409-CEU-brca1.vcf.bgz", package="biovizBase")
vcf <- readVcf(fl.vcf, "hg19")
vr <- as(vcf[, 1:3], "VRanges")
vr <- renameSeqlevels(vr, value = c("17" = "chr17"))
## small region contains data
gr17 <- GRanges("chr17", IRanges(41234400, 41234530))
p.vr <- autoplot(vr, which = wh)
## none geom
p.vr</pre>
```



rect geom
p.vr + xlim(gr17)

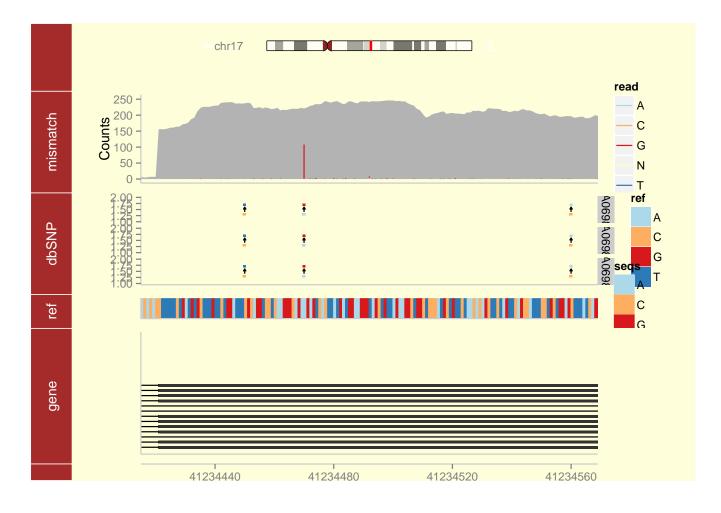


text geom
p.vr + xlim(gr17) + zoom()



```
You can simply overide geom
autoplot(vr, which = wh, geom = "rect", arrow = FALSE)
```

2.6 Building your tracks



Chapter 3

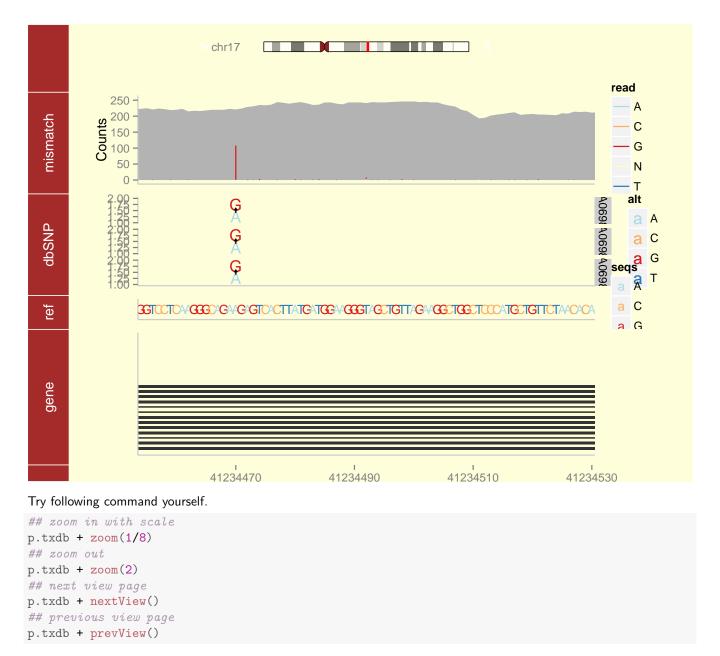
Simple navigation

We try to provide a simple navigation API for your plot, so you could zoom in and zoom out, or go through view chunks one by one.

- zoom: put a factor inside and you can zoom in or zoom out
- nextView: switch to next view
- prevView: switch to previous view

Navigation function also works for tracks plot too.

zoom in
tks + zoom()



Don't forget xlim accept *GRanges* object (single row), so you could simply prepare a *GRanges* to store the region of interests and go through them one by one.

Chapter 4

Overview plots

Overview is a good way to show all events at the same time, give overall summary statics for the whole genome. In this chapter, we will introcue three different layouts that are used a lots in genomic data visualization.

4.1 how to make circular plots

4.1.1 Introduction

Circular view is a special layout in *ggbio*, this idea has been implemented in many different software, for example, the *Circos* project. However, we keep the grammar of graphics for users, so mapping variables to aesthetics is very easy, *ggbio* leverage the data structure defiend in *Bioconductor* to make this process as simple as possible.

4.1.2 Builling circular plot layer by layer

Ok, let's start to process some raw data to the format we want. The data used in this study is from this a paper¹. In this tutorial, We are going to

- 1. Visualize somatic mutation as segment.
- 2. Visualize inter, intro-chromosome rearrangement as links.
- 3. Visualize mutation score as point tracks with grid-background.
- 4. Add scale and ticks and labels.
- 5. To arrange multiple plots and legend. create multiple sample comparison.

All the raw data processed and stored in GRanges ready for use, you can simply load the sample data from biovizBase

data("CRC", package = "biovizBase")

layout_circle is depreicated, because you have to set up radius and trackWidth manually with this function for creating circular plot.

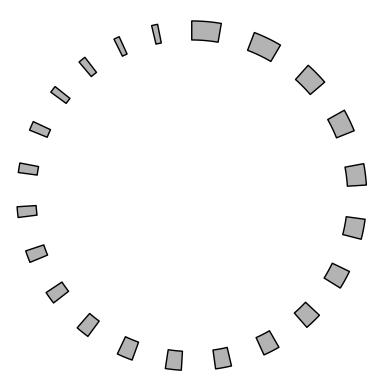
We now present the new circle function, it accepts Granges object, and users don't have to specify radius, track width, you just add them one by one, it will be automatically created from innter circle to outside, unless you specify trackWidth and radius manually. To change default radius and trackWidth for all tracks, you simply put them in ggbio function.

- rule of thumb seqlengths, seqlevels and chromosomes names should be exactly the same.
- to use circle, you have to use ggbio constructor at the beginning instead of ggplot.

¹http://www.nature.com/ng/journal/v43/n10/full/ng.936.html

You can use autoplot to create single track easily like

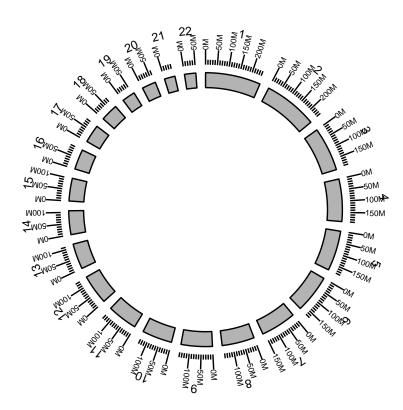
```
head(hg19sub)
## GRanges with 6 ranges and 0 metadata columns:
##
         segnames
                          ranges strand
##
            <Rle>
                       <IRanges> <Rle>
                1 [1, 249250621]
##
     [1]
##
     [2]
                2 [1, 243199373]
                3 [1, 198022430]
##
     [3]
##
     [4]
                4 [1, 191154276]
                                       *
               5 [1, 180915260]
##
     [5]
                                       *
              6 [1, 171115067]
##
     [6]
                                       *
##
     ___
##
     seqlengths:
##
              1
                        2
                                  3
                                             4 ...
                                                          20
                                                                    21
                                                                               22
##
      249250621 243199373 198022430 191154276 ... 63025520 48129895 51304566
autoplot(hg19sub, layout = "circle", fill = "gray70")
```



Hoever, the low level circle function leave you more flexibility to build circular plot one by one. Let's start to add tracks one by one.

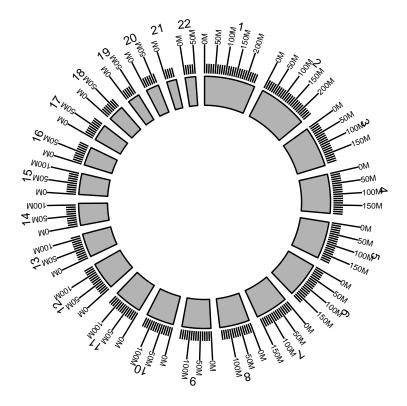
Let's use the same data to create ideogram, label and scale track, it layouts the circle by the order you created from inside to outside.

