1. UEB Tools app

Title and purposes. Start and finish date. Duration

Title: UEB Tools app

Purposes: Development of a web application of microarray post-analysis tools to facilitate the small changes

that the researchers may ask for in the results of the original experiment.

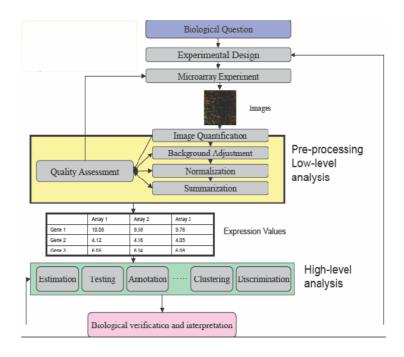
Start date: February 1th, 2019 Finish date: May 20th, 2019 Duration: 4 months approx.

Project description

With the booming omics sciences, today's scientific experiments compile a large amount of data to process and analyze. The necessary software to carry out these analyzes has a very difficult learning curve, since to carry them out requires an advanced level of knowledge in bioinformatics.

In the Statistics and Bioinformatics Unit (UEB) of the Vall d'Hebron Research Institute (VHIR) is offered, among others, the service of performing microarray analysis for biomedical research, but once made and with the results obtained, the researchers usually want to review them and maybe repeat some analyzes or make small changes in the graphs, remove or add samples, or change some parameters (logFC, p-value adjusted ...).

To solve this problem, we propose the creation of a web platform to facilitate the post-analysis previously commented.



Relation of people involved in the project and description of my role

One of the works that the UEB is responsible for is the development of omics analysis and bioinformatics tasks. This web application could help to the UEB workers, specially the bioinformatics section, to do their work easier without having to repeat all the analysis again. This fact demonstrates how important are the software developers in the research world.

Materials required and technical specifications of the project

A RStudio IDE with the last version of R (3.5.1) with different packages installed, the last version of Bioconductor and an Internet connection are required to do this project.

Functional and non-functional requirements

Functional requirements

FR1. Load data

The system will allow to load a file of the Top Tables and Expression Matrix results. High Priority.

FR2. Filter genes selection

The system will allow to define different elements to filter the selection of genes according to choose of the user. To do this, the user will have to choose the data he/she wants to redefine using a numeric input. High Priority.

FR2.1. Set logFC

The system will allow to define the value of the logFC (less than -1 or greater than 1) from which the user wants to name the differentially expressed genes. High Priority.

FR2.2. Set Adjusted P-value

The system will allow to define the value of the adjusted p-value from which the user wishes to name it the genes differentially expressed. High Priority.

FR2.3. Set the selected genes list

The system will allow to set the list of selected genes according the filters applied from which they will be made later analyzes. High Priority

FR3. Do a Volcano Plot

The system will allow to do a Volcano Plot according to the logFC and p-value values from the Top Table.

FR3.1 Show the applied filters in the Volcano Plot

The system will allow to show the applied filters with different colors in the Volcano plot. Low priority.

FR3.2. Choose the number of gene names labels

The system will allow how many gene names labels you want to appear in the plot. Low priority.

FR4. Do a heatmap

The system will allow to do a heatmap from the Expression Matrix with the selected samples. High priority.

FR4.1 Change the colors of the heatmap

The system will allow to change the colors of the heatmap. Low priority.

FR4.2 Change the color input breaks

The system will allow to change the colors input breaks of the heatmap. Low priority.

FR5. Do the Gene Ontology analysis

The system will allow to do the gene ontology analysis with the result of the 3 ontologies: Biological Process, Cellular Component and Molecular Function. High priority.

FR5.1. Do the Gene Ontology analysis table

The system will allow to do the gene ontology analysis table with the result of the 3 ontologies: Biological Process, Cellular Component and Molecular Function. High priority.

FR5.2. Do the Gene Ontology analysis plots

The system will allow to do the gene ontology analysis plots with the result of the 3 ontologies: Biological Process, Cellular Component and Molecular Function. High priority.

