

restfulSE – experiments with HDF5 server content wrapped in SummarizedExperiment

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1 restfulSE

This R package includes proof-of-concept code illustrating several approaches to SummarizedExperiment design with assays stored out-of-memory.

1.1 HDF5 server backed SummarizedExperiment

HDF Server “extends the HDF5 data model to efficiently store large data objects (e.g. up to multi-TB data arrays) and access them over the web using a RESTful API.” In this `restfulSE` package, several data structures are introduced

- to model the server data architecture and
- to perform targeted extraction of numerical data from HDF5 arrays stored on the server.

We work with HDF Object store (<https://www.hdfgroup.org/solutions/hdf-cloud/>).

1.1.1 Illustration with 10x genomics 1.3 million neurons

We used Martin Morgan’s `TENxGenomics` package to transform the sparse-formatted HDF5 supplied by 10x into a dense HDF5 matrix to support natural slicing. Thanks to native compression in HDF5, the data volume expansion is modest.

A helper function in the `restfulSE` package creates a `RESTfulSummarizedExperiment` instance that points to the full numerical dataset.

```
library(restfulSE)
my10x = se1.3M()
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## snapshotDate(): 2019-10-22
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## see ?restfulSEData and browseVignettes('restfulSEData') for documentation
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## downloading 1 resources
## retrieving 1 resource
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## loading from cache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
my10x
## class: SummarizedExperiment
## dim: 27998 1306127
## metadata(0):
## assays(1): counts
## rownames(27998): ENSMUSG00000051951 ENSMUSG00000089699 ...
##   ENSMUSG00000096730 ENSMUSG00000095742
## rowData names(12): ensid seqnames ... symbol entrezid
## colnames(1306127): AACCTGAGATAGGAG-1 AAACCTGAGCGGCTTC-1 ...
```

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```
##    TTTGTCAGTTAAAGTG-133 TTTGTCATCTGAAAGA-133
## colData names(4): Barcode Sequence Library Mouse
```

As an exercise, we acquire the ENSEMBL identifiers for mouse genes annotated to hippocampus development, which has GO ID [GO:0021766](#), and check counts for 10 genes on 6 samples:

```
library(org.Mm.eg.db)
##
hippdev = select(org.Mm.eg.db,
  keys="GO:0021766", keytype="GO", column="ENSEMBL")$ENSEMBL
## 'select()' returned 1:many mapping between keys and columns
hippdev = intersect(hippdev, rownames(my10x))
#unname(assay(my10x[ hippdev[1:10], 10001:10006]))
```

The result:

```
[,1] [,2] [,3] [,4] [,5] [,6]
[1,] 0 0 0 0 0 0
[2,] 0 0 0 0 0 0
[3,] 0 0 0 1 0 0
[4,] 0 1 2 6 5 0
[5,] 0 0 0 0 0 0
[6,] 1 2 4 8 7 3
[7,] 0 0 0 0 0 0
[8,] 0 0 0 0 0 2
[9,] 0 0 0 0 0 0
[10,] 3 0 3 0 1 9
```

1.1.2 Illustration with GTEx tissue expression

We exported the content of the [recount2 GTEx gene-level quantifications](#) to our HDF5 server.
A convenience function is available:

```
tiss = gtexTiss()
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## snapshotDate(): 2019-10-22
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## see ?restfulSEData and browseVignettes('restfulSEData') for documentation
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## downloading 1 resources
## retrieving 1 resource
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## loading from cache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tiss
## class: RangedSummarizedExperiment
## dim: 58037 9662
```

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```
## metadata(0):
## assays(1): recount
## rownames(58037): ENSG00000000003.14 ENSG00000000005.5 ...
##   ENSG00000283698.1 ENSG00000283699.1
## rowData names(3): gene_id bp_length symbol
## colnames(9662): SRR660824 SRR2166176 ... SRR612239 SRR615898
## colData names(82): project sample ... title characteristics
```

We'll use this remote data as a tool for investigating transcriptional patterns in brain anatomy. We can identify the samples from brain using the 'smtsd' colData element:

```
binds = grep("Brain", tiss$smtsd)
table(tiss$smtsd[binds][1:100]) # check diversity in 100 samples
##
##          Brain - Amygdala
##                           4
##  Brain - Anterior cingulate cortex (BA24)
##                           5
##          Brain - Caudate (basal ganglia)
##                           10
##          Brain - Cerebellar Hemisphere
##                           9
##          Brain - Cerebellum
##                           13
##          Brain - Cortex
##                           13
##  Brain - Frontal Cortex (BA9)
##                           10
##          Brain - Hippocampus
##                           8
##          Brain - Hypothalamus
##                           5
##  Brain - Nucleus accumbens (basal ganglia)
##                           7
##          Brain - Putamen (basal ganglia)
##                           4
##  Brain - Spinal cord (cervical c-1)
##                           3
##          Brain - Substantia nigra
##                           9
```

