

Overview of ensemblVEP Pre Ensembl 90

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1 Introduction

Ensembl provides the facility to predict functional consequences of known and unknown variants using the Variant Effect Predictor (VEP). The `ensemblVEP` package wraps Ensembl VEP and returns the results as Robjects or a file on disk. To use this package the Ensembl VEP perl script must be installed in your path. See the package README for details.

NOTE: As of Ensembl version 88 the VEP script has been renamed from `variant_effect_predictor.pl` to `vep`. The `ensemblVEP` package code and documentation have been updated to reflect this change.

Downloads: <http://uswest.ensembl.org/info/docs/tools/vep/index.html>

Complete documentation for runtime options: http://uswest.ensembl.org/info/docs/tools/vep/script/vep_options.html

To test that Ensembl VEP is properly installed, enter the name of the script from the command line:

```
vep
```

2 Results as R objects

```
> library(ensemblVEP)
```

The `ensemblVEP` function can return variant consequences from Ensembl VEP as Robjects (GRanges or VCF) or write them to a file. The default behavior returns a GRanges. Runtime options are stored in a `VEPParam` object and allow a great deal of control over the content and format of the results. See the man pages for more details.

```
> ?ensemblVEP
```

```
> ?VEPParam
```

The default runtime options can be inspected by creating a `VEPParam`.

```
> param <- VEPParam(version=88)
```

```
> param
```

```
class: VEPParam88
identifier(0):
colocatedVariants(0):
dataformat(0):
basic(0):
input(1): species
```

```

cache(3): dir, dir_cache, dir_plugins
output(1): terms
filterqc(0):
database(1): database
advanced(1): buffer_size
version: 88
scriptPath:

```

```
> basic(param)
```

```
$verbose
[1] FALSE
```

```
$quiet
[1] FALSE
```

```
$no_progress
[1] FALSE
```

```
$config
character(0)
```

```
$everything
[1] FALSE
```

```
$fork
numeric(0)
```

Using a vcf file from VariantAnnotation as input, we query Ensembl VEP with the default runtime parameters.

```

> fl <- system.file("extdata", "gl_chrl.vcf", package="VariantAnnotation")
> gr <- ensemblVEP(fl)

```

Consequence data are parsed into the metadata columns of the GRanges. To control the type and amount of data returned see the options in output (VEPParam()).

```
> head(gr, 3)
```

```
GRanges object with 3 ranges and 23 metadata columns:
```

	seqnames	ranges	strand	Allele	
	<Rle>	<IRanges>	<Rle>	<factor>	
rs6054257	20	[14370, 14370]	*	A	
20:17330_T/A	20	[17330, 17330]	*	A	
rs6040355	20	[1110696, 1110696]	*	G	
	Consequence	IMPACT	SYMBOL	Gene	
	<factor>	<factor>	<factor>	<factor>	
rs6054257	intergenic_variant	MODIFIER	<NA>	<NA>	
20:17330_T/A	intergenic_variant	MODIFIER	<NA>	<NA>	
rs6040355	upstream_gene_variant	MODIFIER	PSMF1	ENSG00000125818	
	Feature_type	Feature	BIOTYPE	EXON	
	<factor>	<factor>	<factor>	<factor>	
rs6054257	<NA>	<NA>	<NA>	<NA>	
20:17330_T/A	<NA>	<NA>	<NA>	<NA>	
rs6040355	Transcript	ENST00000479715	processed_transcript	<NA>	
	INTRON	HGVSc	HGVSp	cDNA_position	CDS_position
	<factor>	<factor>	<factor>	<factor>	<factor>
rs6054257	<NA>	<NA>	<NA>	<NA>	<NA>
20:17330_T/A	<NA>	<NA>	<NA>	<NA>	<NA>
rs6040355	<NA>	<NA>	<NA>	<NA>	<NA>
	Protein_position	Amino_acids	Codons	Existing_variation	
	<factor>	<factor>	<factor>	<factor>	

```

rs6054257          <NA>          <NA>          <NA>          <NA>
20:17330_T/A      <NA>          <NA>          <NA>          <NA>
rs6040355         <NA>          <NA>          <NA>          <NA>
          DISTANCE  STRAND      FLAGS  SYMBOL_SOURCE  HGNC_ID
          <factor> <factor> <factor>      <factor> <factor>
rs6054257         <NA>          <NA>          <NA>          <NA>      <NA>
20:17330_T/A     <NA>          <NA>          <NA>          <NA>      <NA>
rs6040355        2610          1          <NA>          HGNC      HGNC:9571
-----