```
p <- ggbio() + circle(hg19sub, geom = "ideo", fill = "gray70") +
    circle(hg19sub, geom = "scale", size = 2) +
    circle(hg19sub, geom = "text", aes(label = seqnames), vjust = 0, size = 3)</pre>
```



To simply override the setting, you can do it globally in ggbio function or individually circle function by specifying parametters trackWidth and radius, you can also specify the global settin for buffer in between in ggbio like example below.

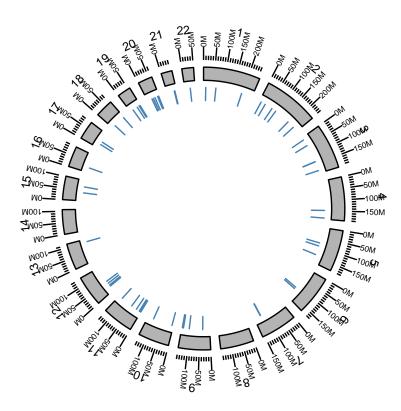
```
p <- ggbio(trackWidth = 10, buffer = 0, radius = 10) + circle(hg19sub, geom = "ideo", fill = "gray70") +
    circle(hg19sub, geom = "scale", size = 2) +
    circle(hg19sub, geom = "text", aes(label = seqnames), vjust = 0, size = 3)
p</pre>
```



Then we add a "rectangle" track to show somatic mutation, this will looks like vertical segments. head(mut.gr)

##	GRange	es with 6	ranges and	10 metadata	columns	5 .	
##	0	seqnames	0				Entrez_Gene_Id
##		<rle></rle>		<iranges></iranges>	<rle></rle>	<pre> < factor></pre>	<integer></integer>
##	[1]	1	[11003085	, 11003085]	+	TARDBP	23435
##	[2]	1	[62352395	, 62352395]	+	I INADL	10207
##	[3]	1	[194960885	, 194960885]	+	CFH	3075
##	[4]	2	[10116508	, 10116508]	-	CYS1	192668
##	[5]	2	[33617747	, 33617747]	+	RASGRP3	25780
##	[6]	2	[73894280	, 73894280]	+	C2orf78	388960
##		Center	NCBI_Build	Strand Va	riant_Cl	assification N	Variant_Type
##		<factor></factor>	<integer></integer>	<factor></factor>		<factor></factor>	<factor></factor>
##	[1]	Broad	36	+		Missense	SNP
##	[2]	Broad	36	+		Missense	SNP
##	[3]	Broad	36	+		Missense	SNP
##	[4]	Broad	36	-		Missense	SNP
##	[5]	Broad	36	+		Missense	SNP
##	[6]	Broad	36	+		Missense	SNP
##		Reference	e_Allele Tur	nor_Seq_Alle	le1 Tumo	or_Seq_Allele2	
##		•	<factor></factor>	<facto< td=""><td>or></td><td><factor></factor></td><td></td></facto<>	or>	<factor></factor>	
##	[1]		G		G	A	
##	[2]		Т		Т	G	
##	[3]		G		G	A	
##	[4]		С		С	Т	

[5] С С Т ## [6] Т Т С ## ## seqlengths: 2 20 ## 1 3 4 . . . 21 22 249250621 243199373 198022430 191154276 ... 63025520 48129895 ## 51304566 p <- ggbio() + circle(mut.gr, geom = "rect", color = "steelblue") +</pre> circle(hg19sub, geom = "ideo", fill = "gray70") + circle(hg19sub, geom = "scale", size = 2) + circle(hg19sub, geom = "text", aes(label = seqnames), vjust = 0, size = 3) р



Next, we need to add some "links" to show the rearrangement, of course, links can be used to map any kind of association between two or more different locations to indicate relationships like copies or fusions. To create a suitable structure to plot, please use another *GRanges* to represent the end of the links, and stored as elementMetadata for the "start point" *GRanges*. Here we named it as "to.gr" and will be used later.

str1

0

0

<integer>

CRC-4

class

> <factor>
1 long_range
1 long_range
0 long_range

deletion

inter_chr

he	ead(crc.g	r)				
##	# GRanges	with 6	ranges and	17 metada	ta columr	ns:
##	t s	eqnames		ranges	strand	individual
##	ŧ	<rle></rle>		<iranges></iranges>	<rle></rle>	<factor></factor>
##	ŧ [1]	18	[56258628,	56258628]	*	CRC-4
##	ŧ [2]	18	[44496014,	44496014]	*	CRC-4
##	ŧ [3]	18	[45023683,	45023683]	*	CRC-4
##	ŧ [4]	8	[52186319,	52186319]	*	CRC-4

8 [37328910, 37328910]

- (

[5]

##

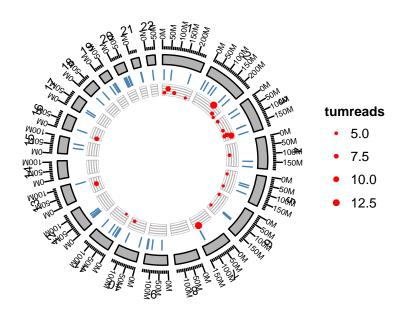
##	[6]	8	[35575394,	35575394]	*	CRC-	-4	0	inter_chr	
##		-		normreads	0	gene2				
##			-	<integer></integer>		<factor></factor>				
##	[1]	2104165	491	2	MC4R	ZCCHC2				
##	[2]	12947165	265	0	KIAA0427	CDH20				
##	[3]	13356670	238	0	DYM	ZCCHC2				
##	[4]	268	94	0	PXDNL	PXDNL				
##	[5]	<na></na>	56	0	ZNF703	PAK7				
##	[6]	<na></na>	53	0	UNC5D	RALGAPB				
##					site	1				
##					<factor< td=""><td>></td><td></td><td></td><td></td><td></td></factor<>	>				
##	[1]		IGR:	69Kb befor	re MC4R(-))				
##	[2]	Intron of	KIAA0427(·	+): 4Kb aft	ter exon 8	3				
##	[3]	Intron	of DYM(-)	: 14Kb afte	er exon 13	3				
##	[4]		IGR: 20	08Kb before	e PXDNL(-))				
##	[5]		IGR: 344	4Kb before	ZNF703(+))				
##	[6]	Intron	of UNC5D(·	+): 3Kb aft	ter exon 4	1				
##					sit	te2				
##					<facto< td=""><td>or></td><td></td><td></td><td></td><td></td></facto<>	or>				
##	[1]	Intron of	E ZCCHC2(+): 222bp be	efore exor	n 4				
##	[2]		IGR:	134Kb befo	ore CDH20	(+)				
##	[3]	Intron of	E ZCCHC2(+): 854bp be	efore exor	n 9				
##	[4]		IGR:	208Kb befo	ore PXDNL	(-)				
##	[5]	Intro	on of PAK7	(-): 11Kb a	after exor	n 4				
##	[6]	Intron of	RALGAPB(+): 839bp at	fter exon	15				
##					fusion	quality	score	В	Presult	
##				4	<factor> <</factor>	<numeric></numeric>	<numeric></numeric>	<i< td=""><td>nteger></td><td></td></i<>	nteger>	
##	[1]				-	1.0000	491.00		-1	
##	[2]				-	0.9944	263.52		1	
##		Protein fi	usion: in :	frame (ZCCI	HC2-DYM)	1.0000	238.00		1	
##	[4]				-	1.0000	94.00		-1	
##	[5]				-	0.9740	54.55		1	
##	[6]			Antisense	e fusion	1.0000	53.00		1	
##			validation	n_result			0	ear	rangements	
##				<factor></factor>			anges>		character>	
##				lidation 18						
##		not_subje	cted_to_va	lidation 18						
##	[3]						30361] int:			
##		not_subject	cted_to_va	lidation 8						
##	[5]								hromosomal	
##	[6]	not_subje	cted_to_va	lidation 20):*:[36598	5752, 3659	95752] int	erc	hromosomal	
##										
##	seq	lengths:								
##		1	2	3	4.		20	21	22	
##	249	9250621 243	3199373 198	8022430 193	1154276 .	630255	520 481298	895	51304566	

Here in this example, we use "intrachromosomal" to label rearrangement within the same chromosomes and use "interchromosomal" to label rearrangement in different chromosomes.

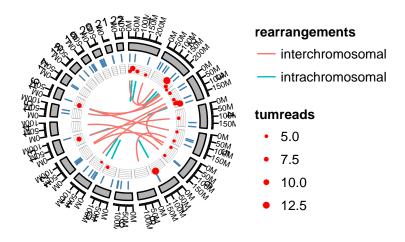
Get subset of links data for only one sample "CRC1"

gr.crc1 <- crc.gr[values(crc.gr)\$individual == "CRC-1"]</pre>

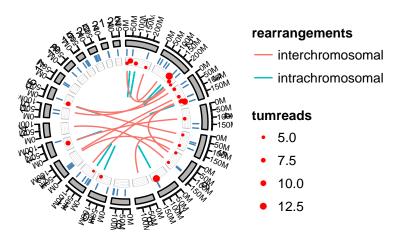
Ok, add a "point" track with grid background for rearrangement data and map 'y' to variable "score", map 'size' to variable "tumreads", rescale the size to a proper size range.



Finally, let's add links and map color to rearrangement types. Remember you need to specify 'linked.to' parameter to the column that contain end point of the data.



All those code could be simply constructed by following code



4.1.3 Complex arragnment of plots

In this step, we are going to make multiple sample comparison, this may require some knowledge about package grid and gridExtra. We will introduce a more easy way to combine your graphics later after this.

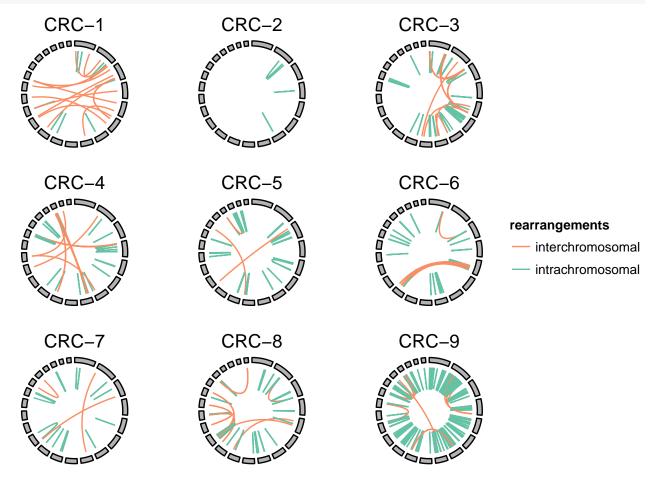
We just want 9 single circular plots put together in one page, since we cannot keep too many tracks, we only keep ideogram and links. Here is one sample.