We'll identify genes annotated to neurotrophic functions using another convenience function in this package:

```
ntgenes = goPatt(termPattern="neurotroph")
## 'select()' returned 1:1 mapping between keys and columns
## 'select()' returned 1:many mapping between keys and columns
head(ntgenes)
##
##          GO EVIDENCE ONTOLOGY      ENSEMBL SYMBOL
## 1 GO:0004897     IBA      MF ENSG00000122756  CNTFR
## 2 GO:0004897     IDA      MF ENSG00000122756  CNTFR
## 3 GO:0004897     IMP      MF ENSG00000160712  IL6R
```

```
## 4 GO:0004897      IDA      MF ENSG00000134352  IL6ST
## 5 GO:0004897      IDA      MF ENSG00000113594  LIFR
## 6 GO:0004897      IBA      MF ENSG00000105246  EBI3
```

2 Some details

2.1 Motivation

Extensive human and computational effort is expended on downloading and managing large genomic data at site of analysis. Interoperable formats that are accessible via generic operations like those in RESTful APIs may help to improve cost-effectiveness of genome-scale analyses.

In this report we examine the use of HDF5 server as a back end for assay data, mediated through the RangedSummarizedExperiment API for interactive use.

A modest server configured to deliver HDF5 content via a RESTful API has been prepared and is used in this vignette.

2.2 Executive summary

We want to provide rapid access to array-like data. We'll work with the Banovich 450k data as there is a simple check against an in-memory representation.

We build a SummarizedExperiment by combining an assay-free RangedSummarizedExperiment with this reference.

```
ehub = ExperimentHub::ExperimentHub()
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## snapshotDate(): 2019-10-22
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tag = names(AnnotationHub::query(ehub, "banoSEMeta"))
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
banoSE = ehub[[tag[1]]]
## see ?restfulSEData and browseVignettes('restfulSEData') for documentation
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## downloading 1 resources
## retrieving 1 resource
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## loading from cache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
ds = HSDSArray(endpoint=URL_hsd(), svrtype="hsds",
               domain="/shared/bioconductor/bano_meQTLex.h5", dsetname="/assay001")
assays(banoSE) = SimpleList(betas=ds)
banoSE
## class: RangedSummarizedExperiment
```

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```
## dim: 329469 64
## metadata(0):
## assays(1): betas
## rownames(329469): cg00000029 cg00000165 ... ch.9.98989607R
##   ch.9.991104F
## rowData names(10): addressA addressB ... probeEnd probeTarget
## colnames(64): NA18498 NA18499 ... NA18489 NA18909
## colData names(35): title geo_accession ... data_row_count naid
```

We can update the SummarizedExperiment metadata through subsetting operations, and then extract the relevant assay data. The data are retrieved from the remote server with the `assay` method.

```
rbanoSub = banoSE[5:8, c(3:9, 40:50)]
## Loading required package: Biostrings
## Loading required package: XVector
##
## Attaching package: 'Biostrings'
## The following object is masked from 'package:base':
##
##     strsplit
assay(rbanoSub)
## <4 x 18> matrix of class DelayedMatrix and type "double":
##           NA18501    NA18502    NA18516 ...    NA19138
## cg00000363  0.325433263  1.377820005  0.596699897  .  0.966695669
## cg00000622  0.003436888 -0.668499289 -1.210634762  .  0.076062477
## cg00000714 -1.184443665 -1.654047967 -0.174729357  .  0.325742947
## cg00000734  0.153831565 -1.299289359  1.903976827  .  1.185320424
##           NA19140
## cg00000363  1.203765271
## cg00000622  0.958031578
## cg00000714 -0.008202908
## cg00000734  0.319937329
```

2.3 10xGenomics examples

2.3.1 t-SNE for a set of genes annotated to hippocampus

We have used Martin Morgan's TENxGenomics package to create a dense HDF5 representation of the assay data, and placed it on the `bigec2` server. The metadata are available as `se100k` in this package; we have used `EnsDb.Mmusculus.v79` to supply gene ranges where available; genes reported but without addresses are addressed at chr1:2 with width 0. The rows are sorted by genomic address within chromosomes.

