Exploring data from the 1000 Genomes project in Bioconductor's *ind1KG* package

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Contents

1 Overview

In this document we will look at high-coverage NGS data obtained on NA19240, because we have the HapMap phase II genotypes (4 mm SNP) for this individual in GGtools/hmyriB36, and we have an affy 6.0 SNP CEL file for this individual (and her cohort) as well.

There are three main objectives discussed in this document.

- We describe how data published in the 1000 genomes (1KG) project can be imported for investigations using R. This involves the use of the *Rsamtools* package. We provide serialized instances of various relevant data elements so that large objects distributed from the project need not be redistributed for these illustrations.

- We describe how information on variants can be related to existing annotation using *rtracklayer* to check for events in regulatory regions, for example.
- We discuss how information in the samtools 'pileup' format can be checked from a statistical perspective to explore how 'known' variants in the sample compare to the putatively newly discovered variants.

The reads examined in the document are all from the Illumina sequencing platform; additional work is sketched facilitating comparison with (released) read libraries based on 454 or ABI platforms.

2 External data acquisition

2.1 Manual extraction of a multi-Mb chunk

We will focus on this individual's chromosome 6. We acquired

```
NA19240.chrom6.SLX.maq.SRP000032.2009_07.bam
```

and the associated bai and bas files from

```
ftp://ftp-trace.ncbi.nih.gov/1000genomes/ftp/pilot_data/data/
     NA19240/alignment/NA19240.chrom6.SLX.maq.SRP000032.2009_07.bam.bas
```

Note that it is possible to work with these files remotely in R, without moving them to the local machine, thanks to the remote access facilities built in to samtools and exposed in R.

We use

```
samtools view \
    NA19240.chrom6.SLX.maq.SRP000032.2009_07.bam |head -3000000 > na19240_3M.sam
```

to obtain a parsable text file, presumably of 3 million reads that aligned, using MAQ, nearest the 5' end of the p arm of chr6. This is because we expect the bam file to be sorted. We picked the number 3 million out of thin air.

This sam format file can be converted to bam format using the samtools import facility. We took chromosome 6 reference sequence from

```
ftp://ftp-trace.ncbi.nih.gov/1000genomes/ftp/technical/
                         reference/human_b36_female.fa.gz
```

and indexed it and used

```
samtools import femchr6.fa.fai na19240_3M.sam na19240_3M.bam
```

to generate the bam file.

We imported this into R using Bioconductor's Rsamtools with a straight application of `scanBam`. The result is saved in the package as `n240`.

```
> library(ind1KG)
> if (!exists("n240")) data(n240)
```

This is a list of lists, and we check on the contents of the elements as follows:

```
> names(n240[[1]])
```

```
 [1] "qname"  "flag"   "rname"  "strand" "pos"    "width"  "mapq"   "cigar"
 [9] "mrnm"   "mpos"   "isize"  "seq"    "qual"
```

We check the classes:

```
> sapply(n240[[1]], class)
```

```
        qname          flag         rname        strand           pos
  "character"     "integer"      "factor"      "factor"     "integer"
        width          mapq         cigar          mrnm          mpos
    "integer"     "integer"       "Cigar"      "factor"     "integer"
        isize           seq          qual
    "integer" "DNAStringSet" "PhredQuality"
```

We get a small number of exemplars from each element:

```
> lapply(n240[[1]], "[", 1:5)
```

```
$qname
[1] "EAS254_13:7:88:1639:15041"    "EAS139_43:2:31:1128:9551"
[3] "EAS254_13:8:68:520:6861"      "BGI-FC20AHFAAXX_6_26_477:352"
[5] "EAS139_43:6:71:1575:10961"

$flag
[1] 35 35 35 16 35

$rname
[1] 6 6 6 6 6
Levels: 6

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

```
$pos
[1] 5001 5002 5004 5004 5005

$width
[1] 51 51 51 36 51

$mapq
[1] 0 0 0 0 0

$cigar
class: Cigar
cigars (5): 51M 51M 51M 36M 51M

$mrnm
[1] 6    6    6    <NA> 6
Levels: 6

$mpos
[1] 5163 5203 5170   NA 5156

$isize
[1] 214 253 218  NA 203

$seq
  A DNAStringSet instance of length 5
    width seq
[1]    51 GATCTTATATAACTGTGAGATTAATCTCAGATAATGACACAAAATATAGTG
[2]    51 ATCTTATATAACTGTGAGATTAATCTCAGATAATGACACAAAATATAGTGA
[3]    51 CTTATATAACTGTGAGATTAATCTCAGATAATGACACAAAATATAGTGAAG
[4]    36 TGTCATTATCTGAGATTAATCTCACAGTTATATAAG
[5]    51 GTATATAACTGTGAGATTAATCTCAGATAATGACACAAAATATAGTGAAGT