```
grl <- split(crc.gr, values(crc.gr)$individual)</pre>
## need "unit", load grid
library(grid)
crc.lst <- lapply(grl, function(gr.cur){</pre>
  print(unique(as.character(values(gr.cur)$individual)))
  cols <- RColorBrewer::brewer.pal(3, "Set2")[2:1]</pre>
  names(cols) <- c("interchromosomal", "intrachromosomal")</pre>
  p <- ggbio() + circle(gr.cur, geom = "link", linked.to = "to.gr",</pre>
                          aes(color = rearrangements)) +
                   circle(hg19sub, geom = "ideo",
                          color = "gray70", fill = "gray70") +
                   scale_color_manual(values = cols) +
                   labs(title = (unique(values(gr.cur)$individual))) +
                   theme(plot.margin = unit(rep(0, 4), "lines"))
})
## [1] "CRC-1"
## [1] "CRC-2"
```

##	[1]	"CRC-3"
##	[1]	"CRC-4"
##	[1]	"CRC-5"
##	[1]	"CRC-6"
##	[1]	"CRC-7"
##	[1]	"CRC-8"
##	[1]	"CRC-9"

We wrap the function in grid level to a more user-friendly high level function, called arrangeGrobByParsingLegend. You can pass your ggplot2 graphics to this function, specify the legend you want to keep on the right, you can also specify the column/row numbers. Here we assume all plots we have passed follows the same color scale and have the same legend, so we only have to keep one legend on the right.

arrangeGrobByParsingLegend(crc.lst, widths = c(4, 1), legend.idx = 1, ncol = 3)



NULL

4.2 How to make grandlinear plots

4.2.1 Introduction

Let's use a subset of *PLINK* output (https://github.com/stephenturner/qqman/blob/master/plink.assoc. txt.gz) as our example test data.