```

seqinfo: 1 sequence from genome

Next we use a vcf of structural variants as input

```
> fl <- system.file("extdata", "structural.vcf", package="VariantAnnotation")
```

and request that a VCF object be returned by setting the vcf option in the dataformat slot to TRUE.

```
> param <- VEPPParam(dataformat=c(vcf=TRUE), version=88)
```

An call to ensemblVEP results in an error.

```
> vcf <- ensemblVEP(fl, param)
2012-12-03 16:40:55 - Starting...
ERROR: Could not detect input file format
```

In most situations Ensembl VEP can auto-detect the input format. In this case, however, it cannot so we explicitly set the format option to 'vcf'.

```
> input(param)$format <- "vcf"
```

Try again.

```
> vep <- ensemblVEP(fl, param)
```

Success! When a VCF is returned, consequence data are included as an unparsed INFO column labeled CSQ.

```
> info(vep)$CSQ
```

```

CharacterList of length 6
[[1]] -|intergenic_variant|MODIFIER|
[[2]] deletion|intron_variant&non_coding_transcript_variant&feature_truncatio...
[[3]] deletion|intergenic_variant|MODIFIER|
[[4]] insertion|intron_variant&feature_elongation|MODIFIER|SETD5|ENSG00000168...
[[5]] duplication|upstream_gene_variant|MODIFIER|RAF1|ENSG00000132155|Transcr...
[[6]] duplication|intron_variant&non_coding_transcript_variant&feature_elonga...

```

The parseCSQToGRanges function parses these data into a GRanges. When the rownames of the original VCF are provided as VCFRowID a metadata column of the same name is included in the output.

```
> vcf <- readVcf(fl, "hg19")
> csq <- parseCSQToGRanges(vep, VCFRowID=rownames(vcf))
> head(csq, 3)
```

GRanges object with 3 ranges and 24 metadata columns:

```

                                     seqnames
                                     <Rle>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C 1
                                     2:321682_T/<DEL> 2
                                     2:321682_T/<DEL> 2
                                     <IRa
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C 2827693-28
                                     2:321682_T/<DEL> 3
                                     2:321682_T/<DEL> 3

```

		strand
		<Rle>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		*
	2:321682_T/	*
	2:321682_T/	*
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		
	2:321682_T/	
	2:321682_T/	
		VCFRowID
		<integer>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		2
	2:321682_T/	3
	2:321682_T/	3
		Allel
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		deletio
	2:321682_T/	deletio
	2:321682_T/	deletio
		interg
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		intron_var
	2:321682_T/	intron_var
	2:321682_T/	intron_var
		IMPAC
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		MODIFIE
	2:321682_T/	MODIFIE
	2:321682_T/	MODIFIE
		SYMB
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	LINC0186
	2:321682_T/	LINC0186
		<chara
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		ENSG000002
	2:321682_T/	ENSG000002
	2:321682_T/	ENSG000002
		Feature_ty
		<characte
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	Transcri
	2:321682_T/	Transcri
		Fe
		<chara
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		ENST000004
	2:321682_T/	ENST000004
	2:321682_T/	ENST000004
		BIOTYP
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	lncRN
	2:321682_T/	lncRN
		EXC
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>

```

2:321682_T/<DEL> <NA>
2:321682_T/<DEL> <NA>
INTRON
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL> 1/
2:321682_T/<DEL> 1/
HGVS
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL> <NA>
2:321682_T/<DEL> <NA>
HGVS
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL> <NA>
2:321682_T/<DEL> <NA>
cDNA_posit
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <
2:321682_T/<DEL> <
2:321682_T/<DEL> <
CDS_positi
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <M
2:321682_T/<DEL> <M
2:321682_T/<DEL> <M
Protein_po
<char
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
Amino_acid
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL> <NA>
2:321682_T/<DEL> <NA>
Codon
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL> <NA>
2:321682_T/<DEL> <NA>
Existing_v
<ch
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
DISTANC
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL> <NA>
2:321682_T/<DEL> <NA>
STRAN
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C <NA>
2:321682_T/<DEL>
2:321682_T/<DEL>
FLAG

