SOFTWARE TOOL ARTICLE

**gganatogram: An R package for modular visualisation of anatograms and tissues based on ggplot2 [version 2; peer review: 2 approved]**

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Abstract
Displaying data onto anatomical structures is a convenient technique to quickly observe tissue related information. However, drawing tissues is a complex task that requires both expertise in anatomy and the arts. While web based applications exist for displaying gene expression on anatograms, other non-genetic disciplines lack similar tools. Moreover, web based tools often lack the modularity associated with packages in programming languages, such as R.

Here I present gganatogram, an R package used to plot modular species anatograms based on a combination of the graphical grammar of ggplot2 and the publicly available anatograms from the Expression Atlas. This combination allows for quick and easy, modular, and reproducible generation of anatograms. Using only one command and a data frame with tissue name, group, colour, and value, this tool enables the user to visualise specific human and mouse tissues with desired colours, grouped by a variable, or displaying a desired value, such as gene-expression, pharmacokinetics, or bacterial load across selected tissues. gganatogram consists of 5 highly annotated organisms, male/female human/mouse, and a cell anatogram. It further consists of 24 other less annotated organisms from the animal and plant kingdom. I hope that this tool will be useful by the wider community in biological sciences. Community members are welcome to submit additional anatograms, which can be incorporated into the package.

A stable version gganatogram has been deposited to neuroconductor, and a development version can be found on github/jespermaag/gganatogram. An interactive shiny app of gganatogram can be found on https://jespermaag.shinyapps.io/gganatogram/, which allows for non-R users to create anatograms.

Keywords
Anatograms, Anatomy, Tissues, Organs, ggplot2, R, Expression Atlas, Shiny
Introduction

Efficiently displaying tissue information in multicellular organisms can be a laborious and time consuming process. Often researchers want to showcase differences in values, such as gene expression or pharmacokinetics between tissues in one organism, or between similar tissues in different groups.

Whereas bar charts and heatmaps provide an informative view of the differences between groups, it can be difficult to immediately observe the biological significance (Figure 1a–b). As compared to an anatogram, where it is easy to quickly spot the differences between tissues or groups, and immediately provide biological context to these observations (Figure 1c). This also has the added benefit that the audience, whether reading a paper or attending a lecture, will have to spend less time and effort to grasp the results.

Figure 1. Comparison between barplot (top left), heatmap (top right), and anatogram (bottom) to display tissue values between groups. The values in the graphs are the same.
Several online tools to display gene expression in different tissues already exist\(^1\)\(^4\). Although these tools provide important information regarding gene expression in various tissues and organisms, other disciplines besides genetics are unable to utilise these applications due to the focus on genes. Moreover, these tools often only include a predefined set of experiments that can be visualised, leading to difficulties in presenting your own data. Other caveats with these tools are that it can be laborious to recreate the plot or automatically create plots from results.

Here I present gganatogram, an open source R package based on ggplot2\(^5\) utilising 28 publicly available anatograms from the Expression Atlas\(^1\)\(^2\), and a cellular anatogram from The Protein Atlas\(^6\). With this package it is easy for any R user to quickly visualise anatograms with specified colours, groups, and values. Using the familiar grammar from ggplot2\(^5\), this program allows for modular anatograms to be generated.

**Methods**

**Implementation**

gganatogram is stored on neuroconductor\(^7\), an open-source platform for rapid testing and dissemination of reproducible computational imaging software. A development version can be found on github/jespermaag/gganatogram, which allows for the community to post issues with the package, submit requests, or add anatograms by creating coordinate files.

```r
source("https://neuroconductor.org/neurocLite.R")
neuro_install("gganatogram")
```

The development version can be installed from github:

```r
develtools::install_github("jespermaag/gganatogram")
```

Briefly, to generate the main list objects that contain all tissue coordinates, I downloaded SVG files from the Expression Atlas (SVG files present here\(^2\). (and processed them using a custom python script (available from GitHub). The script scraped through the SVG files to extract the name, coordinates, and SVG transformations. These were then post-processed in R to create the rda files that make up the tissue coordinates. For the cell, the SVG was downloaded from The Protein Atlas\(^6\). Here, I converted the relative coordinates in the SVG to absolute using Inkscape. I then processed the absolute coordinate SVG using python.