```
snp <- read.table(system.file("extdata", "plink.assoc.sub.txt", package = "biovizBase"),</pre>
                                header = TRUE)
require(biovizBase)
gr.snp <- transformDfToGr(snp, seqnames = "CHR", start = "BP", width = 1)
head(gr.snp)
## GRanges with 6 ranges and 10 metadata columns:
##
                seqnames
                                                           ranges strand |
                                                                                                    CHR
                                                                                                                         SNP
                                                                                                                                             RP
##
                     <Rle>
                                                        <IRanges> <Rle> | <integer>
                                                                                                                <factor> <integer>
                            4 [ 10794096, 10794099]
##
         [1]
                                                                                 * |
                                                                                                      4 rs9291494 10794096
##
         [2]
                          14 [ 55853742, 55853755]
                                                                                   * |
                                                                                                     14 rs1152481 55853742
##
         [3]
                            6 [ 55188853, 55188858]
                                                                                   * |
                                                                                                      6 rs3134708 55188853
                          17 [ 4146033,
         [4]
                                                                                                     17 rs2325988
##
                                                        4146049]
                                                                                  * |
                                                                                                                                   4146033
                           19 [ 46089501, 46089519]
##
         [5]
                                                                                   * |
                                                                                                      19 rs8103444 46089501
                           1 [107051695, 107051695]
                                                                                                      1 rs12072065 107051695
##
         [6]
                                                                                  * |
##
                                          F_A F_U
                                                                                           CHISQ
                                                                                                                    Ρ
                           Α1
                                                                               A2
                                                                                                                                     OR.
##
                <factor> <numeric> <factor> <numeric> <factor> <numeric> <numeric>
                                                                          A
         [1]
                            G
                                     0.3061 0.1341
                                                                                          7.5070 0.006147
##
                                                                                                                              2.8480
##
         [2]
                            G
                                     0.3542 0.2805
                                                                              Α
                                                                                     1.1030 0.293600
                                                                                                                        1.4070
         [3]
                                 0.2500 0.2875
##
                            С
                                                                              A 0.3135 0.575500
                                                                                                                             0.8261
                            G 0.2551 0.2317
##
         [4]
                                                                               A 0.1323 0.716100
                                                                                                                          1.1360
                                     0.3980 0.2927
##
         [5]
                            С
                                                                              A 2.1750 0.140300
                                                                                                                              1.5970
                                                       0.0000
##
         [6]
                                     0.0000
                                                                                С
                                                                                              <NA>
                            0
                                                                                                                <NA>
                                                                                                                                  <NA>
##
         ____
##
         seqlengths:
           4 14 6 17 19 1 9 7 20 11 10 2 18 5 12 22 8 13 3 16 15 21
##
          ##
## change the segname order
require(GenomicRanges)
gr.snp <- keepSeqlevels(gr.snp, as.character(1:22))</pre>
seqlengths(gr.snp)
## 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22
## need to assign seqlengths
data(ideoCyto, package = "biovizBase")
seqlengths(gr.snp) <- as.numeric(seqlengths(ideoCyto$hg18)[1:22])</pre>
## remove missing
gr.snp <- gr.snp[!is.na(gr.snp$P)]</pre>
## transform pvalue
values(gr.snp)$pvalue <- -log10(values(gr.snp)$P)</pre>
head(gr.snp)
## GRanges with 6 ranges and 11 metadata columns:
##
                                                                                                                   SNP
                                                                                                                                       BP
                segnames
                                                         ranges strand |
                                                                                                 CHR
##
                      <Rle>
                                                    <IRanges> <Rle> | <integer> <factor> <integer>
##
         [1]
                            4 [10794096, 10794099]
                                                                               * |
                                                                                                    4 rs9291494 10794096
##
         [2]
                           14 [55853742, 55853755]
                                                                               * |
                                                                                                   14 rs1152481 55853742
##
         [3]
                            6 [55188853, 55188858]
                                                                               * |
                                                                                                   6 rs3134708 55188853
                        17 [ 4146033, 4146049]
                                                                               * |
##
         [4]
                                                                                                  17 rs2325988 4146033
```

ggbio:visualization toolkits for genomic data

##	Г О Т			46089519]	*) rs8103444	46089501
	[6]	9	[81517907,	81517915]	*	9) rs2591	81517907
##		A1	F_A	F_U	A2	CHISQ	Р	OR
##		<factor></factor>	<numeric></numeric>	<numeric></numeric>	<factor></factor>	<numeric></numeric>	<numeric> <</numeric>	<numeric></numeric>
##	[1]	G	0.3061	0.13410	A	7.5070	0.006147	2.8480
##	[2]	G	0.3542	0.28050	A	1.1030	0.293600	1.4070
##	[3]	С	0.2500	0.28750	A	0.3135	0.575500	0.8261
##	[4]	G	0.2551	0.23170	A	0.1323	0.716100	1.1360
##	[5]	С	0.3980	0.29270	A	2.1750	0.140300	1.5970
##	[6]	С	0.1042	0.04878	Т	1.8720	0.171200	2.2670
##		pvalue	ł					
##		<numeric></numeric>						
##	[1]	2.2113	5					
##	[2]	0.5322	1					
##	[3]	0.2400)					
##	[4]	0.1450)					
##	[5]	0.8529)					
##	[6]	0.7665						
##								
##	seql	lengths:						
##		1	2	3	4.	• •	20 2	21 22
##	247	249719 24	2951149 19	9501827 19	1273063 .	624359	64 4694432	49691432
## do	าท.ค							

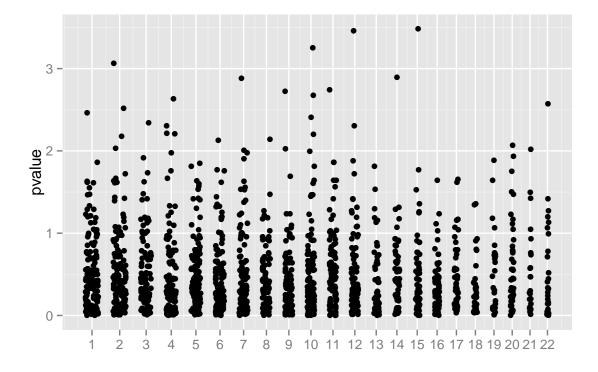
The data is ready, we need to pay attention

- if seqlengths is missing, we use data range, so the chromosome length is not accurate
- use seqlevel to control order of chromosome

4.2.2 Corrdinate genome

In autoplot, argument coord is just used to transform the data, after that, you can use it as common GRanges, all other geom/stat works for it.

autoplot(gr.snp, geom = "point", coord = "genome", aes(y = pvalue))



However, we recommend you to use more powerful function plotGrandLinear to generate manhattan plot introduced in next section.

4.2.3 Convenient plotGrandLinear function

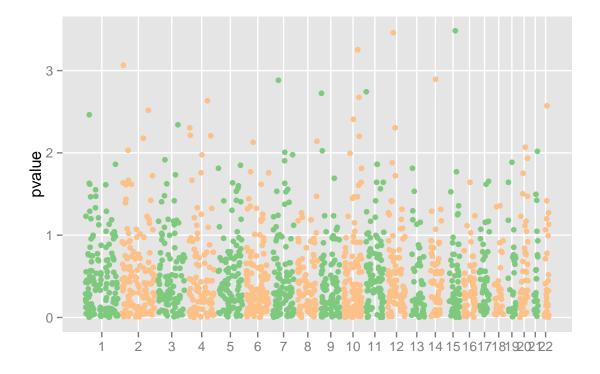
For *Manhattan plot*, we have a function called plotGrandLinear. aes(y =) is required to indicate the y value, e.g. p-value.

Color mapping is automatically figured out by ggbio following the rules

- if color present in aes(), like aes(color = seqnames), it will assume it's mapping to data column called 'seqnames'.
- if color is not wrapped in aes(), then this function will recylcle them to all chromosomes.
- if color is single character representing color, then just use one arbitrary color.

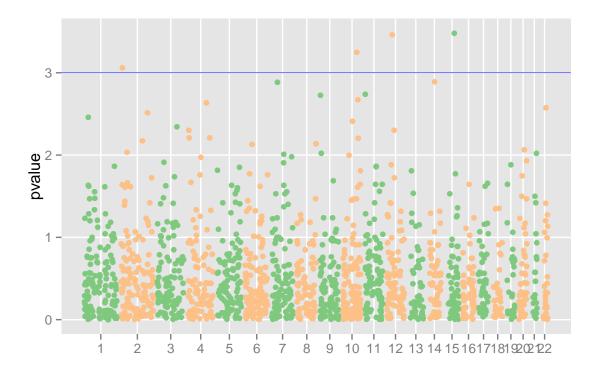
Let's test some examples for controling colors.

