

Overview of **ensemblVEP** Pre Ensembl 90

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Contents

1	Introduction	1
2	Results as R objects	1
3	Write results to a file	6
4	Configuring runtime options	7
5	sessionInfo()	8

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_chr1.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
	<factor>	<factor>	<factor>	<factor>
rs6054257	<NA>	<NA>	<NA>	<NA>
20:17330_T/A	<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 <- VEPParam(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_CCGTGGATGCGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		1
	2:321682_T/	2
	2:321682_T/	2
		ranges
		<IRanges>
1:2827693_CCGTGGATGCGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C	2827693-2827762	
	2:321682_T/	321682
	2:321682_T/	321682

		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
		Allele
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		-
	2:321682_T/	deletion
	2:321682_T/	deletion
		Consequence
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		intergenic_variant
	2:321682_T/	intron_variant&non_c..
	2:321682_T/	intron_variant&non_c..
		IMPACT
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		MODIFIER
	2:321682_T/	MODIFIER
	2:321682_T/	MODIFIER
		SYMBOL
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	LINC01865
	2:321682_T/	LINC01865
		Gene
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	ENSG00000233684
	2:321682_T/	ENSG00000233684
		Feature_type
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	Transcript
	2:321682_T/	Transcript
		Feature
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	ENST00000430529
	2:321682_T/	ENST00000436808
		BIOTYPE
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	lncRNA
	2:321682_T/	lncRNA
		EXON
		<character>
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>

	2:321682_T/	<NA>
	2:321682_T/	<NA>
	INTRON	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	1/1
	2:321682_T/	1/3
	HGVSc	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	HGVSp	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	cDNA_position	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	CDS_position	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	Protein_position	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	Amino_acids	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	Codons	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	Existing_variation	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	DISTANCE	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	<NA>
	2:321682_T/	<NA>
	STRAND	
	<character>	
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C		<NA>
	2:321682_T/	1
	2:321682_T/	1
	FLAGS	

```

1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
SYMBOL_SOURCE
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
HGNC_ID
1:2827693_CCGTGGATGCGGGGACCCGCATCCCCTCTCCCTTCACAGCTGAGTGACCCACATCCCCTCTCCCCTCGCA/C
2:321682_T/<DEL>
2:321682_T/<DEL>
HGNC:52684
HGNC:52684

```

seqinfo: 4 sequences from genome; no seqlengths

The `VCFRowID` columns 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: 24 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 <- VEPPParam(dataformat=c(vcf=TRUE),
+                       input=c(output_file="/path/myfile.vcf"), version=88)
> ## Write a gvf file to myfile.gvf:
> myparam <- VEPPParam(dataformat=c(gvf=TRUE),
+                       input=c(output_file="/path/myfile.gvf"), version=88)
> ## Write a tab delimited file to myfile.txt:
> myparam <- VEPPParam(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 <- VEPPParam(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 <- VEPPParam(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 <- VEPPParam(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 <- VEPPParam(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 4.0.4 (2021-02-15)
```

```
Platform: x86_64-apple-darwin17.0 (64-bit)
```

```
Running under: macOS Mojave 10.14.6
```

```
Matrix products: default
```

```
BLAS: /Library/Frameworks/R.framework/Versions/4.0/Resources/lib/libRblas.dylib
```

```
LAPACK: /Library/Frameworks/R.framework/Versions/4.0/Resources/lib/libRlapack.dylib
```

```
locale:
```

```
[1] C/en_US.UTF-8/en_US.UTF-8/C/en_US.UTF-8/en_US.UTF-8
```

```
attached base packages:
```

```
[1] stats4 parallel stats graphics grDevices utils datasets
```

```
[8] methods base
```

```
other attached packages:
```

```
[1] ensemblVEP_1.32.1 VariantAnnotation_1.36.0  
[3] Rsamtools_2.6.0 Biostrings_2.58.0  
[5] XVector_0.30.0 SummarizedExperiment_1.20.0  
[7] Biobase_2.50.0 MatrixGenerics_1.2.1  
[9] matrixStats_0.58.0 GenomicRanges_1.42.0  
[11] GenomeInfoDb_1.26.4 IRanges_2.24.1  
[13] S4Vectors_0.28.1 BiocGenerics_0.36.0
```

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

```
[1] Rcpp_1.0.6 lattice_0.20-41 prettyunits_1.1.1  
[4] assertthat_0.2.1 utf8_1.2.1 BiocFileCache_1.14.0  
[7] R6_2.5.0 RSQLite_2.2.4 httr_1.4.2  
[10] pillar_1.5.1 zlibbioc_1.36.0 rlang_0.4.10  
[13] GenomicFeatures_1.42.2 progress_1.2.2 curl_4.3  
[16] rstudioapi_0.13 blob_1.2.1 Matrix_1.3-2  
[19] BiocParallel_1.24.1 stringr_1.4.0 RCurl_1.98-1.2  
[22] bit_4.0.4 biomaRt_2.46.3 DelayedArray_0.16.2  
[25] rtracklayer_1.50.0 compiler_4.0.4 pkgconfig_2.0.3  
[28] askpass_1.1 openssl_1.4.3 tidyselect_1.1.0  
[31] tibble_3.1.0 GenomeInfoDbData_1.2.4 XML_3.99-0.5  
[34] fansi_0.4.2 crayon_1.4.1 dplyr_1.0.5  
[37] dbplyr_2.1.0 GenomicAlignments_1.26.0 bitops_1.0-6  
[40] rappdirs_0.3.3 grid_4.0.4 lifecycle_1.0.0  
[43] DBI_1.1.1 magrittr_2.0.1 stringi_1.5.3  
[46] debugme_1.1.0 cachem_1.0.4 xml2_1.3.2  
[49] ellipsis_0.3.1 vctrs_0.3.6 generics_0.1.0  
[52] tools_4.0.4 bit64_4.0.5 BSgenome_1.58.0  
[55] glue_1.4.2 purrr_0.3.4 hms_1.0.0  
[58] fastmap_1.1.0 AnnotationDbi_1.52.0 memoise_2.0.0
```