```
tenx100k = se100k()
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## snapshotDate(): 2019-10-22
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
```

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```
## see ?restfulSEData and browseVignettes('restfulSEData') for documentation
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## downloading 1 resources
## retrieving 1 resource
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## loading from cache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tenx100k
## class: RangedSummarizedExperiment
## dim: 27998 100000
## metadata(1): source
## assays(1): counts
## rownames(27998): ENSMUSG00000109048 ENSMUSG00000109510 ...
##   ENSMUSG0000096768 ENSMUSG00000096850
## rowData names(6): gene_id gene_name ... seq_coord_system symbol
## colnames(100000): AACCTGAGATAGGAG-1 AACCTGAGCGGCTTC-1 ...
##   GACGTTAGTCATACTG-11 GACGTTAGTCCGTGAC-11
## colData names(4): Barcode Sequence Library Mouse
```

We will subset genes annotated to hippocampus development. Here are some related categories:

```
12092 GO:0021766          hippocampus development
12096 GO:0021770          parahippocampal gyrus development
34609 GO:0097410          hippocampal interneuron differentiation
34631 GO:0097432 hippocampal pyramidal neuron differentiation
34656 GO:0097457          hippocampal mossy fiber
35169 GO:0098686          hippocampal mossy fiber to CA3 synapse
42398 GO:1990026          hippocampal mossy fiber expansion
```

```
library(org.Mm.eg.db)
atab = select(org.Mm.eg.db, keys="GO:0021766", keytype="GO", columns="ENSEMBL")
## 'select()' returned 1:many mapping between keys and columns
hg = atab[, "ENSEMBL"]
length(hgok <- intersect(hg, rownames(tenx100k)))
## [1] 61
```

This is a very scattered collection of rows in the matrix. We acquire expression measures for genes annotated to hippocampus on 4000 samples. t-SNE is then used to project the log-transformed measures to the plane.

```
hipn = assay(tenx100k[hgok, 1:4000]) # slow
#d = dist(t(log(1+hipn)), method="manhattan")
#proj = Rtsne(d)

#plot(proj$Y)
```

2.3.2 A set of genes related to the visual cortex

Tasic et al. (Nature neuro 2016, DOI 10.1038/nn.4216) describe single cell analysis of the adult murine brain, identify clusters of cells with distinct transcriptional profiles and anatomic location, and enumerate lists of genes that discriminate these clusters. The tasicST6 DataFrame provides details.

```
#data("tasicST6", package = "restfulSEData")
ehub = ExperimentHub::ExperimentHub()
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## snapshotDate(): 2019-10-22
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tag = names(AnnotationHub::query(ehub, "tasicST6"))
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tasicST6 = ehub[[tag[1]]]
## see ?restfulSEData and browseVignettes('restfulSEData') for documentation
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## downloading 1 resources
## retrieving 1 resource
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## loading from cache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tasicST6
## DataFrame with 49 rows and 4 columns
##           clid      txtype1      txtype2
## <character> <character> <character>
## 1          f01       Vip       Mybpc1
## 2          f02       Vip       Parm1
## 3          f03       Vip       Sncg
## 4          f04       Vip       Chat
## 5          f05       Vip       Gpc3
## ...
## 45         f45   Oligo 9630013A20Rik
## 46         f46   Oligo     Opalin
## 47         f47    Micro      Ctss
## 48         f48    Endo      Xdh
## 49         f49     SMC      Myl9
##
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```

restfulSE – experiments with HDF5 server content wrapped in SummarizedExperiment

```
## 49 c("Bgn", "Nupr1", "Casq2", "Mylk", "Gprc5c", "Slc38a11", "Slc6a20a", "Pcolce", "Vtn", "Cnn2", "Nid1",
```

Key high-level discrimination concerns cells regarded as GABAergic vs. glutamatergic (inhibitory vs excitatory neurotransmission).

2.4 Background

Banovich et al. published a subset of DNA methylation measures assembled on 64 samples of immortalized B-cells from the YRI HapMap cohort.

```
library(restfulSE)
#data("banoSEMeta", package = "restfulSEData")
ehub = ExperimentHub::ExperimentHub()
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## snapshotDate(): 2019-10-22
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
tag = names(AnnotationHub::query(ehub, "banoSEMeta"))
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
banoSEMeta = ehub[[tag[1]]]
## see ?restfulSEData and browseVignettes('restfulSEData') for documentation
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## loading from cache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## Using temporary cache C:\Users\biocbuild\bbs-3.10-bioc\tmpdir\RtmpigIXg6/BiocFileCache
## class: RangedSummarizedExperiment
## dim: 329469 64
## metadata():
## assays(1): betas
## rownames(329469): cg00000029 cg00000165 ... ch.9.98989607R
##   ch.9.991104F
## rowData names(10): addressA addressB ... probeEnd probeTarget
## colnames(64): NA18498 NA18499 ... NA18489 NA18909
## colData names(35): title geo_accession ... data_row_count naid
```

The numerical data have been exported using H. Pages' saveHDF5SummarizedExperiment applied to the banovichSE SummarizedExperiment in the yriMulti package. The HDF5 component is simply copied into the server data space on the remote server.