```
plotGrandLinear(gr.snp, aes(y = pvalue), color = c("#7fc97f", "#fdc086"))
```

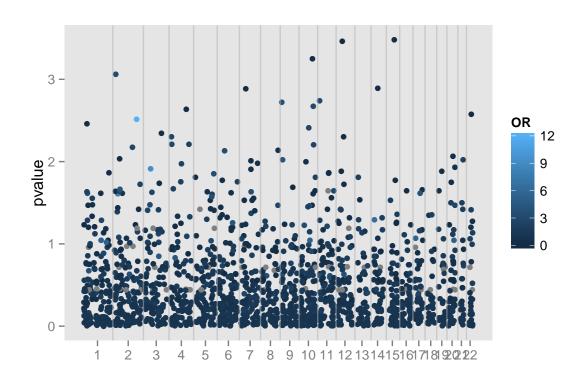


Let's add a cutoff line





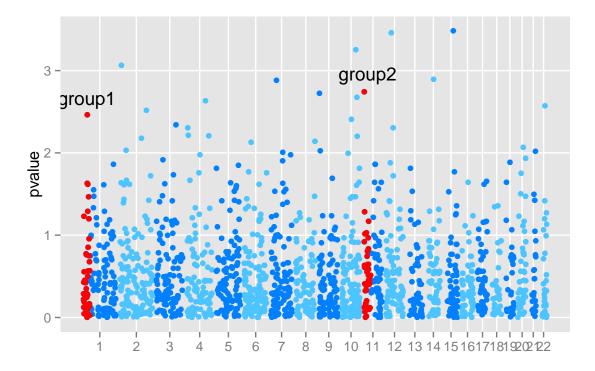
Sometimes you use color to mapping other varibles so you may need a different to separate chromosomes. plotGrandLinear(gr.snp, aes(y = pvalue, color = OR), spaceline = TRUE, legend = TRUE)



4.2.4 How to highlight some points?

You can provide a highlight *GRanges*, and each row highlights a set of overlaped snps, and labeled by rownames or certain columns, there is more control in the function as parameters, with prefix highlight.*, so you could control color, label size and color, etc.

```
gro <- GRanges(c("1", "11"), IRanges(c(100, 2e6), width = 5e7))
names(gro) <- c("group1", "group2")
plotGrandLinear(gr.snp, aes(y = pvalue), highlight.gr = gro)</pre>
```



4.3 How to make stacked karyogram overview plots

4.3.1 Introduction

A karyotype is the number and appearance of chromosomes in the nucleus of a eukaryotic cell². It's one kind of overview when we want to show distribution of certain events on the genome, for example, binding sites for certain protein, even compare them across samples as example shows in this section.

GRanges and Seqinfo objects are an ideal container for storing data needed for karyogram plot. Here is the strategy we used for generating ideogram templates.

- Althouth seqlengths is not required, it's highly recommended for plotting karyogram. If a GRanges object contains seqlengths, we know exactly how long each chromosome is, and will use this information to plot genome space, particularly we plot all levels included in it, **NOT JUST** data space.
- If a GRanges has no seqlengths, we will issue a warning and try to estimate the chromosome lengths from data included. This is **NOT** accurate most time, so please pay attention to what you are going to visualize and make sure set seqlengths before hand.

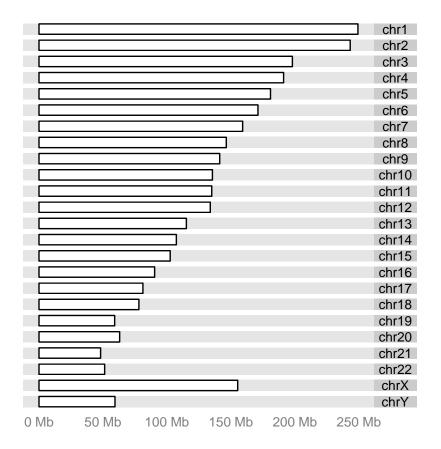
4.3.2 Create karyogram temlate

Let's first introduce how to use autoplot to generate karyogram graphic.

The most easy one is to just plot Seqinfo by using autoplot, if your *GRanges* object has seqinfo with seqlengths information. Then you add data layer later.

²http://en.wikipedia.org/wiki/Karyotype

```
data(ideoCyto, package = "biovizBase")
autoplot(seqinfo(ideoCyto$hg19), layout = "karyogram")
```



To show cytoband, your data need to have cytoband information, we stored some data for you, including hg19, hg18, mm10, mm9.

```
## turn on cytoband if it exists
biovizBase::isIdeogram(ideoCyto$hg19)
```

[1] TRUE

autoplot(ideoCyto\$hg19, layout = "karyogram", cytoband = TRUE)

chr1	
chr2	
chr3	
chr4	
chr5	gieStain
chr6	gicotain
chr7	acen
chr8	
chr9	gneg
chr10	gpos100
chr11	gpositoo
chr12	gpos25
chr13	
chr14	gpos50
chr15	an 225
chr16	gpos75
chr17	gvar
chr18	gvai
chr19	stalk
chr20	
chr21	
chr22	
chrX	
chrY	

0 Mb 50 Mb 100 Mb 150 Mb 200 Mb 250 Mb

To change order or only show a subset of the karyogram, you have to manipulate seqlevels, please check out manual for keepSeqlevels, seqlevels in *GenomicRanges* package for more information. Or you could read the example below.

4.3.3 Add data on karyogram layout

If you have single data set stored as *GRanges* to show on a karyogram layout, autoplot function is enough for you to plot the data on it.

We use a default data in package *biovizBase*, which is a subset of RNA editing set in human. The data involved in this GRanges is sparse, so we cannot simply use it to make karyogram template, otherwise, the estimated chromosome lengths will be very rough and inaccurate. So what we need to do first is to *add seglength information to this object*.