$qual
  A PhredQuality instance of length 5
    width seq
[1]    51 C<@?AB?A@B@A?C@B@?@?BC@BB@C@A??B@AB<??@?AABAA?@>=??
[2]    51 B???A@A@A@A?;??@?@>ACABAAC@;@=BABB@=@A@?A?>A?A?<@9>
[3]    51 B@@=A@A@@@B@A@=@?AB<AB@B@@@>A@AB@>@=?@A:AB@A??@>;?>
[4]    36 C=:>A=>>==A=8?7>@?@=:@>?8;>9?8>9><60
[5]    51 +?>@?A?A?B@A????BC@ABACA???B@BB@=?A@ABAAB@B@?A?=?=A
```

We can use R at this point to do matching to reference and filtering and so forth, but we will only do this in a *post mortem* fashion, as it seems to make more sense to use samtools directly to do, for example, SNP calling.

2.2 Programmatic extraction of annotated regions

(This code segment suggested by Martin Morgan.)

We can use the *GenomicFeatures* package to obtain intervals defining various genomic elements.

```
> library(GenomicFeatures)
> library(GenomicFeatures.Hsapiens.UCSC.hg18)
> library(IRanges)
> genes = geneHuman()
> genes[1:2, ]

        name chrom strand txStart txEnd cdsStart cdsEnd exonCount
1 uc001aaa.2  chr1      +    1116  4121     1116   1115         3
2 uc009vip.1  chr1      +    1116  4272     1116   1115         2
      exonStarts       exonEnds proteinID    alignID
1 1116,2476,3084 2090,2584,4121      <NA> uc001aaa.2
2      1116,2476      2090,4272      <NA> uc009vip.1
```

The `transcripts` method will obtain ranges of transcripts with constraints.

```
> tx6 <- transcripts(genes, proximal = 300)
> chr6a <- ranges(tx6)[["chr6"]][1:50]
> chr6a

IRanges of length 50
       start     end  width
[1]   237101  296355  59255
[2]   249628  296353  46726
[3]   336752  347637  10886
[4]   336752  356443  19692
[5]   342056  347637   5582
[6]   430138  638109 207972
[7]   600939  601964   1026
[8]   654105  656405   2301
[9]   720456  725214   4759
...      ...     ...    ...
[42] 2945066 2964993  19928
[43] 2990617 2991033    417
[44] 3009121 3060420  51300
[45] 3021997 3060420  38424
```

```
[46] 3064041 3098281  34241
[47] 3098901 3101481   2581
[48] 3098901 3102782   3882
[49] 3128053 3141000  12948
[50] 3169514 3172870   3357
```

With a local BAM file, the following counting procedure is quick. Note that `fl` could be a URL beginning

```
ftp://ftp-trace.ncbi.nih.gov/1000genomes/ftp/pilot_data/data/NA19240/alignment...
```

and the computations would work, but completion speed would depend upon server load and network throughput.

```
> library(Rsamtools)
> p1 <- ScanBamParam(which = RangesList(`6` = chr6a))
> fl = "/mnt/data/stvjc/1000GENOMES/NA19240.chrom6.SLX.maq.SRP000032.2009_07.bam"
> unix.time(cnt <- countBam(fl, param = p1))
> sum(cnt$records)
```

The following scan will yield a list with read and quality information on the 50 transcript regions requested in `chr6a` allocated to 50 list elements.

```
> res <- scanBam(fl, param = p1)
> length(res)
> names(res[[1]])
```

3 Exploring a samtools pileup

The pileup output derived from the 3 million reads is a 17GB (sic) text file derived as follows:

```
samtools pileup -cf femchr6.fa \
      NA19240.chrom6.SLX.maq.SRP000032.2009_07.bam > na19240F.pup
```

First 10 lines:

```
6       5001    G       G       4       0       0       1       ^!.     C
6       5002    A       A       7       0       0       2       .^!.    <B
6       5003    T       T       7       0       0       2       ..      @?
6       5004    C       C       14      0       0       4       ..^!.^!,        ??B0
6       5005    T       T       4       0       0       5       ...,^!G A?@6+
6       5006    T       T       15      0       0       5       ...,.   BA@<?
6       5007    A       A       15      0       0       5       ...,.   ?@=>>
6       5008    T       T       15      0       0       5       ...,.   AAA9@
6       5009    A       A       15      0       0       5       ...,.   @@@>?
6       5010    T       T       17      0       0       6       ...,.^!.        BAA8AC
```

The total number of lines is not quite 200 million, so it might be handled directly in R on a reasonably sized machine. We have isolated the first 10 million records and restricted attention to those locations where the individual NA19240 differs from the reference sequence.

```
> data(pup240_disc)
> pup240_disc[1:5, ]
```

```
        loc ref indiv depth                          pileup
7961  12961   C     G     4                               g
9074  14074   G     T     4                              T$
11171 16172   C     A     2                           ,$.$a
23462 28466   A     C     6                              C,
28697 33701   A     G    21 ,,,,$,,,,.,,,,,.,..,..g.,,gGggggGgggg
```

Some of these variants are denoted with asterisk, suggesting evidence of deletion. We will omit these for now. There are also some non-nucleotide-valued markers, omitted.