FR6. Output results

FR6.1 Output filters applied

The system will allow to output the result of the filters applied. High priority.

FR6.2. Output selected plots

The system will allow to output the result of the selected plots. High priority.

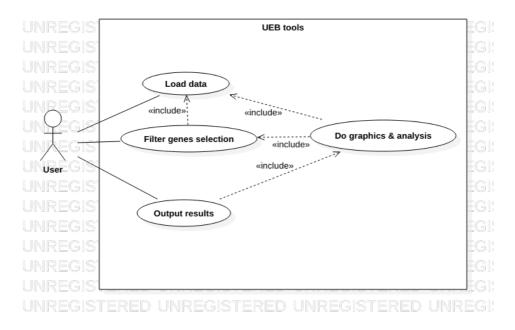
FR6.3. Output analysis results

The system will allow to output the result of the analysis done. High priority.

Nonfunctional requirements

R, Shiny, bioconductor

Use case diagram and their textual description



USE CASE CARD		
ID	1	
NAME	Load data	
DESCRIPTION	The user wants to load a file containing data to analyze	
	NORMAL FLOW	
ACTORS	The final user of the application	
PRECONDITIONS	Not damaged file	
ACTIVATION	User loads the file	
DESCRIPTION	1- User clicks the Browse button	
	2- User selects the file	
	3- User selects the type of separator and decimal	
	4- User clicks the submit button	
	ALTERNATIVE FLOW 1	
DESCRIPTION	The file is not correct	
POSTCONDITION	A message of "Please, upload Express&Top Files" will appear in the check table of below	
	ALTERNATIVE FLOW 2	
DESCRIPTION	The separator and decimal are not correct	
POSTCONDITION	The user will see the wrong format in the check table of below	

	USE CASE CARD
ID	2
NAME	Filter genes selection
DESCRIPTION	The user wants to filter genes by Adj p-value and by logFC
	NORMAL FLOW
ACTORS	The final user of the application
PRECONDITIONS	A file has been uploaded
ACTIVATION	User selects the Adj p-value and logFC values

DESCRIPTION	1- User checks the Adj p-value value	
	2- User checks the logFC value	
	3- User clicks the submit button	
	ALTERNATIVE FLOW 1	
DESCRIPTION	There is no gene with these values	
POSTCONDITION	A message of "There is no data with these values" will appear	
	ALTERNATIVE FLOW 2	
DESCRIPTION	The filters do not run	
POSTCONDITION	The selected genes table will not change after clicking the submit button	

	USE CASE CARD
ID	3
NAME	Doing graphics & analysis
DESCRIPTION	The user wants to see the different plots
	NORMAL FLOW
ACTORS	The final user of the application
PRECONDITIONS	A file has been uploaded and the filters have been selected
ACTIVATION	User loads the file and clicks the section of the graphic he/she wants to see
DESCRIPTION	1- User loads the file
	2- User checks the filters
	3- User clicks the section of the graphic he/she wants to see
	ALTERNATIVE FLOW 1
DESCRIPTION	The graphic is not shown because of an error
POSTCONDITION	Shiny will show a specific error
	ALTERNATIVE FLOW 2
DESCRIPTION	User has not selected filters
POSTCONDITION	Graphics will appear with the default filters

	USE CASE CARD
ID	4
NAME	Output & Download results
DESCRIPTION	The user wants to see the different results and download them
	NORMAL FLOW
ACTORS	The final user of the application
PRECONDITIONS	A file has been uploaded and the filters have been selected
ACTIVATION	User loads the file and clicks the section of the results he/she wants to see
DESCRIPTION	1- User loads the file
	2- User checks the filters
	3- User clicks the section of the graphic he/she wants to see
	4- User clicks the download button
	ALTERNATIVE FLOW 1
DESCRIPTION	The download does not run
POSTCONDITION	Shiny will show a specific error of why it does no run

There is not the test data because I have not worked with databases in this project.