```
data(darned_hg19_subset500, package = "biovizBase")
dn <- darned_hg19_subset500
library(GenomicRanges)
seqlengths(dn)
##
   chr1 chr10 chr11 chr12 chr13 chr14 chr15 chr16 chr17 chr18 chr19 chr2 chr20
##
     NA
           NA
                 NA
                       NA
                             NA
                                 NA
                                       NA
                                               NA
                                                   NA
                                                         NA
                                                               NA
                                                                      NA
                                                                              NA
## chr21 chr22
               chr3 chr4
                           chr5 chr6 chr7
                                             chr8 chr9
                                                          chrX
##
     NA
           NA
                 NA
                       NA
                             NA
                                    NA
                                          NA
                                                NA
                                                      NA
                                                            NA
## add seqlengths
## we have seglegaths information in another data set
seqlengths(dn) <- seqlengths(ideoCyto$hg19)[names(seqlengths(dn))]</pre>
## then we change order
dn <- keepSeqlevels(dn, paste0("chr", c(1:22, "X")))</pre>
seqlengths(dn)
##
       chr1
                  chr2
                            chr3
                                      chr4
                                                chr5
                                                          chr6
                                                                    chr7
                                                                              chr8
## 249250621 243199373 198022430 191154276 180915260 171115067 159138663 146364022
```

chr10 chr12 chr15 chr9 chr11 chr13 chr14 chr16 ## 141213431 135534747 135006516 133851895 115169878 107349540 102531392 90354753 ## chr17 chr18 chr19 chr20 chr21 chr22 chrX 81195210 78077248 59128983 ## 63025520 48129895 51304566 155270560

autoplot(dn, layout = "karyogram")

	_
	chr1
	chr2
	chr3
	chr4
	chr5
	chr6
	chr7
	chr8
	chr9
	chr10
	chr11
	chr12
	chr13
	chr14
	chr15
	chr16
	chr17
	chr18
	chr19
	chr20
	chr21
	chr22
	chrX
0 Mb 50 Mb 100 Mb 150 Mb 200 Mb 25	0 Mb

Then we take one step further, the power of *ggplot2* or *ggbio* is the flexible multivariate data mapping ability in graphics, make data exploration much more convenient. In the following example, we are trying to map a categorical variable 'exReg' to color, this variable is included in the data, and have three levels, '3' indicate 3' utr, '5' means 5' utr and 'C' means coding region. We have some missing values indicated as NA, in default, it's going to be shown in gray color, and keep in mind, since the basic geom(geometric object) is rectangle, and genome space is very large, so change both color/fill color of the rectangle to specify both border and filled color is necessary to get the data shown as different color, otherwise if the region is too small, border color is going to override the fill color.

```
## since default is geom rectangle, even though it's looks like segment
## we still use both fill/color to map colors
autoplot(dn, layout = "karyogram", aes(color = exReg, fill = exReg))
```

chr1	
chr2	
chr3	
chr4	
chr5	
chr6	
chr7	
chr8	
chr9	-
chr10	exReg
chr11	3
chr12	5
chr13	
chr14	C
chr15	
chr16	
chr17	
chr18	
chr19	
chr20	
chr21	
chr22	
chrX	

0 Mb 50 Mb 100 Mb 150 Mb 200 Mb 250 Mb

Or you can set the missing value to particular color yo u want (NA values is not shown on the legend).

```
## since default is geom rectangle, even though it's looks like segment
## we still use both fill/color to map colors
autoplot(dn, layout = "karyogram", aes(color = exReg, fill = exReg), alpha = 0.5) +
scale_color_discrete(na.value = "brown")
```

chr1	
chr2	
chr3	
chr4	
chr5	
chr6	
chr7	
chr8	
chr9	avDag
chr10	exReg
chr11	- 3
chr12	5
chr13	
chr14	<u> </u>
chr15	
chr16	
chr17	
chr18	
chr19	
chr20	
chr21	
chr22	
chrX	

0 Mb 50 Mb 100 Mb 150 Mb 200 Mb 250 Mb

Well, sometimes we have too many values, we want to separate them by groups and show them at diffent height, below is a hack for that purpose and in next section, we will introduce a more flexible and general way to add data layer by layer.

Template chromosome y limits is [0, 10], that's why this hack works

4.3.4 Add more data using layout_karyogram function

In this section, a lower level function layout_karyogram is going to be introduced. This is convenient API for constructing karyogram plot and adding more data layer by layer. Function ggplot is just to create blank object to add layer on.

You need to pay attention to

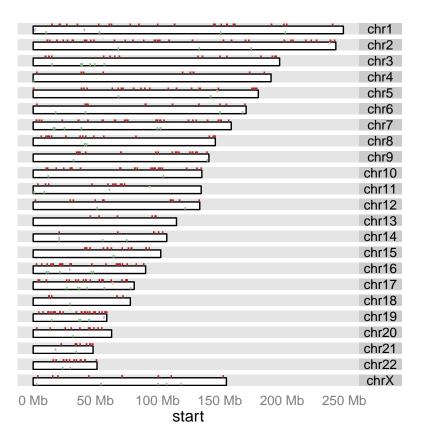
- when you add plots layer by layer, seqnames of different data must be the same to make sure the data are mapped to the same chromosome. For example, if you name chromosome following schema like *chr1* and use just number 1 to name other data, they will be treated as different chromosomes.
- cannot use the same aesthetics mapping multiple time for different data. For example, if you have used aes(color =), for one data, you cannot use aes(color =) anymore for mapping variables from other add-on data, this is

currently not allowed in *ggplot2*, even though you expect multiple color legend shows up, this is going to confuse people which is which. HOWEVER, color or fill without aes() wrap around, is allowed for any track, it's set single arbitrary color.

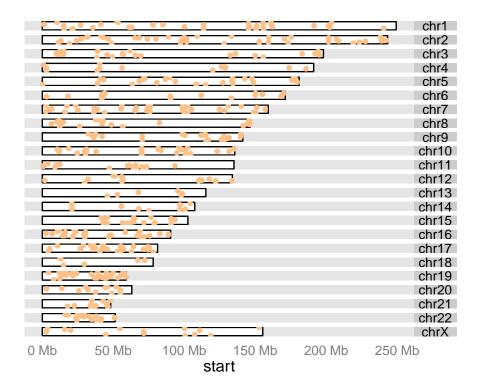
• Default rectangle y range is [0, 10], so when you add on more data layer by layer on existing graphics, you can use ylim to control how to normalize your data and plot it relative to chromosome space. For example, with default, chromosome space is plotted between y [0, 10], if you use ylim = c(10, 20), you will stack data right above each chromosomes and with equal width. For geom like 'point', which you need to specify 'y' value in aes(), we will add 5% margin on top and at bottom of that track.

Many times we overlay different datas sets, so let's break down the previous samples into 4 groups and treat them as different data and build them layer by layer, assign the color by hand. You could use ylim to control where they are ploted.

