

# Identifying proteins and metabolic pathways associated to the neuroprotective response mediated by tibolone in astrocytes under an induced inflammatory model

Presented by:

**Daniel Camilo Osorio Hurtado**

in partial fulfillment of requirements for the degree of

**Master in Bioinformatics**

Advisors: **Janneth Gonzalez PhD.** and **Andrés Pinzon PhD.**  
Bioinformatics and Computational Systems Biology Lab



**Universidad Nacional de Colombia**  
**Engineering Faculty - Department of Systems and Industrial Engineering**  
**Bogotá, Colombia**

## Objectives:

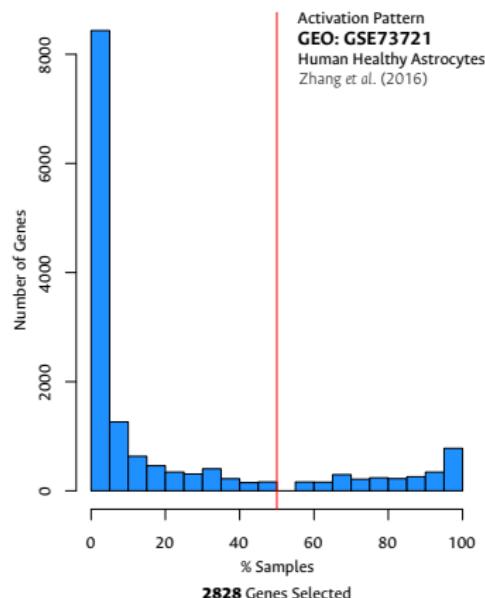
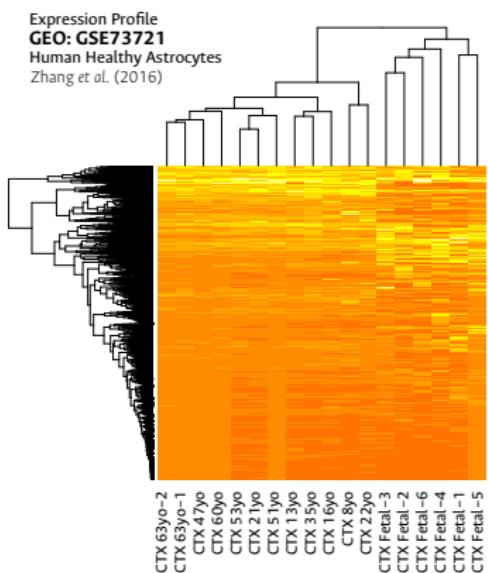
To identify proteins and metabolic pathways involved in the neuroprotective effects of tibolone in human astrocytes based in metabolic scenarios comparation we set:

- ▶ Build a tissue specific computational model of astrocytes metabolism using gene expression data integration.
- ▶ Evaluate the effects caused by the increase of free fatty acids and tibolone presence in astrocytes metabolism.
- ▶ Determine metabolic pathways and relevant functional products in response to steroid tibolone through systems biology approximations.
- ▶ Evaluate the importance of proteins and metabolic pathways previously identified on the dynamics of the metabolic model.

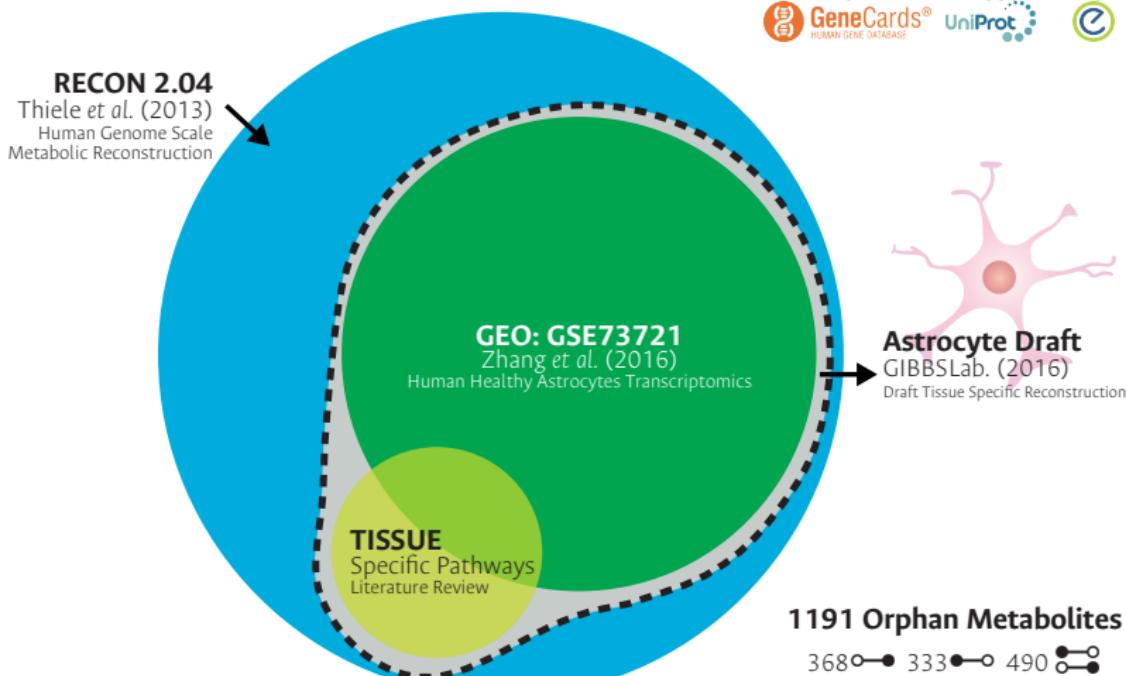
## OBJECTIVE 1:

Build a tissue specific computational model of astrocytes metabolism using gene expression data integration.

# Healthy Human Astrocytes Gene Expression Data



# Mapping Reactions



# Gap-Find and Gap-Fill Available Algorithms

ALGORITHM	ENVIRONMENT	HOW IT WORKS
SMILEY	Python - OpenSource	<ul style="list-style-type: none"><li>Optimization based.</li><li><b>Fills one metabolite per time.</b></li></ul>
gap-Find/Fill	GAMS - OpenSource	<ul style="list-style-type: none"><li>Optimization based.</li><li><b>Makes several intra model modifications.</b></li></ul>
growMatch	Python - OpenSource	<ul style="list-style-type: none"><li>Optimization based.</li><li><b>Fills one objective function per time.</b></li></ul>
fastGapFill	MATLAB - Privative	<ul style="list-style-type: none"><li>Optimization based.</li><li>Multiobjective.</li></ul>

# Finding and Filling Gaps



## 'g2f' Package

An R Package to Find and Fill Gaps for genome-scale metabolic networks

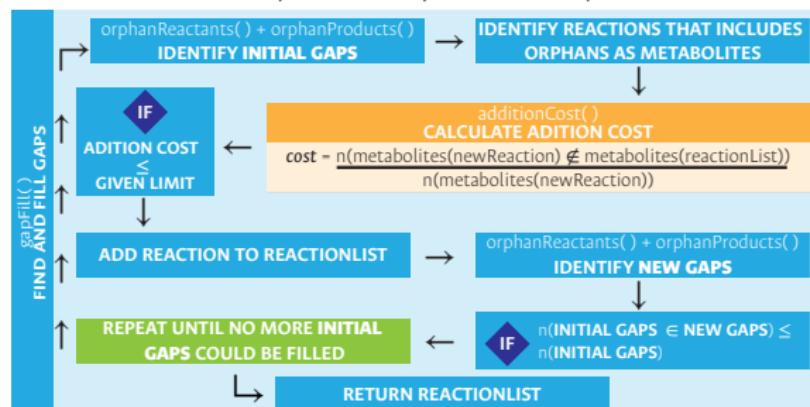
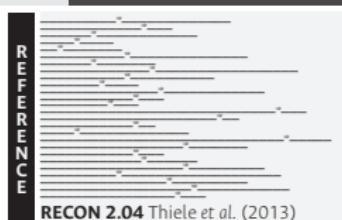
Kelly Botero, Daniel Osorio, Janneth Gonzalez and Andres Pinzón-Velasco.

Language: R  
 Stable: CRAN  
 Development: gibbslab/g2f  
 License: GPL-2  
 Binaries: Windows - Linux - Mac