```
> pup240_disc = pup240_disc[pup240_disc$ref != "*", ]
> pup240_disc = pup240_disc[pup240_disc$ref %in% c("A", "C", "T",
+     "G"), ]
> table(pup240_disc$indiv)
```

```
   A    C    G    K    M    R    S    T    W    Y 
1593 1926 1861 1039  958 4247 1068 1593  982 4233 
```

How many of the calls that disagree with reference are present at locations not already identified as polymorphic by dbSNP?

```
> data(c6snp)
> sum(!(pup240_disc$loc %in% c6snp$chrPosFrom))
```

```
[1] 4075
```

How many of these possibly novel variants are sites of heterozygosity?

```
> nov = pup240_disc[!(pup240_disc$loc %in% c6snp$chrPosFrom), ]
> table(nov$indiv)
```

```
  A   C   G   K   M   R   S   T   W   Y 
251 247 193 309 264 985 279 238 330 979 
```

4 Checking samtools-based calls against other calls

4.1 HapMap Phase II calls

We include information from the phase II HapMap calls for NA19240. We have a `snp.matrix` instance with the full genotyping for chromosome 6 and location information as supplied by HapMap.

```
> data(yri240_6)
> names(yri240_6)

[1] "hm2"  "supp"
```

The following code gets all relevant HapMap calls in a generic format and isolates the SNP at which NA19240 is heterozygous.

```
> snps = as(yri240_6[[1]], "character")
> hets = which(snps == "A/B")
> rshet = colnames(snps)[hets]
> smet = yri240_6[[2]]
> smethet = smet[hets, ]
> smethet[1:5, ]

           dbSNPalleles Assignment Chromosome Position Strand
rs12192290          A/T        A/T       chr6    95272      +
rs1929630           A/C        A/C       chr6    99536      +
rs719065            A/G        A/G       chr6   110632      +
rs12209455          A/G        A/G       chr6   112510      +
rs6909153           A/G        A/G       chr6   119769      +
```

We also have the full pileup information for the first 500K loci computed by samtools pileup.

```
> data(pup240_500k)
> pup240_500k[1:2, ]

    V2 V3 V4 V5   V9
1 5001  G  G  4  ^!.
2 5002  A  A  7 .^!.
```

This include some duplicated locations, which we remove.

```
> pup240_500ku = pup240_500k[!duplicated(pup240_500k[, 1]), ]
```

The pileups at which HapMap says our subject is heterozygous are

```
> hpup = pup240_500ku[pup240_500ku[, 1] %in% smethet[, "Position"],
+     ]
```

Are there any loci (in this very small region of chromosome 6) that HapMap says are heterozygous, but that are found to be homozygous by sequencing?

```
> hom = hpup[hpup[, 2] == hpup[, 3], ]
> hom

           V2 V3 V4 V5
158570 163386  C  C 15
169883 174667  T  T 89
219091 223773  C  C  4
                                                                             V9
158570                                         ,,,....,,t.,..,t..T.TtTa..,^~,^~T
169883             ..,,,$.$,,.,..,,...,..C,....,CAcc,,C,,,C,.C..,..,Cc,..^<,^?,
219091 ,.t..,,,,$.Gt,,tt,,t,.,,..,,t..,.,g.A,,.,tt,,..,,.T,t.,,,.,Tt...,.gt.^,g

> smethet[smethet[, "Position"] %in% hom[, 1], ]

          dbSNPalleles Assignment Chromosome Position Strand
rs1418703          C/T        C/T       chr6   163386      +
rs6915606          C/T        C/T       chr6   174667      +
rs815571           C/T        C/T       chr6   223773      +
```

4.2 Affy SNP 6.0 chip calls

We ran `crlmm` to genotype all 90 YRI samples from 6.0 chips distributed by Affymetrix. The data for NA19240 chromosome 6 are available in the ind1KG package:

```
> data(gw6c6.snp240)
> gw6c6.snp240[1:4, ]

    man_fsetid dbsnp_rs_id physical_pos strand allele_a allele_b call240
1 SNP_A-1984753    rs719065       110632      1        C        T       2
2 SNP_A-1984758   rs6927090       197145      1        A        C       3
3 SNP_A-1984759    rs815583       230695      1        C        T       2
4 SNP_A-1984760   rs1514346       334630      0        A        G       2
```

The heterozygous loci are

```
> hloc6 = gw6c6.snp240[gw6c6.snp240$call240 == 2, "physical_pos"]
```

Let's see if the sequencing leads to the same decisions (at least with regard to heterozygous vs. homozygous):

```
> inds = which(pup240_500k[, 1] %in% hloc6)
> table(pup240_500k[inds, 3])

 K  M  R  S  Y
 6  2 17  2 14
```

For the loci called homozygous by crlmm, we have:

```
> oloc6 = gw6c6.snp240[gw6c6.snp240$call240 %in% c(1, 3), "physical_pos"]
> oinds = which(pup240_500k[, 1] %in% oloc6)
> table(pup240_500k[oinds, 3])

 A  C  G  T  Y
21 49 38 22  1
```

5 Relating possibly novel variants to existing annotation

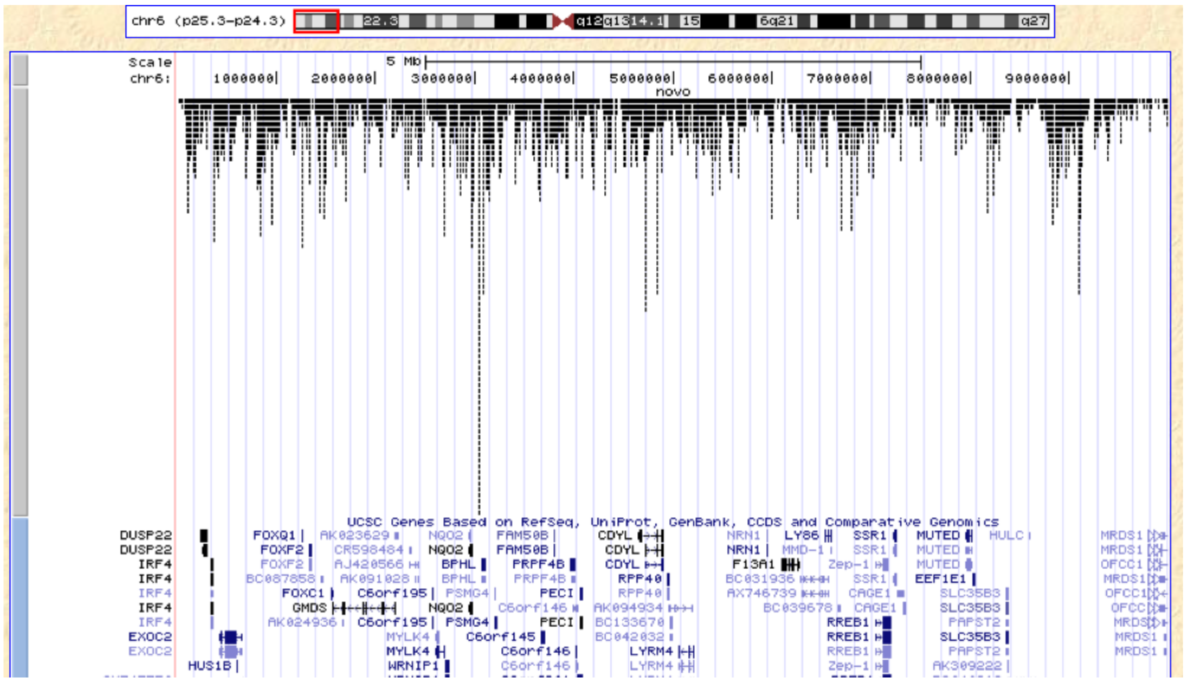
5.1 Browser-based visualization

There are many ways to use Bioconductor annotation resources to learn about contexts of variants. However, the UCSC genome browser is probably the most efficient place to start. We can convert our vector of locations of apparently new variants to a browser track as follows; this code is not executed in vignette construction, but you may run it manually if suitably networked.

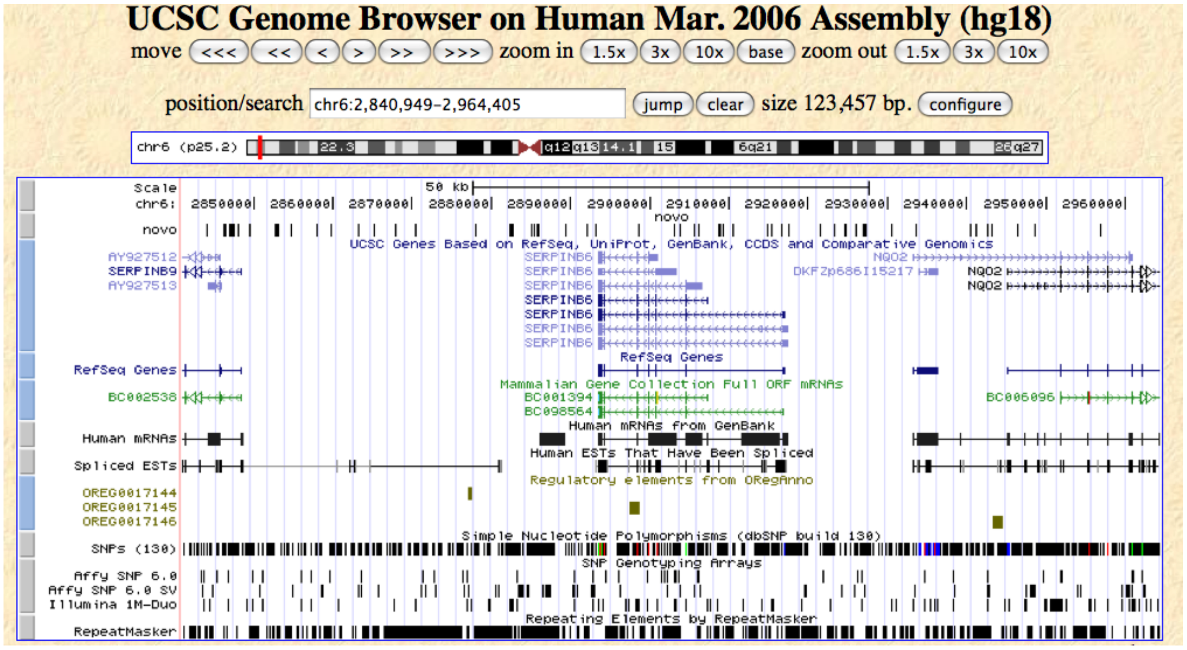
```
> library(IRanges)
> nloc = nov$loc
> nrd = RangedData(IRanges(nloc, nloc))
> names(nrd) = "chr6"
> library(rtracklayer)
> br = browserSession("UCSC")
> br[["novo"]] = nrd
> v1 = browserView(br, GenomicRanges(1, 1e+07, "chr6"))
```

This arranges the browser so that the custom track at the top of the display, 'novo', gives the locations of the possible novel variants.

Overall, we see that these novel variants occur regularly across the 10MB.


chr6 (p25.3-p24.3)
Scale
5 Mb
chr6:
1000000
2000000
3000000
4000000
5000000
6000000
7000000
8000000
9000000
novo
UCSC Genes Based on RefSeq, UniProt, GenBank, CCDS and Comparative Genomics
DUSP22
IRF4
EXOC2
HUS1B
FOXQ1
FOXF2
FOXC1
GMDS
NQO2
BPHL
MYLK4
WRNIP1
C6orf195
PSMG4
FAM50B
PRPF4B
PECI
C6orf145
C6orf146
CDYL
RPP40
LYRM4
NRN1
F13A1
LY86
SSR1
RREB1
CAGE1
MUTED
EEF1E1
SLC35B3
PAPST2
MRDS1
OFCC1
HULC

We can zoom in to the region around a given gene, here SERPINEB6.


UCSC Genome Browser on Human Mar. 2006 Assembly (hg18)
move <<< << < > >> >>> zoom in 1.5x 3x 10x base zoom out 1.5x 3x 10x
position/search chr6:2,840,949-2,964,405 jump clear size 123,457 bp. configure
chr6 (p25.2)
Scale
chr6:
novo
UCSC Genes Based on RefSeq, UniProt, GenBank, CCDS and Comparative Genomics
SERPINB9
SERPINB6
NQO2
RefSeq Genes
Mammalian Gene Collection Full ORF mRNAs
Human mRNAs from GenBank
Human ESTs That Have Been Spliced
Regulatory elements from ORegAnno
Simple Nucleotide Polymorphisms (dbSNP build 130)
SNP Genotyping Arrays
Affy SNP 6.0
Affy SNP 6.0 SV
Illumina 1M-Duo
Repeating Elements by RepeatMasker
RepeatMasker

5.2 Browser-based annotation extraction and comparison

Because the rtracklayer gives a bidirectional interface, it is possible to programmatically check for coincidence of variant locations, gene regions, or regulatory elements, for example.

We can learn the names of all available tracks for the current session via code like the following.