```
## prepare the data
dn3 <- dn.nona[dn.nona$exReg == '3']
dn5 <- dn.nona[dn.nona$exReg == '5']
dnC <- dn.nona[dn.nona$exReg == 'C']
dn.na <- dn[is.na(dn$exReg)]
## now we have 4 different data sets
autoplot(seqinfo(dn3), layout = "karyogram") +
layout_karyogram(data = dn3, geom = "rect", ylim = c(0, 10/3), color = "#7fc97f") +
layout_karyogram(data = dn5, geom = "rect", ylim = c(10/3, 10/3*2), color = "#beaed4") +
layout_karyogram(data = dnC, geom = "rect", ylim = c(10/3*2, 10), color = "#fdc086") +
layout_karyogram(data = dn.na, geom = "rect", ylim = c(10, 10/3*4), color = "brown")
```



What's more, you could even chagne the geom for those data



4.3.5 More flexible layout of karyogram

p.ylim + facet_wrap(~seqnames)

chr1	chr2	chr3	chr4	chr5	
chr6	chr7	chr8	chr9	chr10	
u u					ovPog
chr11	chr12	chr13	chr14	chr15	exReg 3
					5
chr16	chr17	chr18	chr19	chr20	C
chr21	chr22	chrX			

O 500005R0195130 ISONO05R0195130 ISONO05R0195131 IM b

Chapter 5

Link ranges to your data

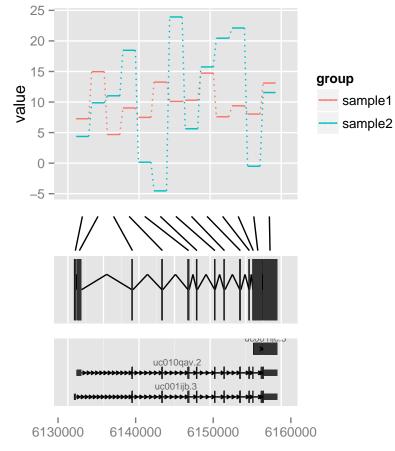
Plot GRanges object structure and linked to a even spaced paralell coordinates plot which represting the data in elementeMetadata.

```
library(TxDb.Hsapiens.UCSC.hg19.knownGene)
library(ggbio)
data(genesymbol, package = "biovizBase")
txdb <- TxDb.Hsapiens.UCSC.hg19.knownGene</pre>
model <- exonsBy(txdb, by = "tx")</pre>
model17 <- subsetByOverlaps(model, genesymbol["RBM17"])</pre>
exons <- exons(txdb)
exon17 <- subsetByOverlaps(exons, genesymbol["RBM17"])</pre>
## reduce to make sure there is no overlap
## just for example
exon.new <- reduce(exon17)</pre>
## suppose
values(exon.new)$sample1 <- rnorm(length(exon.new), 10, 3)</pre>
values(exon.new)$sample2 <- rnorm(length(exon.new), 10, 10)</pre>
values(exon.new)$score <- rnorm(length(exon.new))</pre>
values(exon.new)$significant <- sample(c(TRUE,FALSE), size = length(exon.new),replace = TRUE)</pre>
## data readu
exon.new
## GRanges with 13 ranges and 4 metadata columns:
       seqnames ranges strand |
<Rle> <IRanges> <Rle> |
                                                            sample1
##
##
                                                            <numeric>
##
       [1] chr10 [6130949, 6131156] + | 7.26895725483784
                                             + | 14.9766691947961
##
      [2] chr10 [6131309, 6131934]
      [3]chr10[6139011, 6139151]++4.6851558464563[4]chr10[6143234, 6143350]++9.02997654442108[5]chr10[6146894, 6147060]++7.48731547091016
##
##
##
##
      . . .
             . . .
                                     . . .
                                            . . . . . .
                                                                   . . .
           chr10 [6154173, 6154324]
                                          + | 14.7316906767153
##
      [9]
##
     [10] chr10 [6155471, 6155544]
                                             + | 7.58742745452715
##
     [11] chr10 [6156012, 6156110]
                                             + | 9.38550268352287
     [12] chr10 [6156126, 6157274]
                                             + | 8.03439235975778
##
##
     [13] chr10 [6157416, 6159422]
                                             + | 13.1087718583769
                    sample2 score significant
<numeric> <numeric> <logical>
##
##
      [1] 4.36852305658921 -2.96791738482802
##
```

##	[2]	9.87036450259352	0.101747387012454	0	
##	[3]	11.0197811445135	-0.597429436326523	0	
##	[4]	18.4654156968838	1.11616833300151	1	
##	[5]	0.155813603844239	0.487526566098763	1	
##					
##	[9]	15.7429658816936	0.721778444658207	1	
##	[10]	20.4460170390669	-1.65781546925978	1	
##	[11]	22.1135443543151	-1.81953389443168	0	
##	[12]	-0.503535023101866	1.91602272974415	1	
##	[13]	11.5534416861042	0.468713318813882	1	
##					
##	seal	engths:			
##	1	chr1	chr2	chrUn_g1000249	
##		249250621	243199373	38502	

Make the plots, you can pass a list of annotation tracks too.