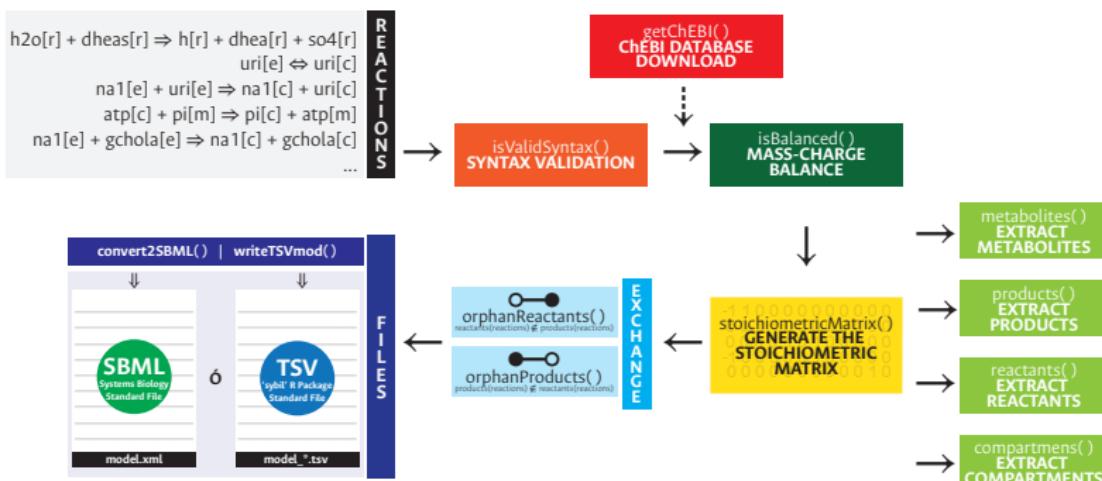
```
h2o[r] + dheas[r] ⇒ h[r] + dhea[r] + so4[r]
uri[e] ⇔ uri[c]
na1[e] + uri[e] ⇒ na1[c] + uri[c]
atp[c] + pi[m] ⇒ pi[c] + atp[m]
na1[e] + gchola[e] ⇒ na1[c] + gchola[c]
```

**ASTROCYTE DRAFT** GiBBS Lab (2016)

R  
E  
A  
C  
T  
I  
O  
N  
L  
I  
S  
T



# Syntax, Mass-Charge Validation and SBML files



# Metabolic Model Debugging



## 'g2f' Package

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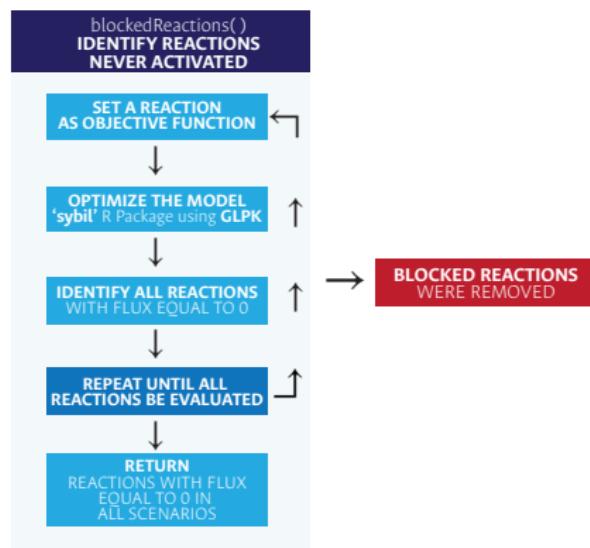
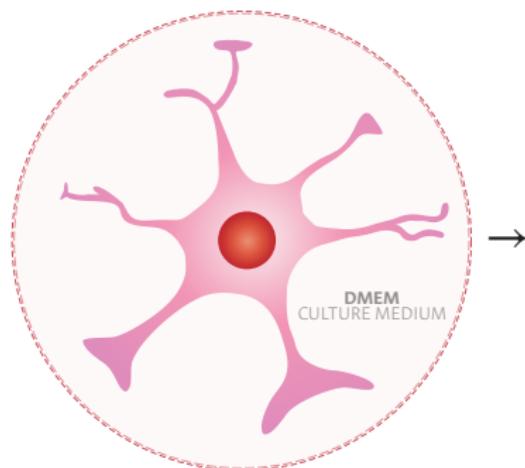
Language: R

Stable: CRAN

Development: gibbslab/g2f

License: GPL-2

Binaries: Windows - Linux - Mac



# Gene Expression Integration Available Methods

METHOD	ENVIRONMENT	HOW IT WORKS
GIMME	MATLAB <b>Privative</b>	<ul style="list-style-type: none"><li>· Binary Discretization</li><li>· Ensures flux for a selected objective function</li></ul>
iMAT	MATLAB <b>Privative</b>	<ul style="list-style-type: none"><li>· Integration proportional to gene-expression (H, M and L categorization)</li><li>· Not objective function required</li></ul>
E-FLUX	<b>Not implemented</b>	<ul style="list-style-type: none"><li>· Requires a user-given threshold</li><li>· Continuous Integration</li></ul>
PROM	MATLAB <b>Privative</b>	<ul style="list-style-type: none"><li>· Requires a user-given regulatory network</li><li>· Constraints are setting according to the associated transcript. factor</li></ul>

