R for cytometry - manual gates

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Installing packages

```
# install from bioconductor
library(BiocInstalller)
biocLite(CytoML)
# update to newest version
library(devtools)
install_github("blasern/CytoML", ref="trunk",
               dependencies=FALSE)
```

Download data from cytobank

```
# load CRAN package
require(CytobankAPI)
# Authenticate via authentication token
cyto_session <- authenticate(site="cellmass",
                             auth_token="xxx")
# specify experiment id
experiment_id <- 7266
# specify data directory
data_dir <- "fcs_from_cytobank"
dir.create(data_dir, recursive = TRUE)
# list fcs files
files <- fcs_files.list(cyto_session,
                        experiment_id = experiment_id)
```

Download data from cytobank

```
# download fcs files
downloaded_zip <- fcs_files.download_zip(
  UserSession = cyto_session,
  experiment_id = experiment_id,
  fcs_files = files[, "id"],
  directory = data_dir,
  timeout = 60*nrow(files))
# unzip files
unzip(downloaded_zip, exdir = data_dir)
# list fcs files
fcs_files <- paste0(
  data_dir, "/",
  list.files(data_dir, pattern="\\.fcs"))
```

Download manual gates

```
# download gates
gates_list <- gates.list(cyto_session,
                         experiment_id = experiment_id)
gates_file <- gates.gatingML_download(
  cyto_session, experiment_id,
  directory = data_dir,
  timeout = 60*nrow(gates))
```

Read fcs files and gates

```
library(flowWorkspace)
library(CytoML)

gs <- cytobank2GatingSet(gates_file, fcs_files)
```

Combine to one matrix

```
# extract groups from gating set
cells <- fsApply(as.flowSet(gs@data), identity,
                 use.exprs=TRUE)
groups <- do.call(rbind, lapply(gs, function(x)
  sapply(getNodes(x)[-1], function(y)
    getIndices(x, y))))
combined <- cbind(cells, groups)
```

From previous classes

- Plot tSNE with different cell populations colored
- Compare with SPADE clusters
- Add combined data to a flowSet
- Upload final data to cytobank