```
> tn = trackNames(br)
> grep("Genes", names(tn), value=TRUE) # many different gene sets
> tn["UCSC Genes"]    # resolve indirection
```

For example, to get the symbols for genes in the 10 million bp excerpt that we are working with, we can use

```
> rsg = track(br, "refGene")
> rsgdf = as.data.frame(rsg)
```

This data frame has been serialized with the ind1KG package.

```
> data(rsgdf)
> names(rsgdf)
```

```
 [1] "space"        "start"        "end"          "width"        "name"
 [6] "score"        "strand"       "thickStart"   "thickEnd"     "color"
[11] "blockCount"   "blockSizes"   "blockStarts"
```

```
> rsgdf[1:3, 1:7]

  space  start    end  width      name score strand
1  chr6 237101 296355  59255 NM_020185     0      +
2  chr6 336752 356443  19692 NM_002460     0      +
3  chr6 430138 638109 207972 NM_018303     0      -
```

We see that the 'names' here are RefSeq identifiers. We may be able to resolve them to Entrez Gene Ids, and thence to symbols, as follows:

```
> library(org.Hs.eg.db)
> rsgn = as.character(rsgdf$name)
> eid = mget(rsgn, revmap(org.Hs.egREFSEQ), ifnotfound = NA)
> eid = na.omit(unlist(eid))
> sym = mget(eid, org.Hs.egSYMBOL, ifnotfound = NA)
> unlist(sym)[1:10]