**Operation**

gganatogram requires an installation of R\(\geq 3.0.0\), ggplot2\(^5\) v.3.0.0 and ggpolypath\(^8\) v.0.1.0. The program should be able to run on any computer with the system requirements for R. The online shiny version does not require an installation of R since it is run on the server side.

Plots can be generated using a basic data.frame containing organ name, colour, type, or value, with the specified column names below. Organs are plotted one at a time based on the order of the data.frame. The tissue of each consecutive row will be layered on top of the previous. The gganatogram package provides 29 such data.frames containing all tissues available to plot, one for each human and mouse, divided by sex, one cell, and 24 other organisms (Table 1).

```r
hgMale_key, hgFemale_key, mmMale_key, mmFemale_key, cell_key["cell"]
```

These data frames have already specified colour, type, and an assigned random number to facilitate the start of plotting.

```r
head(hgFemale_key)
organ  colour      type     value
1        pancreas  orange digestion 10.373146
2           liver  orange digestion 19.723172
3           colon  orange digestion 14.853335
4     bone_marrow #41ab5d     other 19.681587
5    urinary_bladder  orange digestion 14.914273
6         stomach  orange digestion  2.667599
```
Table 1. Showing all the organisms in gganatogram, how to call their keys, and the number of features per organisms.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Key</th>
<th>Number of features</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Human male</td>
<td>hgMale_key</td>
<td>68</td>
</tr>
<tr>
<td>2 Human female</td>
<td>hgFemale_key</td>
<td>70</td>
</tr>
<tr>
<td>3 Mouse male</td>
<td>mmMale_key</td>
<td>45</td>
</tr>
<tr>
<td>4 Mouse female</td>
<td>mmFemale_key</td>
<td>47</td>
</tr>
<tr>
<td>5 cell</td>
<td>cell_key[&quot;cell&quot;]</td>
<td>24</td>
</tr>
<tr>
<td>6 anolis_carolinensis</td>
<td>other_key[[&quot;anolis_carolinensis&quot;]</td>
<td>4</td>
</tr>
<tr>
<td>7 arabidopsis_thaliana</td>
<td>other_key[[&quot;arabidopsis_thaliana&quot;]</td>
<td>4</td>
</tr>
<tr>
<td>8 bos_taurus</td>
<td>other_key[&quot;bos_taurus&quot;]</td>
<td>10</td>
</tr>
<tr>
<td>9 brachypodium_distachyon.flower_parts</td>
<td>other_key[[&quot;brachypodium_distachyon.flower_parts&quot;]</td>
<td>5</td>
</tr>
<tr>
<td>10 brachypodium_distachyon.whole_plant</td>
<td>other_key[[&quot;brachypodium_distachyon.whole_plant&quot;]</td>
<td>3</td>
</tr>
<tr>
<td>11 gallus_gallus</td>
<td>other_key[&quot;gallus_gallus&quot;]</td>
<td>8</td>
</tr>
<tr>
<td>12 hordeum_vulgare.flower_parts</td>
<td>other_key[&quot;hordeum_vulgare.flower_parts&quot;]</td>
<td>6</td>
</tr>
<tr>
<td>13 hordeum_vulgare.whole_plant</td>
<td>other_key[&quot;hordeum_vulgare.whole_plant&quot;]</td>
<td>5</td>
</tr>
<tr>
<td>14 macaca_mulatta</td>
<td>other_key[&quot;macaca_mulatta&quot;]</td>
<td>6</td>
</tr>
<tr>
<td>15 monodelphis_domestica</td>
<td>other_key[&quot;monodelphis_domestica&quot;]</td>
<td>6</td>
</tr>
<tr>
<td>16 oryza_sativa.flower_parts</td>
<td>other_key[&quot;oryza_sativa.flower_parts&quot;]</td>
<td>5</td>
</tr>
<tr>
<td>17 oryza_sativa.whole_plant</td>
<td>other_key[&quot;oryza_sativa.whole_plant&quot;]</td>
<td>5</td>
</tr>
<tr>
<td>18 papio_anubis</td>
<td>other_key[&quot;papio_anubis&quot;]</td>
<td>15</td>
</tr>
<tr>
<td>19 rattus_norvegicus</td>
<td>other_key[&quot;rattus_norvegicus&quot;]</td>
<td>10</td>
</tr>
<tr>
<td>20 solanum_lycopersicum.flower_parts</td>
<td>other_key[&quot;solanum_lycopersicum.flower_parts&quot;]</td>
<td>7</td>
</tr>
<tr>
<td>21 solanum_lycopersicum.whole_plant</td>
<td>other_key[&quot;solanum_lycopersicum.whole_plant&quot;]</td>
<td>5</td>
</tr>
<tr>
<td>22 sorghum_bicolor.flower_parts</td>
<td>other_key[&quot;sorghum_bicolor.flower_parts&quot;]</td>
<td>7</td>
</tr>
<tr>
<td>23 sorghum_bicolor.whole_plant</td>
<td>other_key[&quot;sorghum_bicolor.whole_plant&quot;]</td>
<td>7</td>
</tr>
<tr>
<td>24 tetraodon_nigroviridis</td>
<td>other_key[&quot;tetraodon_nigroviridis&quot;]</td>
<td>4</td>
</tr>
<tr>
<td>25 triticum_aestivum.flower_parts</td>
<td>other_key[&quot;triticum_aestivum.flower_parts&quot;]</td>
<td>11</td>
</tr>
<tr>
<td>26 triticum_aestivum.whole_plant</td>
<td>other_key[&quot;triticum_aestivum.whole_plant&quot;]</td>
<td>4</td>
</tr>
<tr>
<td>27 xenopus_tropicalis</td>
<td>other_key[&quot;xenopus_tropicalis&quot;]</td>
<td>5</td>
</tr>
<tr>
<td>28 zea_mays.flower_parts</td>
<td>other_key[&quot;zea_mays.flower_parts&quot;]</td>
<td>9</td>
</tr>
<tr>
<td>29 zea_mays.whole_plant</td>
<td>other_key[&quot;zea_mays.whole_plant&quot;]</td>
<td>7</td>
</tr>
</tbody>
</table>