```

```

1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
SYMBOL_SOURCE
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
HGNC_ID
<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL> HGNC:5268
2:321682_T/<DEL> HGNC:5268

```

```
-----
seqinfo: 4 sequences from genome; no seqlengths
```

The `VCFRowID` column maps the expanded `CSQ` data back to the rows in the `VCF` object. This index can be used to subset the original `VCF`.

```

> vcf[csq$"VCFRowID"]

class: CollapsedVCF
dim: 81 1
rowRanges(vcf):
  GRanges with 5 metadata columns: paramRangeID, REF, ALT, QUAL, FILTER
info(vcf):
  DataFrame with 10 columns: BKPTID, CIEND, CIPOS, END, HOMLEN, HOMSEQ, IMPR...
info(header(vcf)):
  Number Type Description
BKPTID . String ID of the assembled alternate allele in the asse...
CIEND 2 Integer Confidence interval around END for imprecise var...
CIPOS 2 Integer Confidence interval around POS for imprecise var...
END 1 Integer End position of the variant described in this re...
HOMLEN . Integer Length of base pair identical micro-homology at ...
HOMSEQ . String Sequence of base pair identical micro-homology a...
IMPRECISE 0 Flag Imprecise structural variation
MEINFO 4 String Mobile element info of the form NAME,START,END,P...
SVLEN . Integer Difference in length between REF and ALT alleles
SVTYPE 1 String Type of structural variant
geno(vcf):
  List of length 4: GT, GQ, CN, CNQ
geno(header(vcf)):
  Number Type Description
GT 1 String Genotype
GQ 1 Float Genotype quality
CN 1 Integer Copy number genotype for imprecise events
CNQ 1 Float Copy number genotype quality for imprecise events

```

3 Write results to a file

In the previous section we saw Ensembl VEP results returned as `R` objects in the workspace. Alternatively, these results can be written directly to a file. The flag that controls how the data are returned is the `output_file` flag in the `input` options.

When `output_file` is an empty character (default), the results are returned as either a `GRanges` or `VCF` object.

```

> input(param)$output_file

character(0)

```

To write results directly to a file, specify a file name for the `output_file` flag.

```
> input(param)$output_file <- "/mypath/myfile"
```

The file can be written as a *vcf* or *gvf* by setting the options in the *dataformat* slot to TRUE. If neither of *vcf* or *gvf* are TRUE the file is written out as tab delimited.

```
> ## Write a vcf file to myfile.vcf:
> myparam <- VEPParam(dataformat=c(vcf=TRUE),
+                       input=c(output_file="/path/myfile.vcf"), version=88)
> ## Write a gvf file to myfile.gvf:
> myparam <- VEPParam(dataformat=c(gvf=TRUE),
+                       input=c(output_file="/path/myfile.gvf"), version=88)
> ## Write a tab delimited file to myfile.txt:
> myparam <- VEPParam(input=c(output_file="/path/myfile.txt"), version=88)
```

4 Configuring runtime options

The Ensembl VEP web page has complete descriptions of all runtime options. http://uswest.ensembl.org/info/docs/tools/vep/script/vep_options.html Below are examples of how to configure the runtime options in the *VEP-Param* for specific situations. Investigate the differences in results using a sample file from *VariantAnnotation*.

```
> fl <- system.file("extdata", "ex2.vcf", package="VariantAnnotation")
```

- Add regulatory region consequences:

```
> param <- VEPParam(output=c(regulatory=TRUE), version=88)
> gr <- ensemblVEP(fl, param)
```

- Specify input file format as VCF, add HGNC gene identifiers, output SO consequence terms:

```
> param <- VEPParam(input=c(format="vcf"),
+                   output=c(terms="so"),
+                   identifiers=c(symbol=TRUE), version=88)
> gr <- ensemblVEP(fl, param)
```

- Check for co-located variants, output only coding sequence consequences, output HGVS names:

```
> param <- VEPParam(filterqc=c(coding_only=TRUE),
+                   colocatedVariants=c(check_existing=TRUE),
+                   identifiers=c(symbol=TRUE), version=88)
> gr <- ensemblVEP(fl, param)
```