Entity-relation diagram

As it is not an application where there are relations between different classes, an entity-relation diagram will not be attached.

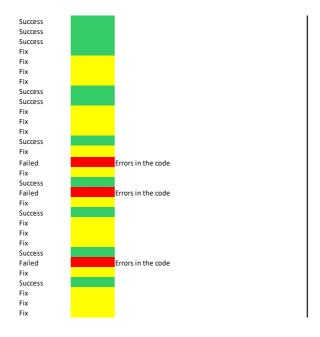
Class diagram

Data input

As it is not an application where there are relations between different classes, a class diagram will not be attached.

Test cases and Test Data

t	topTable&ExpMat i	illes (.csv) 2- The actor has to upload Express&t	topfiles. (1)upload ExpressAndTop.csv (2) upload myfile.txt (3) upload myfile2.pdf (4) upload myfile3.odt (5) file not load	Success Error: Not valid format Error: Not valid format Error: Not valid format Error: No load file
	Success Fix Fix Fix Fix	errors with t	the format	
	Filters from the top table	The actor chooses filtering the topTable by p-value (<0.05 or <0.01), and/or logFC (<-1,>1)	(1) Applying filters with a topTable file lo (2.0) no filter choosen (2.1) logFC>1 (2.2) logFC<-1 (2.3) logFC=5 (2.4) logFC=a (2.5) logFC=% (2.6) p-value<0.01 (2.7) p-value<10.05 (2.8) p-value=1 (2.9) p-value=a (3) Applying filters without a topTable file	Success: we do not want to apply any filter Success Success Error: filter not valid Error: filter not valid Error: filter not valid Success Success Error: filter not valid Error: filter not valid
	Selected genes from the topTable	According the filters selected previously, the app generates a new table	(4) Table appears with filters applyed (5) Table appears without filters applyed (6) No table appears (7) No table appears because any file has be	Success Error: filters has not been applyed correctly Error: An error has ocurred
	Volcano plot	According the selected genes table, a volcano plot will be generated.	(8) Volcano plot appears (9) Volcano plot not appears (10) Volcano plot not appears because any fi	Success Error: An error has ocurred
	Number of genes filtered for the volcano plot	The actor chooses the number of genes he/she wants to be plotted	(11) numGenes=1 (12) numGenes=% (13) numGenes=-1 (14) numGenes=a	Success Error: filter not valid Error: filter not valid Error: filter not valid Error: filter not valid
	Heatmap	Acording the expression matrix loaded, a heapmap will be generated	(15) Heapmap appears (16) Heapmap not appears (17) Heapmap not appears because any file ha	Success Error: An error has ocurred asError: File not loaded
	Number of genes filtered for the heatmap	The actor chooses the number of genes he/she wants to be plotted	(18) numGenes=1 (19) numGenes=% (20) numGenes=-1 (21) numGenes=a	Success Error: filter not valid Error: filter not valid Error: filter not valid



Functional Analysis

GO Analysis

According the selected genes table, a GO analysis table will be generated.

According the selected genes table, the GO analysis plots will be generated.

(1) A results table appears Success

(2) A results table not appears Error: An error has ocurred

(3) A results table not appears because the :Error: An error has ocurred (4) A results table not appears because any :Error: File not loaded

(5) The results plots appear Success

(6) The results plots not appear Error: An error has ocurred

(7) The results plots not appear because the Error: An error has ocurred

(8) The results plots not appear because any ${\it Error: File\ not\ loaded}$



UEB TOOLS TEST CASE REPORT

Name: TC01-Input Data Creator: Elisabet Aguayo Version: 28/04/2019

Description/purpose: User loads files **Actor:** Researcher who uses the app

Test Data: topTable files, expression matrix files **Precondition:** Both files must have .csv format

Dependencies: None

Steps:

- 1-The actor visits the input data page
- 2-The actor loads the topTable and/or expression matrix files
- 3- The actor clicks on the submit button

Expected result:

- 1-The files are loaded correcty
- 2-The files can be read and its information can be processed

Condition of entry that discusses	Classes Valid for the condition analyzed	Classes not Valid for the condition analyzed
TopTable & ExpMat files	.csv2 files (2)	Insert .txt files (3) Insert .pdf files (4) Insert .odt files (5) Insert .doc files (6)

Valid proba case TC-01 Input Data	Classes for the condition analyzed	Result
.csv	(1)	Success
.csv2	(2)	Success
.txt	(3)	Error: Not valid format.
.pdf	(4)	Error: Not valid format.
.odt	(5)	Error: Not valid format.
.doc	(6)	Error: Not valid format.

Name: TC02-Graphical Analysis Creator: Elisabet Aguayo Version: 28/04/2019

Description/purpose: System generates different tables and plots according the input files

Actor: Researcher who uses the app

Test Data: filters for the topTable, selected genes from the topTable, volcano plot, number of genes filtered

for the volcano plot, heatmap, number of genes for the heatmap

Precondition: Files must be loaded previously

Dependencies: None

Steps:

filters for the topTable

- 1-The actor visits the graphical analysis page
- 2-The actor choose the filters he/she wants to apply to the topTable data
- 3- The actor clicks on the submit button

selected genes from the topTable

1- According the filters selected previously, the systems generates a new table

volcano plot

1-According the selected genes table, a volcano plot 1- the volcano plot is generated correctly will be generated.

number of genes filtered for the volcano plot

- 1-The actor choose the number of genes he/she wants to apply to the topTable data
- 2- The actor clicks on the submit button

heatmap

1-According the expression matrix table, a heatmap 1- the heatmap is generated correctly will be generated.

number of genes filtered for the heatmap

1-The actor choose the number of genes he/she wants to apply to the expression matrix table data

Expected result:

filters for the topTable

- 1-The filters are applyed correcty
- 2-The selected genes table will have different view according to the filters applied

selected genes from the topTable

- 1- the new table are generated correctly
- 2- the new generated data are available to be processed

volcano plot

number of genes filtered for the volcano plot

- 1-The filter is applyed correcty
- 2-The volcano plot will have different view according to the filter applied

heatmap

number of genes filtered for the heatmap

- 1-The filter is applyed correcty
- 2-The heatmap will have different view according to

Condition of entry that discusses	Classes Valid for the condition analyzed	Classes not Valid for the condition analyzed
Filters for the topTable	LogFC >1 (1) LogFC <-1 (2) p-value<0.05 (3) p-value<0.01 (4)	LogFC =1 (5) LogFC =a (6) LogFC =% (7) p-value=1 (8) p-value=a (9) p-value=% (10)
Selected genes table	Table appears with filters applied(11)	Table appears without filters applied(12) No table appears(13) No table appears because any file has been loaded(14)
Volcano Plot	Volcano plot appears (15)	Volcano plot not appears (16) Volcano plot not appears because any file has been loaded(17)
Filter of the number of genes for the volcano plot	NumGenes=1 (18)	NumGenes=-1 (19) NumGenes=a (20) NumGenes=% (21)
Heatmap	Heatmap appears (22)	Heatmap not appears (23) Heatmap not appears because any file has been loaded(24)
Filter of the number of genes for the heatmap	NumGenes=1 (25)	NumGenes=-1 (26) NumGenes=a (27) NumGenes=% (28)

