hpar: The Human Protein Atlas in R

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Abstract

The Human Protein Atlas (HPA) is a systematic study oh the human proteome using antibody-based proteomics. Multiple tissues and cell lines are systematically assayed affinity-purified antibodies and confocal microscopy. The hpar package is an R interface to the HPA project. It distributes three data sets, provides functionality to query these and to access detailed information pages, including confocal microscopy images available on the HPA web page.

Keywords: infrastructure, bioinformatics, proteomics, microscopy

Contents

1 Introduction

1.1 The HPA project

From the Human Protein Atlas[1] (Uhlén et al., 2005; Uhlen et al., 2010) site:

> The Swedish Human Protein Atlas project, funded by the Knut and Alice Wallenberg Foundation, has been set up to allow for a systematic exploration of the human proteome using Antibody-Based Proteomics. This is accomplished by combining high-throughput generation of affinity-purified antibodies with protein profiling in a multitude of tissues and cells assembled in tissue microarrays. Confocal microscopy analysis using human cell lines is performed for more detailed protein localisation. The program hosts the Human Protein Atlas portal with expression profiles of human proteins in tissues and cells.

The hpar package provides functionality to use HPA data from the R interface. It also distributes three data sets available from the HPA site.

Normal tissue data Expression profiles for proteins in human tissues based on immunohistochemisty using tissue micro arrays. The `dataframe` includes Ensembl gene identifier ("Gene"), tissue name ("Tissue"), annotated cell type ("Cell.type"), expression value ("Level"), the type of annotation (annotated protein expression (APE), based on more than one antibody, or staining, based on one antibody only) ("Expression.type"), and the reliability or validation of the expression value ("Reliability").

Subcellular location data Subcellular localisation of proteins based on immunofluorescently stained cells. The `dataframe` includes Ensembl gene identifier ("Gene"), main subcellular location of the protein ("Main.location"), other locations ("Other.location"), the type of annotation (annotated protein expression (APE), based on more than one antibody, or staining, based on one antibody only) ("Expression.type"), and the reliability or validation of the expression value ("Reliability").

RNA data RNA levels in three different cell lines, based on RNA-seq. The `dataframe` includes Ensembl gene identifier ("Gene"), analysed cell line ("Cell.line"), number of reads per kilobase gene model and million reads ("RPKM"), and abundance class ("Abundance").

1.2 HPA data usage policy

The use of data and images from the HPA in publications and presentations is permitted provided that the following conditions are met:

- The publication and/or presentation are solely for informational and non-commercial purposes.
- The source of the data and/or image is referred to the HPA site (`www.proteinatlas.org`) and/or one or more of our publications are cited.

1.3 Installation

hpar is available through the Bioconductor project. Details about the package and the installation procedure can be found on its page[2]. To install using the dedicated Bioconductor infrastructure, run :

[1] http://www.proteinatlas.org/

[2] `http://bioconductor.org/packages/devel/bioc/html/hpar.html`

```
> source("http://bioconductor.org/biocLite.R")
> ## or, if you have already used the above before
> library("BiocInstaller")  ## and to install the package
> biocLite("hpar")
```

After installation, hpar will have to be explicitly loaded with

```
> library("hpar")

This is hpar 1.0.1.  For more information,
please type '?hpar' or 'vignette('hpar')'.
```

so that all the package's functionality and data is available to the user.

2 The hpar package

2.1 Data sets

The three data sets, named `hpaNormalTissue`, `hpaSubcellularLoc` and `hpaRna` in the package can be loaded with the `data` function, as illustrated below for `hpaNormalTissue` below. Each data set is a `dataframe` and can be easily manipulated using standard `R` functionality. The code chunk below illustrates some of its properties.

```
> data(hpaNormalTissue)
> dim(hpaNormalTissue)

[1] 1101631       6

> names(hpaNormalTissue)

[1] "Gene"            "Tissue"          "Cell.type"
[4] "Level"           "Expression.type" "Reliability"

> ## Number of genes
> length(unique(hpaNormalTissue$Gene))

[1] 14079

> ## Number of cell types
> length(unique(hpaNormalTissue$Cell.type))

[1] 43

> head(levels(hpaNormalTissue$Cell.type))

[1] "adipocytes"                  "bile duct cells"
[3] "cells in endometrial stroma" "cells in glomeruli"
[5] "cells in granular layer"     "cells in molecular layer"

> ## Number of tissues
> length(unique(hpaNormalTissue$Tissue))

[1] 48

> head(levels(hpaNormalTissue$Tissue))
```

```
[1] "adrenal gland" "appendix"      "bone marrow"   "breast"
[5] "bronchus"      "cerebellum"

> table(hpaNormalTissue$Expression.type)


     APE Staining
  254533   847098
```

2.2 HPA interface

The package provides a interface to the HPA data. The `getHpa` allows to query the data sets described in section 2.1. It takes three arguments, `id`, `hpadata` and `type`, that control the query, what data set to interrogate and how to report results respectively. The HPA data uses Ensembl gene identifiers and `id` must be a valid identifier.`hpadata` must be one of `"NormalTissue"`, `"Rna"` or `"SubcellularLoc"`. `type` can be `data` or `details`. The former is the default and returns a `dataframe` containing the information relevant to `id`. It is also possible to obtained detailed information, (including cell images) as web pages, directly from the HPA web page, using `details`.