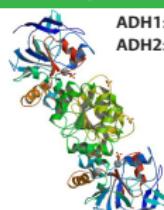
# Constraining the Metabolic Model



## 'exp2flux' Package

An R Package to convert expression data to FBA fluxes  
*Daniel Osorio, Kelly Botero, Janneth Gonzalez and Andres Pinzón-Velasco.*

E.C: 1.1.1.1



ADH1: |||||  
 ADH2: |||||



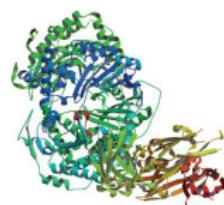
**(ADH2 or ADH1)**

sum (ExprADH2 + ExprADH1)

E.C: 3.4.21.5



IDE.A: |||||  
 IDE.B: |||||  
 IDE.C: |||||



**3.4.21.5**

**IDE.A and IDE.B and IDE.C**

min (ExprIDE.A, ExprIDE.B, ExprIDE.C)

GENE EXPRESSION  
DATA



exp2flux()  
CONVERT GENE  
EXPRESSION DATA  
TO FBA FLUXES



CONSTRAINED  
METABOLIC MODEL

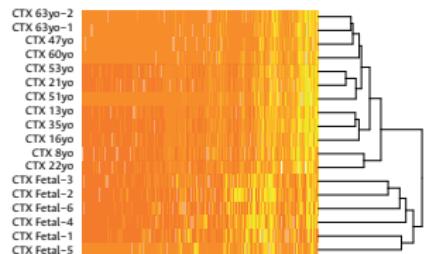


fluxDifferences()  
COMPUTE FOLDCHANGE  
OF FLUXES BETWEEN  
METABOLIC SCENARIOS

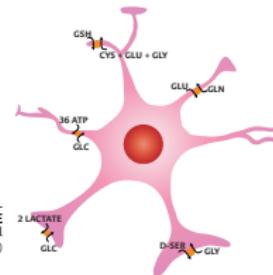
METABOLIC MODEL  
WITH GPR



# Human Healthy Mature Astrocyte Model



Expression Profile  
GEO: GSE73721  
Human Healthy Astrocytes  
Zhang et al. (2016)



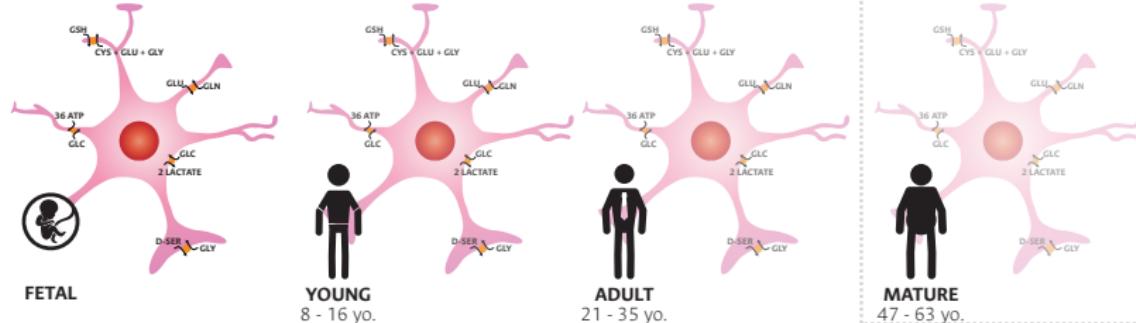
SBML  
**ASTROCYTE**  
Tissue Specific Model  
GIBBSLab. (2016)



**'exp2flux' Package**

An R Package to convert expression data to FBA fluxes  
Daniel Osorio, Kelly Botero, Janneth Gonzalez and Andres Pinzón-Velasco.