Valid proba case TC-02 Graphical Analysis	Classes for the condition analyzed	Result
LogFC >1	(1)	Success
LogFC <-1	(2)	Success
p-value<0.05	(3)	Success
p-value<0.01	(4)	Success
LogFC =1	(5)	Error: Not valid format.
LogFC =a	(6)	Error: Not valid format.
LogFC =%	(7)	Error: Filter not valid.
p-value=1	(8)	Error: Filter not valid.
p-value=a	(9)	Error: Filter not valid.
p-value=%	(10)	Error: Filter not valid.
Table appears with filters applied	(11)	Success
Table appears without filters applied	(12)	Error: filters has not been applyed correctly
No table appears	(13)	Error: An error has occurred
No table appears because any file	(14)	Error: File not loaded

has been loaded		
plot appears	(15)(22)	Success
plot not appears (16)	(16)(23)	Error: An error has occurred
plot not appears because any file has been loaded	(17)(24)	Error: File not loaded
NumGenes=1	(18)(25)	Success
NumGenes=-1	(19)(26)	Error: Filter not valid.
NumGenes=a	(20)(27)	Error: Filter not valid.
NumGenes=%	(21)(28)	Error: Filter not valid.

Name: TC03-Functional Analysis

Creator: Elisabet Aguayo **Version:** 28/04/2019

Description/purpose: System generates different analysis according the input files

Actor: Researcher who uses the app

Test Data: Go Analysis, GSEA, Veen Diagram **Precondition:** Files must be loaded previously

Dependencies: None

Steps: GO Analysis	Expected result: GO Analysis
1- According the selected genes table, a GO analysis is generated	

Condition discusses	of	entry	that	Classes Valid for the condition analyzed	Classes not Valid for the condition analyzed
GO				GO analysis table appears(1)	GO analysis table/plot not appears (2) GO analysis table/plot not appears because the selected genes table has not been generated (3) GO analysis table/plot not appears because any file has been loaded(4)

Valid proba case TC-03 Functional Analysis	Classes for the condition analyzed	Result
Plot/table results appears	(1)(5)(8)	Success
Plot/table results not appears	(2)(6)(9)	Error: An error has occurred
Plot/table results not appears because the selected genes table has not been generated		Error: An error has occurred
Plot/table results not appears because any file has been loaded	(4)(7)(11)	Error: File not loaded

<u>Description of methodology used and project planning (Agile or classic approaching with GANTT: dates, tasks and resources)</u>

I used classic methodologies with GANTT to plan the project.

It is defined in the following points to develop in 4 phases of the project (4 sprints):

Sprint 1

- Bibliographical review (February 1 to February 8)

An extensive bibliographic review has been carried out on analysis of microarrays and pipelines of R to understand its operation to carry out the post-analyzes requested.

- Definition of application parameters (February 11 to February 13)

I have defined what parameters can be controlled by the user to be able to redefine the analyzes again and make them based on these chosen parameters.

Sprint 2

- Layout design of the web application (February 14 to February 21)

The distribution of the web application in Shiny has been theoretically designed.

- Definition of technical aspects of the web application (February 21 to March 20)

Each action of the application has been defined, the necessary files must be selected to test the operation, and the necessary R packages must be defined to carry out the post-analyzes.

Sprint 3

- Schedule the application (March 20 to May 15)

The script in R must be developed based on the characteristics defined above.

- Programming script to the web application (March 20 to May 15)

All the components that will be necessary to create an application in Shiny of the designed application must be programmed.

Sprint 4

- Proof of robustness (May 15 to May 20)

You must verify locally that the application works correctly with the selected data.

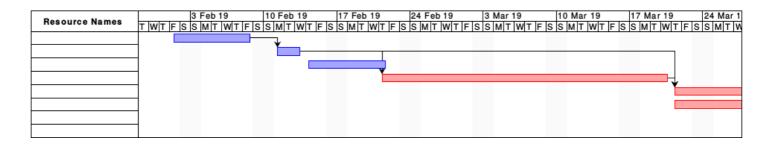
- Deployment on the server (May 20 to May 28)

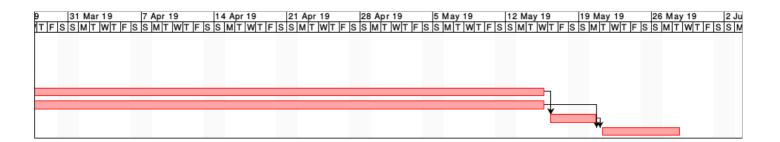
The application must be deployed on the free server that has R-Studio for Shiny.