The main function is called gganatogram(). By default, and without any arguments, it plots the outline of a male human with standard ggplot2 parameters. By adding just a few options, it is possible to quickly change to female, fill specified organs by selected colour, or fill the organs based on a value (Figure 2).

```r
library(gganatogram)
library(gridExtra)
organPlot <- data.frame(organ = c("heart", "leukocyte", "nerve", "brain", "liver", "stomach", "colon"),
type = c("circulation", "circulation", "nervous_system", "nervous_system", "digestion", "digestion", "digestion"),
colour = c("red", "red", "purple", "purple", "orange", "orange", "orange"),
value = c(10, 5, 15, 8, 10, 0, 10),
stringsAsFactors=F)
```
A <- gganatogram() + ggtitle("A")
B <- gganatogram(fillOutline="#a6bddb", sex = "female") + ggtitle("B")
C <- gganatogram(data=organPlot, fillOutline="#a6bddb", organism="human", sex="female", fill="colour")+ theme_void() + ggtitle("C")
D <- gganatogram(data=organPlot, fillOutline="#a6bddb", organism="human", sex="female", fill="value") + theme_void() + scale_fill_distiller(palette = "Reds", direction=1) + ggtitle("D")
grid.arrange(A, B, C, D, ncol=4)

Figure 2. (A) Default plot generated by calling gganatogram(), (B) adding female, plotting specified organs by (C) colour, (D) value.

Use cases
This section provides additional plotting examples.