We will illustrate this functionality with using the E74-like factor 3 gene (ENSG00000163435) as example.

```
> id <- "ENSG00000163435"
> head(getHpa(id, hpadata = "NormalTissue"))

                  Gene        Tissue           Cell.type Level
670255 ENSG00000163435 adrenal gland     glandular cells  None
670256 ENSG00000163435      appendix     glandular cells  High
670257 ENSG00000163435      appendix     lymphoid tissue  None
670258 ENSG00000163435   bone marrow hematopoietic cells  None
670259 ENSG00000163435        breast          adipocytes  None
670260 ENSG00000163435        breast     glandular cells  None
       Expression.type Reliability
670255             APE        High
670256             APE        High
670257             APE        High
670258             APE        High
670259             APE        High
670260             APE        High

> getHpa(id, hpadata = "SubcellularLoc")

                Gene                       Main.location
7025 ENSG00000163435 Nucleus but not nucleoli;Cytoplasm
     Other.location Expression.type Reliability
7025                            APE        High

> head(getHpa(id, hpadata = "Rna"))

                 Gene   sample Value Unit Abundance
32722 ENSG00000163435    A-431  54.1 FPKM      high
32723 ENSG00000163435   U-2 OS   0.1 FPKM       low
32724 ENSG00000163435 U-251 MG   0.5 FPKM       low
```

If we ask for `detail`, a browser page pointing to the relevant page is open (see figure 1)

```
> getHpa(id, type = "details")
```

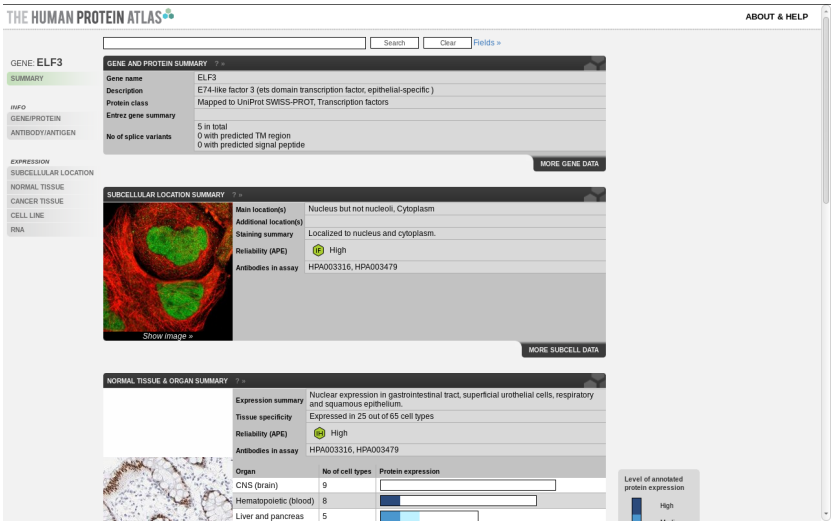

Figure 1: The HPA web page for the E74-like factor 3 gene (ENSG00000163435) gene.

If a user is interested specifically in one data set, it is possible to set `hpadata` globally and omit it in `getHpa`. This is done by setting the `hpar` options `hpardata` with the `setHparOptions` function. The current default data set can be tested with `getHparOptions`.

```
> getHparOptions()

$hpar
$hpar$hpadata
[1] "NormalTissue"

> setHparOptions(hpadata = "SubcellularLoc")
> getHparOptions()

$hpar
$hpar$hpadata
[1] "SubcellularLoc"

> getHpa(id)

                Gene                        Main.location
7025 ENSG00000163435 Nucleus but not nucleoli;Cytoplasm
     Other.location Expression.type Reliability
7025                            APE        High
```

2.3 HPA release information

Information about the HPA release used to build the installed `hpar` package can be accessed with `getHpaVersion`, `getHpaDate` and `getHpaEnsembl`. Full release details can be found on the HPA release history[3] page.