      56940        3662       55770      135458      285768      285768
   "DUSP22"      "IRF4"     "EXOC2"     "HUS1B" "LOC285768" "LOC285768"
      94234        2295        2296        2762
    "FOXQ1"     "FOXF2"     "FOXC1"      "GMDS"
```

These names are consistent with what we see on the browser displays shown above.

We can use IRanges infrastructure to check for intersection between novel variant locations and gene occupancy regions.

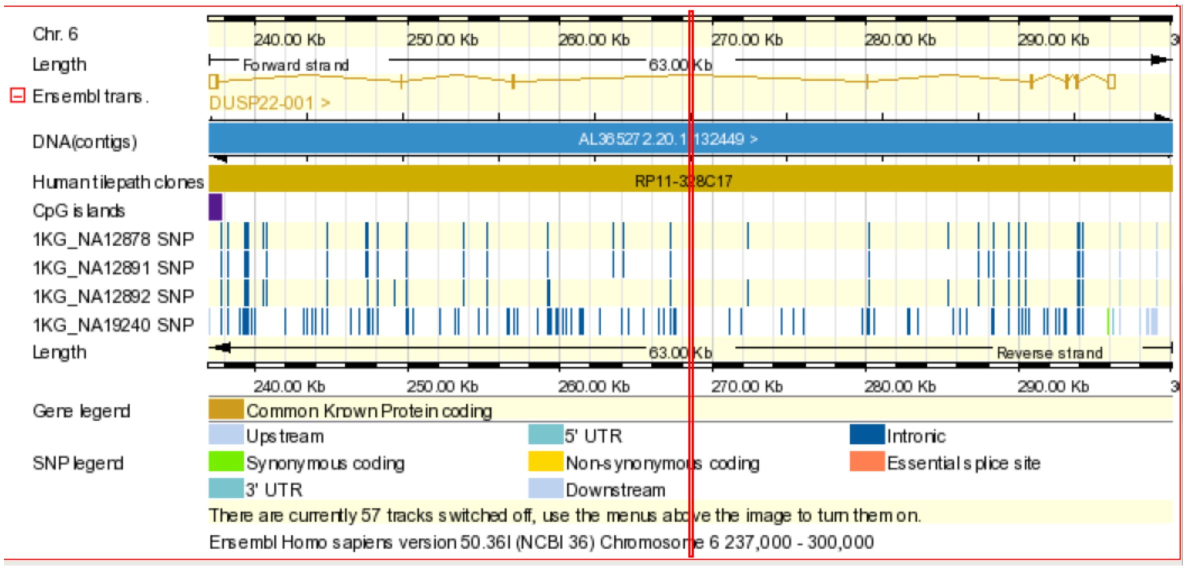
```
> nloc = nov$loc  # this one is evaluated
> nranges = IRanges(nloc, nloc)
> granges = IRanges(rsgdf$start, rsgdf$end) # no guarantee of annotation
> length(nranges)

[1] 4075

> length(granges)

[1] 73

> sum(nranges %in% granges)