To plot all tissues per organism, use the provided key files that exist per organism and sex. This displays all tissues in the order of each data frame. To change the order in which organs are layered on top of each other, reorder the data frame to have those tissues at the bottom (Figure 3).

library(gganatogram)
library(gridExtra)
hgMale <- gganatogram(data=hgMale_key, fillOutline="#a6bddb", organism="human", sex="male", fill="colour") + theme_void()+ coord_fixed()
hgFemale <- gganatogram(data=hgFemale_key, fillOutline="#a6bddb", organism="human", sex="female", fill="colour") + theme_void()+ coord_fixed()
mmMale <- gganatogram(data=mmMale_key, fillOutline="#a6bddb", organism="mouse", sex="male", fill="colour") + theme_void()+ coord_fixed()
mmFemale <- gganatogram(data=mmFemale_key, outline = T, fillOutline="#a6bddb", organism="mouse", sex="female", fill="colour") + theme_void()+ coord_fixed()
cell <- gganatogram(data=cell_key["cell"], outline = T, fillOutline="#a6bddb", organism="cell", fill="colour") + theme_void()+ coord_fixed()
lay <- rbind(c(1,2,3), c(4,5, 5))
grid.arrange(hgMale, hgFemale, mmMale, mmFemale, cell, layout_matrix=lay)
Figure 3. Displaying all tissues available for human and mouse, male and female. The colours are specified in the provided key data frames.

To compare anatograms, e.g. draw one specific anatogram side by side and compare values, a long table has to be created with the type column changed to the variables to compare. The following code recreates (Figure 1c).

```r
normal <- data.frame(organ = c("heart", "leukocyte", "nerve", "brain", "liver", "stomach", "colon"),
            value = c(10, 5, 1, 2, 2, 5, 5),
            type = rep("Normal", 7),
            stringsAsFactors=F)

cancer <- data.frame(organ = c("heart", "leukocyte", "nerve", "brain", "liver", "stomach", "colon"),
            value = c(5, 5, 10, 12, 15, 5, 10),
            type = rep("Cancer", 7),
            stringsAsFactors=F)

compareGroups <- rbind(normal, cancer)

gganatogram(data=compareGroups, fillOutline="white", organism="human",
            sex="male", fill="value") +
    theme_void() +
    facet_wrap(~type) +
    scale_fill_distiller(palette = "Reds", direction=1)
```
To change the order of how organs are layered on top of each other, change the order of the data frame. The organs to plot on the top layer should be on the end of the data frame (Figure 4).

```
organPlot <- data.frame(organ = c("heart", "leukocyte", "nerve", "brain", "stomach", "colon", "lung", "kidney", "liver"),
value = c(10, 5, 15, 8, 10, 0, 3, 8, 3),
stringsAsFactors=F)
A <- gganatogram(data=organPlot, fillOutline="grey", organism="human", sex="female", fill="value") + theme_void() +
scale_fill_distiller(palette = "Reds", direction=1)
organReorder <- c("lung", "liver", "colon", "nerve", "leukocyte", "kidney", "stomach", "brain", "heart")
organPlotReorder <- organPlot[match(organReorder, organPlot$organ),]
B <- gganatogram(data=organPlotReorder, fillOutline="grey", organism="human", sex="female", fill="value") + theme_void() +
scale_fill_distiller(palette = "Reds", direction=1)
grid.arrange(A, B, ncol=2)
```

![Image](image.png)

**Figure 4.** Changing the order of the data frame results in change in the layer of organs to plot.

Organs can also be separated by faceting, as per standard ggplot2 using facet_wrap (Figure 5). This can help to display organs that are nested on top of each other. On the left, lungs and liver hides other plotted organs. This is corrected to the right in order to show the organs of interest.

```
library(gganatogram)
gganatogram(hgMale_key, fillOutline="#a6bde", organism="human", sex="male", fill="colour") +
theme_void() +
facet_wrap(~type)
```

The cell diagram will be useful for users to plot cellular sub-locations of proteins, mRNAs, or other molecules (Figure 6).
Figure 5. Faceting tissues based on type and displaying the corresponding colour.