[3] http://www.proteinatlas.org/about/releases

```
> getHpaVersion()

[1] "Protein Atlas version 10.0"

> getHpaDate()

[1] "2012.09.12"

> getHpaEnsembl()

[1] "67.37"
```

3 A small use case

Let's compare the subcellular localisation annotation obtained from the HPA subcellular location data set and the information available in the Bioconductor annotation packages. The HPA query shown below indicates that the HECW1 (ENSG00000002746) gene main locations are nucleus (but not nucleoli) and cytoplasm.

```
> id <- "ENSG00000002746"
> getHpa(id, "SubcellularLoc")

              Gene                      Main.location Other.location
24 ENSG00000002746 Nucleus but not nucleoli;Cytoplasm
   Expression.type Reliability
24             APE        High
```

Below, we first extract all cellular component GO terms available for ENSG00000002746 from the org.Hs.eg.db human annotation and then retrieve their term definitions using the GO.db database, indicating concordant results. The IDA evidence code indicates that this information is inferred from direct assay.

```
> library(org.Hs.eg.db)
> library(GO.db)
> ans <- select(org.Hs.eg.db, keys = id, cols = c("ENSEMBL", "GO", "ONTOLOGY"),
+     keytype = "ENSEMBL")

Warning:  'select' resulted in 1:many mapping between keys and return
rows

> ans <- ans[ans$ONTOLOGY == "CC", ]
> ans

          ENSEMBL         GO EVIDENCE ONTOLOGY
2 ENSG00000002746 GO:0005634      IDA       CC
3 ENSG00000002746 GO:0005737      IDA       CC

> sapply(as.list(GOTERM[ans$GO]), slot, "Term")

 GO:0005634  GO:0005737
  "nucleus" "cytoplasm"
```

Session information

- R version 2.15.2 (2012-10-26), `x86_64-unknown-linux-gnu`
- Locale: `LC_CTYPE=en_US.UTF-8`, `LC_NUMERIC=C`, `LC_TIME=en_US.UTF-8`, `LC_COLLATE=C`, `LC_MONETARY=en_US.UTF-8`, `LC_MESSAGES=en_US.UTF-8`, `LC_PAPER=C`, `LC_NAME=C`, `LC_ADDRESS=C`, `LC_TELEPHONE=C`, `LC_MEASUREMENT=en_US.UTF-8`, `LC_IDENTIFICATION=C`
- Base packages: base, datasets, grDevices, graphics, methods, stats, utils
- Other packages: AnnotationDbi 1.20.3, Biobase 2.18.0, BiocGenerics 0.4.0, DBI 0.2-5, GO.db 2.8.0, RSQLite 0.11.2, codetools 0.2-8, hpar 1.0.1, knitr 1.0, org.Hs.eg.db 2.8.0
- Loaded via a namespace (and not attached): IRanges 1.16.4, digest 0.6.0, evaluate 0.4.3, formatR 0.7, parallel 2.15.2, stats4 2.15.2, stringr 0.6.2, tools 2.15.2

References

Mathias Uhlén, Erik Björling, Charlotta Agaton, Cristina Al-Khalili A. Szigyarto, Bahram Amini, Elisabet Andersen, Ann-Catrin C. Andersson, Pia Angelidou, Anna Asplund, Caroline Asplund, Lisa Berglund, Kristina Bergström, Harry Brumer, Dijana Cerjan, Marica Ekström, Adila Elobeid, Cecilia Eriksson, Linn Fagerberg, Ronny Falk, Jenny Fall, Mattias Forsberg, Marcus Gry G. Björklund, Kristoffer Gumbel, Asif Halimi, Inga Hallin, Carl Hamsten, Marianne Hansson, My Hedhammar, Görel Hercules, Caroline Kampf, Karin Larsson, Mats Lindskog, Wald Lodewyckx, Jan Lund, Joakim Lundeberg, Kristina Magnusson, Erik Malm, Peter Nilsson, Jenny Odling, Per Oksvold, Ingmarie Olsson, Emma Oster, Jenny Ottosson, Linda Paavilainen, Anja Persson, Rebecca Rimini, Johan Rockberg, Marcus Runeson, Asa Sivertsson, Anna Sköllermo, Johanna Steen, Maria Stenvall, Fredrik Sterky, Sara Strömberg, Mårten Sundberg, Hanna Tegel, Samuel Tourle, Eva Wahlund, Annelie Waldén, Jinghong Wan, Henrik Wernérus, Joakim Westberg, Kenneth Wester, Ulla Wrethagen, Lan Lan L. Xu, Sophia Hober, and Fredrik Pontén. A human protein atlas for normal and cancer tissues based on antibody proteomics. *Molecular & cellular proteomics : MCP*, 4(12): 1920–1932, December 2005. ISSN 1535-9476. doi: 10.1074/mcp.M500279-MCP200. URL http://dx.doi.org/10.1074/mcp.M500279-MCP200.

Mathias Uhlen, Per Oksvold, Linn Fagerberg, Emma Lundberg, Kalle Jonasson, Mattias Forsberg, Martin Zwahlen, Caroline Kampf, Kenneth Wester, Sophia Hober, Henrik Wernerus, Lisa Björling, and Fredrik Ponten. Towards a knowledge-based Human Protein Atlas. *Nature biotechnology*, 28(12):1248–1250, December 2010. ISSN 1546-1696. doi: 10.1038/nbt1210-1248. URL http://dx.doi.org/10.1038/nbt1210-1248.