Trello has been used as project management software to manage sprints.

Project memo: initial planning, diary log, problems and solutions, final conclusions

	(1)	Name	Duration	Start	Finish	Predecessors
1	o	Revisió bibliogràfica	5.875 da	2/1/19 9:00 AM	2/8/19 5:00 PM	
2	Ö	Definició de paràmetres de l'aplicació	2.5 days?	2/11/19 8:00 AM	2/13/19 1:00 PM	1
3		Disseny layout	6 days?	2/14/19 8:00 AM	2/21/19 5:00 PM	
4		Definició aspectes tècnics	20 days?	2/21/19 8:00 AM	3/20/19 5:00 PM	2
5	o i	Programar aplicació	40 days?	3/21/19 8:00 AM	5/15/19 5:00 PM	2;4
6	O	Implementar script	40 days?	3/21/19 8:00 AM	5/15/19 5:00 PM	
7	Ö	Prova robustesa	3 days?	5/16/19 8:00 AM	5/20/19 5:00 PM	5;6
8		Desplegament en servidor	6 days?	5/20/19 5:00 PM	5/28/19 5:00 PM	6;7





The main problem I have had in the whole project is, on the one hand, the fact that I did not know anything about microarrays so far, and when Alex Sánchez told me about this proposal of designing an application of microarrays post-analysis, I did not know how start. For this reason, I started designing the application without knowing if those I was doing was correct.

On the other hand, I had never worked with Shiny and Bioconductor, and I had to learn by myself.

As the application is not very big, I have finished the most I had proposed, but I started to really understand about all at the end of April, for this reason this is not as good as I would like that.

The unique functionality that it lacks it is the GSEA analysis, because I do not understand it and I have not had any time to implement it.

Additionally, I have not deployed the application to the server because, as is an application from a private company, they have not asked to me anything about that, and I do not want to do anything without permission.

Correspondence with initial functionalities

All functionalities respond as expected initially, except for GSEA, that is missing because of lack of time and its complexity.

Innovation

To do this application I have had to learn about R, Bioconductor, Shiny and Microarrays, all in a self-taught way. In general, I am happy with it and proud of myself..

Installation Guide

To run this application is necessary to install the RStudio IDE 1.1.463 with the R 3.5.1 version. Moreover, you must install these packages: DT, shinycssdownloaders, RColorBrewer, colourpicker, gplots, ggplot2, calibrate, xtable, digest.

On the other hand, you must install Bioconductor 3.8 version with these bioconductor packages: limma, GOfuncR, TxDb.Hsapiens.UCSC.hg19.knownGene, Homo.sapiens, stringi, ggrepel.

Use of OOP style: encapsulation, inheritance and polymorphism

In this case, in R all are objects. Despite, the OOP style in R is not the same that in other programming languages.

Software architectural pattern used

I used the two scripts structure: ui.R for the views and server.R for the controllers. I think this is what mvc looks like the most.

Internal documentation

You can see the internal documentation in the attached code.

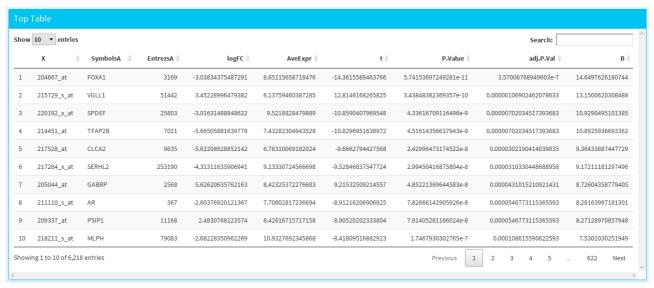
Acquired knowledge related to the professional cycle