[1] 1506

> match(nranges,granges)[1:200]
```

```
  [1] NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA
 [26] NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA
 [51] NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA
 [76] NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA
[101] NA NA NA NA NA NA NA NA  1  1  1  1  1  1  1  1  1  1  1  1  1  1  1  1  1
[126]  1  1  1  1  1 NA NA NA NA NA NA NA NA  2  2 NA NA NA NA NA NA NA NA NA NA
[151] NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA
[176] NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA NA
```

We can see that there is a batch of variants present in the first gene, and this is confirmed by checking the 1KG browser.

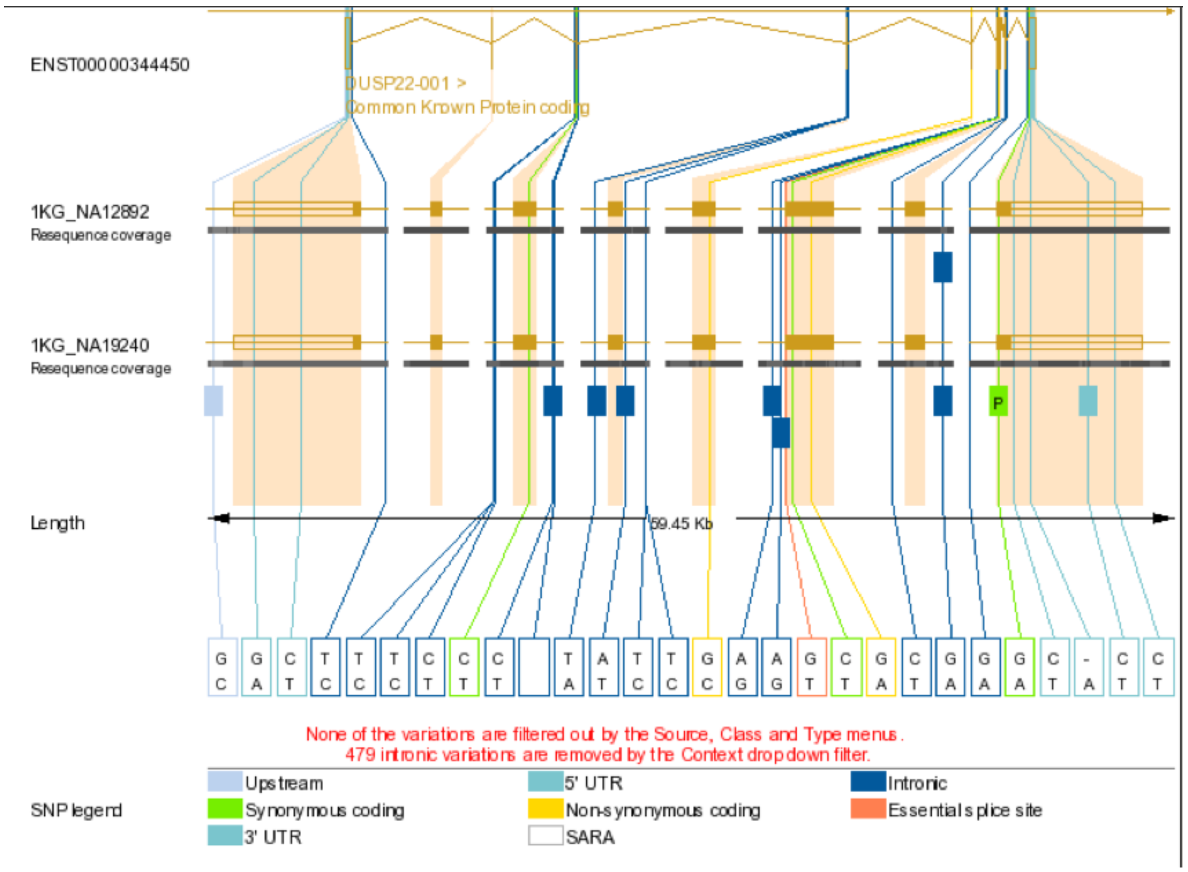
```
NCBI36        2201 TGGGTAATAAATTTCTTGACTTTTATTATCTTGTGAATAGAGACTCACAG  2250
1KG_NA19240   2201 TGGGTAATAAATTTCTTGACTTTTATTATCTTGTGAATAGAGACTCACAG  2250

NCBI36        2251 TGTATCAATGAAGAAATACATGTATAGAGAGATTTAGAGACTTCCCTGCT  2300
1KG_NA19240   2251 TGTATCAATGAAGAAATAYATGTATAGAGAGATTTAGAGACTTCCCTGCT  2300

NCBI36        2301 TCAACACTTATTGATTATGTACATACTATGAGCCAGGTGTTGTGCTAGTG  2350
1KG_NA19240   2301 TCAACACTTATTGATTATGTACATACTATGAGCCAGGTGTTGTGCTAGTG  2350

NCBI36        2351 GGTGGGGAGGGGGCAAAGGGAGTACAGTGGTAAATGACCCAAAGATTCTT  2400
1KG_NA19240   2351 GGTGGGGAGGGGGCAAAGGGAGTACAGTGGTAAATGACCCAAAGATTCTT  2400

NCBI36        2401 CACAGAACCTTCAGTACAGGTCAGCATTGTCCAGCACTAATATGATGCGA  2450
1KG_NA19240   2401 CACAGAACCTTCAGTAYAGGTCARCATTGTCCAGCACTAATATGATGCGA  2450

NCBI36        2451 GCTATATATGTAACTTAAAATGTTCCGGTAGCCACATTAAAAAATAAACC  2500
1KG_NA19240   2451 GCTATATRTGTAACTTAAAATGTTCCGGTAGCCACATTAAAAAATAAACC  2500

NCBI36        2501 GGTGAAATTAATTTTAATAATATATTTTATTTAACAATATATTAAAACTA  2550
1KG_NA19240   2501 GGTGAAATTAATTTTAATAATATATTTTATTTAACAATATATTAAAACTA  2550

NCBI36        2551 TTGTTTCAACATGTAACCAATATATAAAGTTATTCATGGAAGATTCCATG  2600
1KG_NA19240   2551 TTGTTTCAACATGTAAYCAATATATAAAGTTATTCATGGAAGATTCCATG  2600