Figure 6. Cell diagram to plot values at specific cellular sub-locations.
library(viridis)
library(dplyr)

normal <- cell_key[["cell"]]
normal$type <- "Normal"
cancer <- cell_key[["cell"]]

cellCompartments <- c("intermediate_filaments", "endoplasmic_reticulum", "centrosome", "microtubules", "nucleoplasm", "mitochondria", "endosomes", "lipid_droplets")
cancer[match(cellCompartments, cancer$organ),]$value <- c(2, 0, rep(15, 6))
cancer$type <- "Cancer"

plotCell <- rbind(normal, cancer)
plotCell %>%
  mutate(type = factor(type, levels=c("Normal", "Cancer")))%>%
gganatogram(outline = F, organism="cell", fill="value") + theme_void() +
  coord_fixed() +
  scale_fill_viridis() +
  facet_wrap(~type)

Because I elected to use ggplot2 for the package, the user can add additional layers from standard plots. This can be useful to show highlight features, such as metastasis, location of tissue biopsies, or gene expression of specific biopsies (Figure 7).

**Figure 7.** Geom points added to a gganatogram to show the location of tissue biopsies (top left) along with a barplot of biopsy expression for an example gene (bottom). Another option is to fill both tissues and points by value (top right). Red colour around plot added for emphasis.
library(gganatogram)
library(dplyr)
library(gridExtra)

biopsies <- data.frame(biopsy = c("liver", "heart", "prostate", "stomach", "brain"),
  x = c(50, 55, 53, 60, 57),
  y = c(60, 48, 95, 68, 10),
  value = c(10, 15, 5, 2, 15))
p <- hgMale_key %>%
dplyr::filter(organ %in% c("liver", "heart", "prostate", "stomach", "brain")) %>%
gganatogram(fillOutline="lightgray", organism="human", sex="male",
  fill="colour") + theme_void() +
ggtitle("Position of biopsies")

p <- p + geom_point(data = biopsies, pch=21, size=2, aes(x =x, y = -y,
  fill = biopsy, colour= biopsy))
p2 <- ggplot(biopsies, aes(x = biopsy, y = value, fill = biopsy)) +
  geom_bar(stat= "identity", col="black") +
  theme_minimal() +
  theme(legend.position= "none") +
  theme(axis.text.x = element_text(angle = 60, hjust = 1)) +
  ggtitle("Gene1 expression")

p3 <- hgMale_key%>%
dplyr::filter(organ %in% c("liver", "heart", "prostate", "stomach", "brain")) %>%
gganatogram(fillOutline="lightgray", organism="human", sex="male",
  fill="value") + theme_void() +
ggtitle("Value of biopsies") +
  geom_point(data = biopsies, pch=21, size=3, aes(x =x, y = -y,
  fill = value), colour="red")

lay <- rbind(c(1,2), c(1,2), c(3, NULL))
ggrid.arrange(p, p3, p2, layout_matrix = lay)

Other than human, mouse, and the cell diagram, gganatogram consists of 24 other organisms which can be called with the other_key (Table 1). These consists of a mix of animals and plants. Unfortunately, these organisms are not as detailed which is apparent from the lower number of features to plot (Table 1, Figure 8).

library(gridExtra)
plotList <- list()
for (organism in names(other_key)) {
  plotList[[organism]] <- gganatogram(data=other_key[[organism]], outline = T,
    fillOutline="white", organism=organism, fill="colour") +
    theme_void() +
    ggtile(organism) +
    theme(plot.title = element_text(hjust=0.5, size=9)) +
    coord_fixed()
}
do.call(grid.arrange, c(plotList, ncol=6))

Furthermore, gganatogram has an online shiny app which can be used without any R installation. This app let people can select organisms, colour palette, select tissues and adjust values for the tissues. This should allow for researchers without any experience in R to be able to use gganatogram. The online shiny app is located at https://jespermaag.shinyapps.io/gganatogram/.
Figure 8. All 24 other organisms present in gganatogram with all their tissues plotted.

For R users, the app can easily be run locally with the following command:

```r
library(shiny)
runGitHub("gganatogram", "jespermaag", subdir = "shiny")
```

This command checks for all required packages and installs them if needed.

Summary

In summary, I have designed and implemented an R package to easily visualise anatograms based on ggplot2 and the anatograms from Expression Atlas, which when combined create a powerful tool to plot and display tissue information.