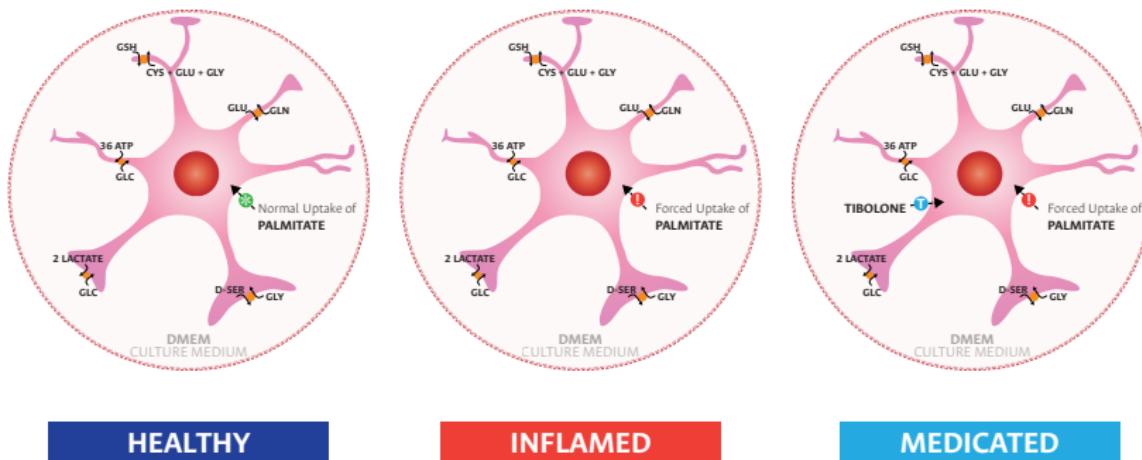
`exp2flux()`  
CONVERT GENE  
EXPRESSION DATA  
TO FBA FLUXES



## OBJECTIVE 2:

Evaluate the effects caused by the increase of free fatty acids and tibolone presence in astrocytes metabolism.