- Add SIFT score and prediction, PolyPhen prediction only, output results as VCF:

```
fl <- system.file("extdata", "chr22.vcf.gz", package="VariantAnnotation")
param <- VEPParam(output=c(sift="b", polyphen="p"),
                  dataformat=c(vcf=TRUE), version=88)
vcf <- ensemblVEP(fl, param)
csq <- parseCSQToGRanges(vcf)

> head(levels(mcols(csq)$SIFT))
[1] "deleterious(0.01)" "deleterious(0.02)" "deleterious(0.03)"
[4] "deleterious(0.04)" "deleterious(0.05)" "deleterious(0)"

> levels(mcols(csq)$PolyPhen)
[1] "benign" "possibly_damaging" "probably_damaging"
[4] "unknown"
```

5 sessionInfo()

```
> sessionInfo()
```

```
R version 4.2.1 (2022-06-23)  
Platform: x86_64-pc-linux-gnu (64-bit)  
Running under: Ubuntu 20.04.4 LTS
```

```
Matrix products: default
```

```
BLAS: /home/biocbuild/bbs-3.15-bioc/R/lib/libRblas.so
```

```
LAPACK: /home/biocbuild/bbs-3.15-bioc/R/lib/libRlapack.so
```

```
locale:
```

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

```
[1] stats4      stats      graphics  grDevices  utils      datasets  methods  
[8] base
```

```
other attached packages:
```

```
[1] ensemblVEP_1.38.0          VariantAnnotation_1.42.1  
[3] Rsamtools_2.12.0          Biostrings_2.64.0  
[5] XVector_0.36.0            SummarizedExperiment_1.26.1  
[7] Biobase_2.56.0            MatrixGenerics_1.8.1  
[9] matrixStats_0.62.0        GenomicRanges_1.48.0  
[11] GenomeInfoDb_1.32.2       IRanges_2.30.0  
[13] S4Vectors_0.34.0          BiocGenerics_0.42.0
```

```
loaded via a namespace (and not attached):
```

```
[1] Rcpp_1.0.9                 lattice_0.20-45          prettyunits_1.1.1  
[4] png_0.1-7                  assertthat_0.2.1        digest_0.6.29  
[7] utf8_1.2.2                  BiocFileCache_2.4.0     R6_2.5.1  
[10] RSQLite_2.2.14             httr_1.4.3              pillar_1.7.0  
[13] zlibbioc_1.42.0            rlang_1.0.4             GenomicFeatures_1.48.3  
[16] progress_1.2.2             curl_4.3.2              blob_1.2.3  
[19] Matrix_1.4-1               BiocParallel_1.30.3    stringr_1.4.0  
[22] RCurl_1.98-1.7             bit_4.0.4               biomaRt_2.52.0  
[25] DelayedArray_0.22.0        rtracklayer_1.56.1     compiler_4.2.1  
[28] pkgconfig_2.0.3            tidyselect_1.1.2        KEGGREST_1.36.3  
[31] tibble_3.1.7               GenomeInfoDbData_1.2.8  codetools_0.2-18  
[34] XML_3.99-0.10             fansi_1.0.3             crayon_1.5.1  
[37] dplyr_1.0.9                dbplyr_2.2.1            GenomicAlignments_1.32.0  
[40] bitops_1.0-7              rappdirs_0.3.3         grid_4.2.1  
[43] lifecycle_1.0.1           DBI_1.1.3               magrittr_2.0.3  
[46] cli_3.3.0                  stringi_1.7.8           cachem_1.0.6  
[49] xml2_1.3.3                 ellipsis_0.3.2         filelock_1.0.2  
[52] vctrs_0.4.1                generics_0.1.3         rjson_0.2.21  
[55] restfulr_0.0.15           tools_4.2.1            bit64_4.0.5  
[58] BSgenome_1.64.0           glue_1.6.2             purrr_0.3.4  
[61] hms_1.1.1                  yaml_2.3.5             parallel_4.2.1  
[64] fastmap_1.1.0             AnnotationDbi_1.58.0    memoise_2.0.1  
[67] BiocIO_1.6.0
```