- Development of interactive web applications
- Development of web applications embedded in trademark languages
- Elaboration of the documentation of the web application
- Installation, configuration and use of a version control system
- Verification of the execution of web applications
- Verification of the levels achieved through the use of external tests
- Verification of the visualization of the interface with different browsers and technologies.
- Modification of the web interface to match the target you are pursuing and the users you are targeting.
- Verification of the ease of navigation of a web document through different peripherals.
- Development of programs for data processing in the field of bioinformatics.
- Analysis of requirements according to the needs of the client / user of the application.
- Planning the development of the application, both with regard to the technical aspects of design, as well as its sequencing and timing.
- Development of the development report and documentation for implementation and configuration of the application, as well as user guides.

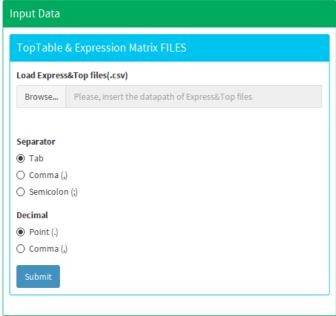
Technical knowledge acquired

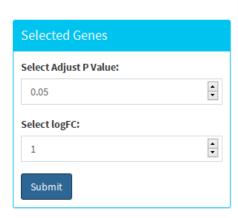
R, Shiny, Bioconductor, Microarrays Analysis

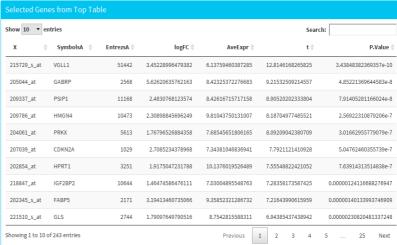
Screenshots of the final application

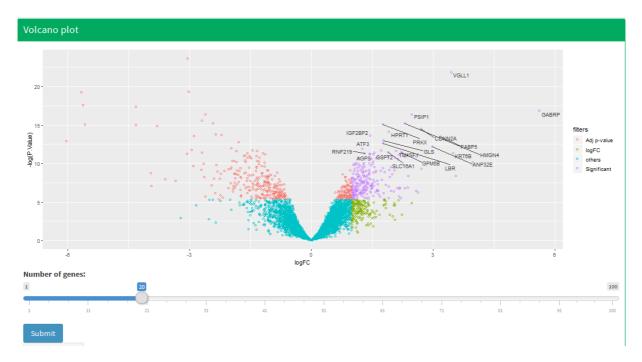


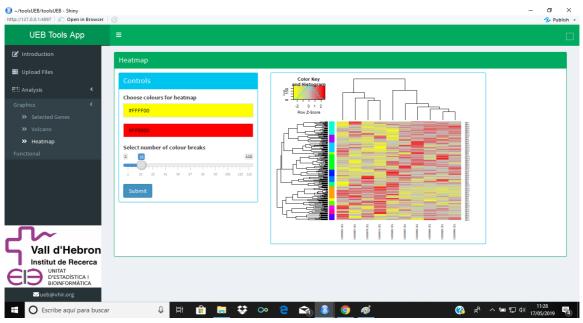












10	▼ entries			Search:	
	go_id	♦ gene			
L	GO:0003824	AACS	catalytic activity	molecular_function	
2	GO:0003824	ABCA12	catalytic activity	molecular_function	
3	GO:0003824	ABCC2	catalytic activity	molecular_function	
ı	GO:0003824	ABHD11	catalytic activity	molecular_function	
5	GO:0003824	ACAD8	catalytic activity	molecular_function	
5	GO:0003824	ACOX2	catalytic activity	molecular_function	
7	GO:0003824	ACOX3	catalytic activity	molecular_function	
3	GO:0003824	ACSF2	catalytic activity	molecular_function	
)	GO:0003824	AKR1A1	catalytic activity	molecular_function	
10	GO:0003824	AKR1B10	catalytic activity	molecular_function	