```
p17 <- autoplot(txdb, genesymbol["RBM17"])
plotRangesLinkedToData(exon.new, stat.y = c("sample1", "sample2"), annotation = list(p17))</pre>
```



For more information, check the manual.

Chapter 6

Miscellaneous

Every plot object produced by *ggplot2* is essentially a *ggplot2* object, so you could use all the tricks you know with *ggplot2* on *ggbio* plots too, including scales, colors, themes, etc.

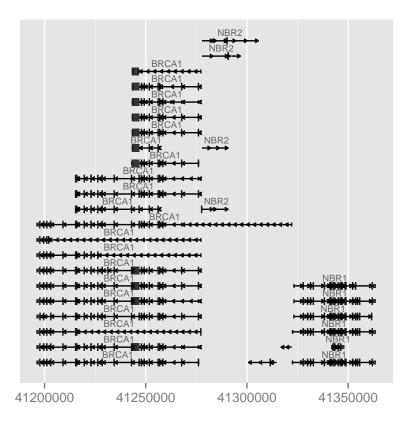
6.1 Themes

In ggbio, we developed some more themes to make things easier.

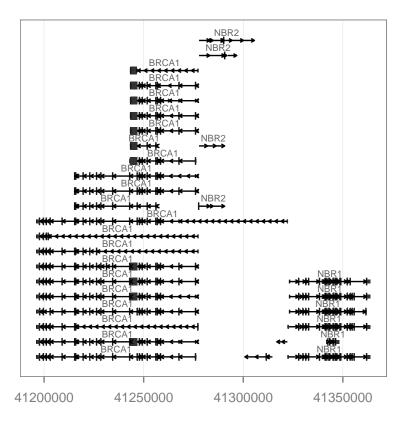
6.1.1 Plot theme

Plot level themes are like any other themes defined in *ggplot2*, simply apply it to a plot.

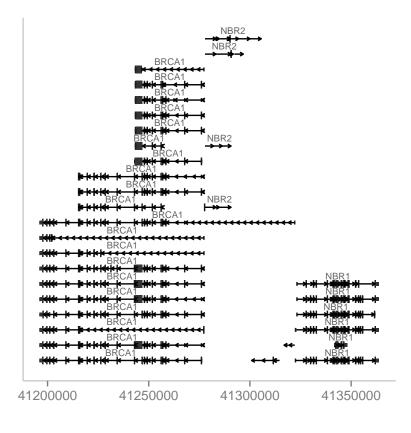
p.txdb



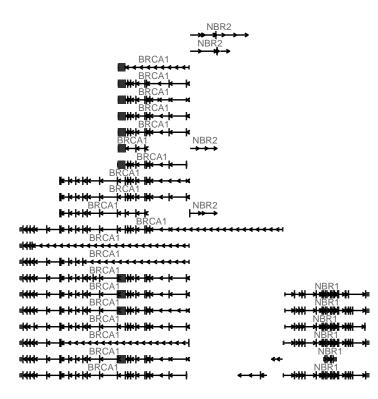
p.txdb + theme_alignment()



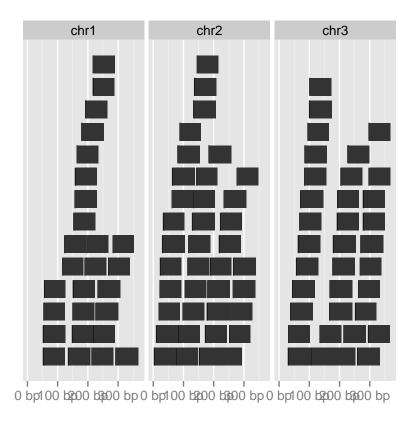
p.txdb + theme_clear()



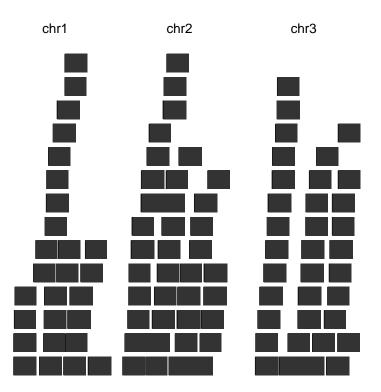
p.txdb + theme_null()



When you have multiple chromosomes encoded in seqnames, you could use theme_genome to make a 'fake' linear view of genome coordinates quickly by applying this theme, because it's not equal to chromosome lengths, it's simply



autoplot(gr) + theme_genome()



6.1.2 Track theme

Track level themes are more complex, it controls whole looking of the tracks, it's essentially a theme object with some attributes controlling the tracks appearance.

See how we make a template, you could customize in the same way

```
theme_tracks_sunset
## function (bg = "#fffedb", alpha = 1, ...)
## {
##
       res <- theme_clear(grid.x.major = FALSE, ...)</pre>
##
       attr(res, "track.plot.color") <- sapply(bg, scales::alpha,</pre>
##
            alpha)
##
       attr(res, "track.bg.color") <- bg</pre>
       attr(res, "label.text.color") <- "white"</pre>
##
       attr(res, "label.bg.fill") <- "#a52a2a"</pre>
##
##
       res
## }
## <environment: namespace:ggbio>
```

The attributes you could control is basically passed to tracks() constructor, including

character
character
character
numeric
characterORNULL
characterORNULL
unit

Table 6.1: tracks attributes

Chapter 7

Session Information

sessionInfo()

```
## R version 3.1.1 (2014-07-10)
## 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=en_US.UTF-8
                                 LC_NAME=C
## [9] LC_ADDRESS=C
                                  LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
                parallel stats graphics grDevices utils
## [1] grid
                                                                  datasets
## [8] methods
                base
##
## other attached packages:
## [1] VariantAnnotation_1.10.5
## [2] Rsamtools 1.16.1
## [3] BSgenome.Hsapiens.UCSC.hg19_1.3.1000
## [4] BSgenome_1.32.0
## [5] Biostrings_2.32.1
## [6] biovizBase_1.12.3
## [7] XVector_0.4.0
## [8] Homo.sapiens_1.1.2
## [9] TxDb.Hsapiens.UCSC.hg19.knownGene_2.14.0
## [10] org.Hs.eg.db_2.14.0
## [11] GO.db_2.14.0
## [12] RSQLite_0.11.4
## [13] DBI_0.2-7
## [14] OrganismDbi_1.6.0
## [15] GenomicFeatures_1.16.2
## [16] AnnotationDbi_1.26.0
## [17] Biobase_2.24.0
## [18] GenomicRanges_1.16.4
## [19] GenomeInfoDb_1.0.2
## [20] IRanges_1.22.10
```

```
## [21] ggbio_1.12.10
## [22] ggplot2_1.0.0
## [23] BiocGenerics_0.10.0
## [24] knitr_1.6
##
## loaded via a namespace (and not attached):
                       BatchJobs_1.3
## [1] BBmisc_1.7
                                                    BiocParallel_0.6.1
## [4] BiocStyle_1.2.0
                            Formula_1.1-2
                                                    GenomicAlignments_1.0.6
                                                    RBGL_1.40.1
## [7] Hmisc_3.14-4
                            MASS_7.3-34
## [10] RColorBrewer_1.0-5
                            RCurl_1.95-4.3
                                                    Rcpp_0.11.2
## [13] XML_3.98-1.1
                             biomaRt_2.20.0
                                                    bitops_1.0-6
## [16] brew_1.0-6
                             checkmate_1.3
                                                    cluster_1.15.2
## [19] codetools_0.2-9
                            colorspace_1.2-4
                                                    dichromat_2.0-0
## [22] digest_0.6.4
                                                    fail_1.2
                             evaluate_0.5.5
## [25] foreach_1.4.2
                            formatR_1.0
                                                    graph_1.42.0
## [28] gridExtra_0.9.1
                            gtable_0.1.2
                                                   highr_0.3
## [31] iterators_1.0.7
                             labeling_0.3
                                                    lattice_0.20-29
## [34] latticeExtra_0.6-26
                             munsell_0.4.2
                                                    plyr_1.8.1
## [37] proto_0.3-10
                             reshape2_1.4
                                                    rtracklayer_1.24.2
## [40] scales_0.2.4
                             sendmailR_1.1-2
                                                    splines_3.1.1
## [43] stats4_3.1.1
                             stringr_0.6.2
                                                    survival_2.37-7
## [46] tools_3.1.1
                             zlibbioc_1.10.0
```