NCBI36        2601 TTTTTTACTATGTCTTTGTGAAAGGAAAAATCTCGGGACCCCCAGATCAC  2650
1KG_NA19240   2601 TTTTTTACTATGTCTTTGTGAAAGGAAAAATCTCGGGACCCCCAGATCAC  2650
```

Looking in more detail, we have



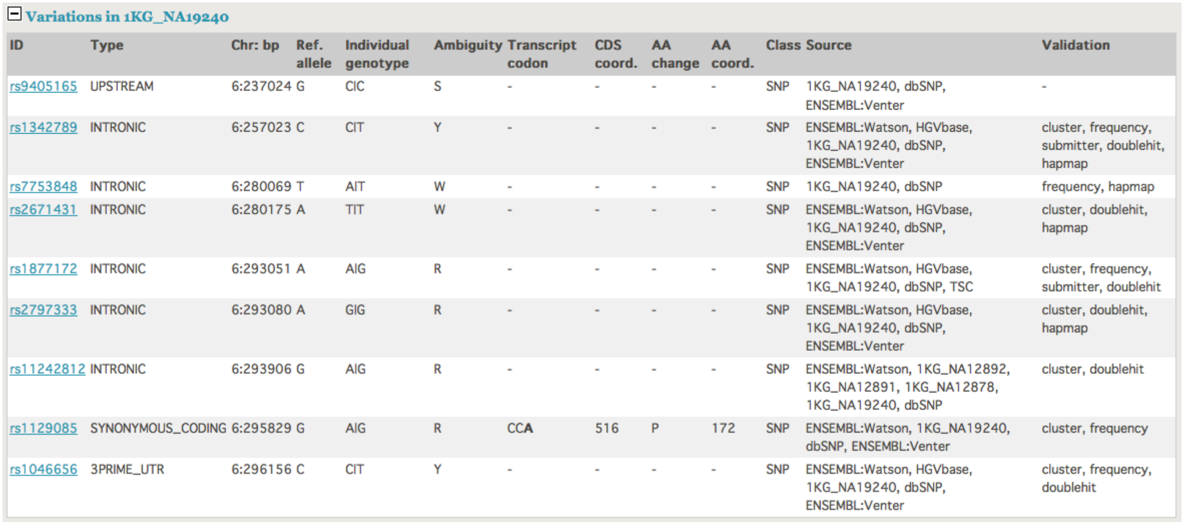
There are currently 57 tracks switched off, use the menus above the image to turn them on.
Ensembl Homo sapiens version 50.36l (NCBI 36) Chromosome 6 237,000 - 300,000

and this can be exploded into the Ensembl variant browser view



None of the variations are filtered out by the Source, Class and Type menus.
479 intronic variations are removed by the Context drop down filter.

with textual metadata view



Variations in 1KG_NA19240

ID	Type	Chr: bp	Ref. allele	Individual genotype	Ambiguity	Transcript codon	CDS coord.	AA change	AA coord.	Class	Source	Validation
rs9405165	UPSTREAM	6:237024	G	C|C	S	-	-	-	-	SNP	1KG_NA19240, dbSNP, ENSEMBL:Venter	-
rs1342789	INTRONIC	6:257023	C	C|T	Y	-	-	-	-	SNP	ENSEMBL:Watson, HGVbase, 1KG_NA19240, dbSNP, ENSEMBL:Venter	cluster, frequency, submitter, doublehit, hapmap
rs7753848	INTRONIC	6:280069	T	A|T	W	-	-	-	-	SNP	1KG_NA19240, dbSNP	frequency, hapmap
rs2671431	INTRONIC	6:280175	A	T|T	W	-	-	-	-	SNP	ENSEMBL:Watson, HGVbase, 1KG_NA19240, dbSNP, ENSEMBL:Venter	cluster, doublehit, hapmap
rs1877172	INTRONIC	6:293051	A	A|G	R	-	-	-	-	SNP	ENSEMBL:Watson, HGVbase, 1KG_NA19240, dbSNP, TSC	cluster, frequency, submitter, doublehit
rs2797333	INTRONIC	6:293080	A	G|G	R	-	-	-	-	SNP	ENSEMBL:Watson, HGVbase, 1KG_NA19240, dbSNP, ENSEMBL:Venter	cluster, doublehit, hapmap
rs11242812	INTRONIC	6:293906	G	A|G	R	-	-	-	-	SNP	ENSEMBL:Watson, 1KG_NA12892, 1KG_NA12891, 1KG_NA12878, 1KG_NA19240, dbSNP	cluster, doublehit
rs1129085	SYNONYMOUS_CODING	6:295829	G	A|G	R	CCA	516	P	172	SNP	ENSEMBL:Watson, 1KG_NA19240, dbSNP, ENSEMBL:Venter	cluster, frequency
rs1046656	3PRIME_UTR	6:296156	C	C|T	Y	-	-	-	-	SNP	ENSEMBL:Watson, HGVbase, 1KG_NA19240, dbSNP, ENSEMBL:Venter	cluster, frequency, doublehit

So it seems DUSP22 resides over plenty of known SNP; our computations are supposed to reveal hitherto unknown variants in this region for this individual.

5.3 Exercises

1. The `oregdf` data frame is supplied in *ind1KG*, containing information on regulatory elements annotated in oreganno. How many novel variants for NA19240 lie in oreganno regulatory regions? What types of regions are occupied?

2. Derive a data frame for regions of nucleosome occupancy in our 10 Mb segment, and check how many of the novel variants lie in such regions.