The one line command to generate these plots should allow for users with even limited R knowledge to create informative anatograms for publications or presentations.

Software availability

1. Link to version control repository containing the source code:
   http://neuroconductor.org:8080/package/gganatogram
2. Link to development version:
   github/jespermaag/gganatogram
3. Link to archived source code as at time of revision:
   https://zenodo.org/record/1477479

Software license: GPL-2
Author contributions
JLVM conceptualised the study, decided the methodology, wrote the code, and drafted the manuscript.

Grant information
The author(s) declared that no grants were involved in supporting this work.

Acknowledgments
I would like to thank the neuroconductor team: Ciprian Crainiceanu, John Muschelli, Brian Caffo, and Adi Gherman for storing gganatogram on their repository.

Paul Brennan (@brennanpcardiff) for adding additional checks to the package.

I would like to thank Irene Papatheodorou and the Expression Atlas team at EMBL-EBI for making the anatograms available.

I would also like to thank Anna Antoniak for editing the manuscript, and Stephen Rudley for manuscript feedback.

References

Open Peer Review

Current Peer Review Status: ✔ ✔

Version 2

Reviewer Report 13 December 2018
https://doi.org/10.5256/f1000research.18533.r41909

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Helder I. Nakaya
University of São Paulo, São Paulo, Brazil

Really liked the improved version.

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Systems biology, computational tools, vaccinology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Reviewer Report 21 November 2018
https://doi.org/10.5256/f1000research.18533.r40885

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Saskia Freytag
Population Health and Immunity Division, Walter and Eliza Hall Institute (WEHI), Department of Medical Biology, University of Melbourne, Parkville, Vic, Australia

Really nice update to the article!

Competing Interests: No competing interests were disclosed.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.
This article describes a new R package, which allows easy plotting of discrete and continuous measurements onto human and mouse anatomy.

This is a really valuable R package contribution, as it fills a real void in the current infrastructure. I found the code examples in the manuscript intuitive and easy to run. It is great that the author adopts the popular ggplot2 grammar as well as a tidy data structures.

Minor comments:
1. It would be useful to know how many tissues (and which) can be plotted using this package.
2. An example of changing the order of the data frame to change the layering should be added.
3. I encountered the following error when trying to install the package through neuroconductor:
   Error in latest_neuroc_release(release = "stable") :
   unused argument (release = "stable")

Thank you for making all code (even for processing) publicly available.

Is the rationale for developing the new software tool clearly explained?
Yes

Is the description of the software tool technically sound?
Yes

Are sufficient details of the code, methods and analysis (if applicable) provided to allow replication of the software development and its use by others?
Yes

Is sufficient information provided to allow interpretation of the expected output datasets and any results generated using the tool?
Yes
Are the conclusions about the tool and its performance adequately supported by the findings presented in the article?
Yes

**Competing Interests:** No competing interests were disclosed.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Helder I. Nakaya

University of São Paulo, São Paulo, Brazil

The author describes an R package that displays discrete and continuous data onto anatomical structures. The structures are based on mouse and human anatograms from the Expression Atlas project and the grammar from ggplot2 R library. The code example was easy to run and the necessary input data was intuitive and simple.

The author can increase its usage by providing a webtool (even one based on shiny) that takes as input a csv or tsv table with predefined columns. This would allow physicians and scientists with no background in bioinformatics to easily display their data.

Also, it would be useful to create an anatogram for the human brain and one for the different compartments of a cell (nucleus, mitochondria, etc).

Is the rationale for developing the new software tool clearly explained?
Yes

Is the description of the software tool technically sound?
Yes

Are sufficient details of the code, methods and analysis (if applicable) provided to allow replication of the software development and its use by others?
Yes

Is sufficient information provided to allow interpretation of the expected output datasets and any results generated using the tool?
Yes
Are the conclusions about the tool and its performance adequately supported by the findings presented in the article?
Yes

**Competing Interests:** No competing interests were disclosed.

**Reviewer Expertise:** Systems biology, computational tools, vaccinology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

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