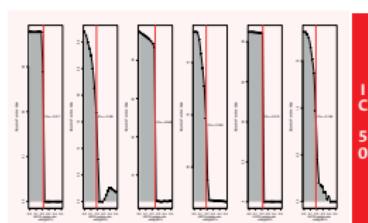
# Metabolic Scenarios



**MAIN OBJECTIVE FUNCTION:**  
Generic Human Biomass Reaction included in RECON 2.04  
(Thiele *et al.*, 2013)

# Inflamed Metabolic Scenario

Normal Uptake of PALMITATE



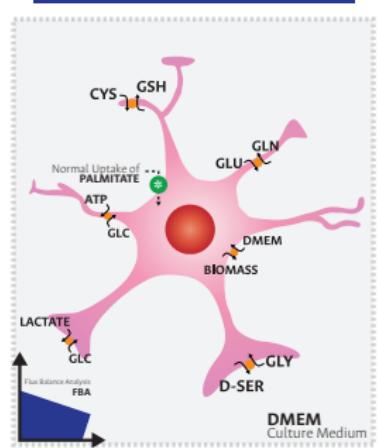
Force Uptake of PALMITATE



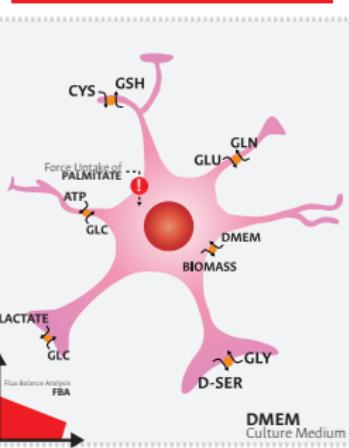
TIBOLONE



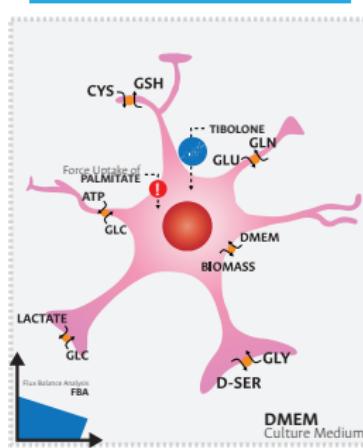
HEALTHY



INFLAMMATED

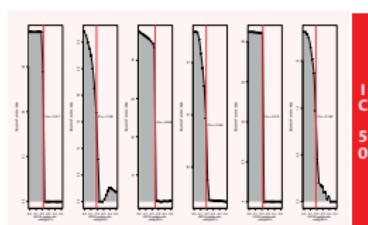


MEDICATED



# Medicated Metabolic Scenario

Normal Uptake of PALMITATE



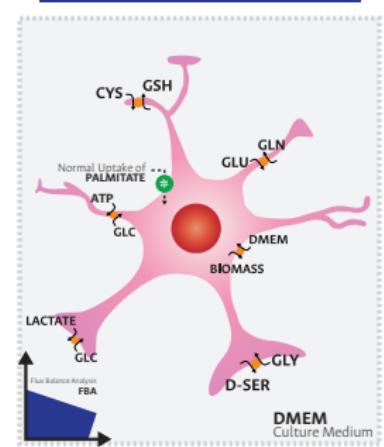
Force Uptake of PALMITATE



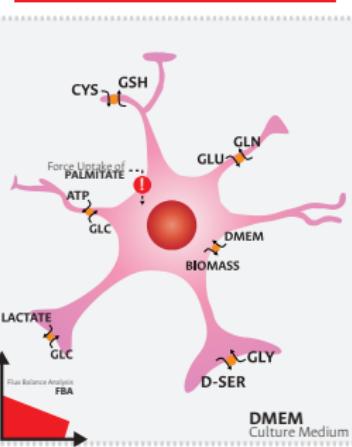
TIBOLONE



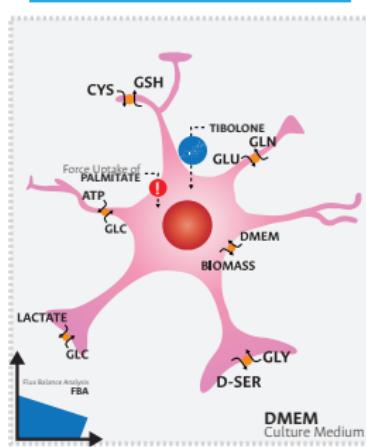
HEALTHY



INFLAMMATED



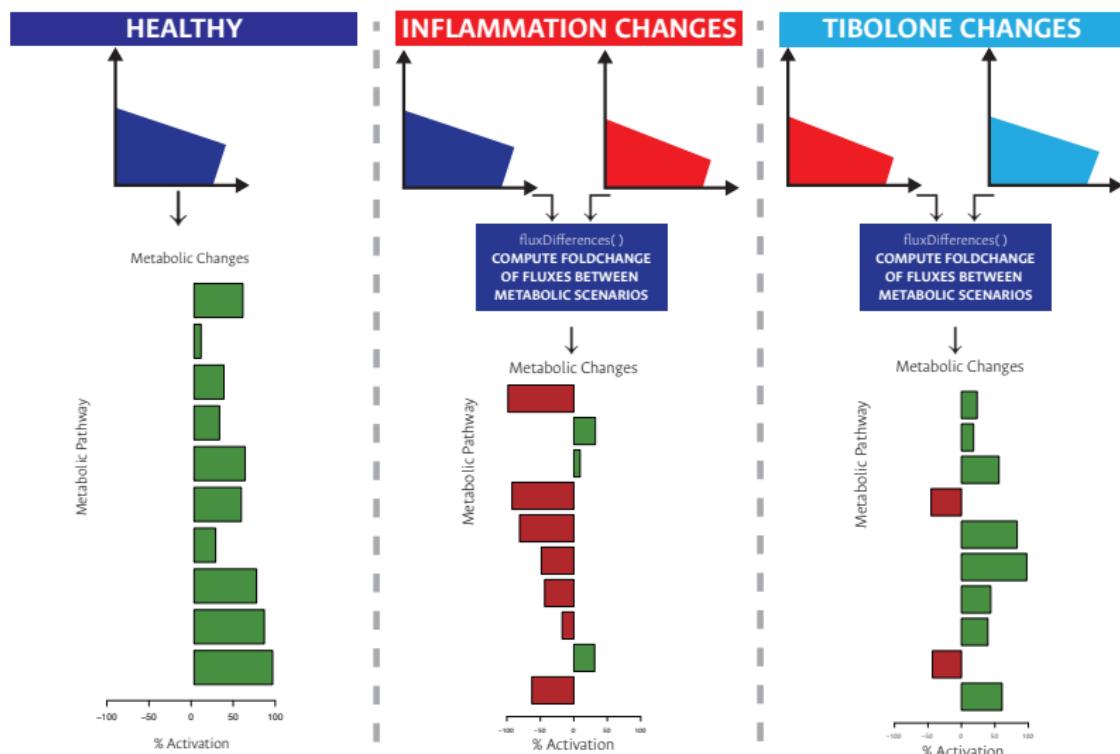
MEDICATED



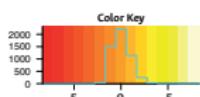
## **OBJECTIVE 3:**

Determine metabolic pathways and relevant functional products in response to steroid tibolone through systems biology approximations.

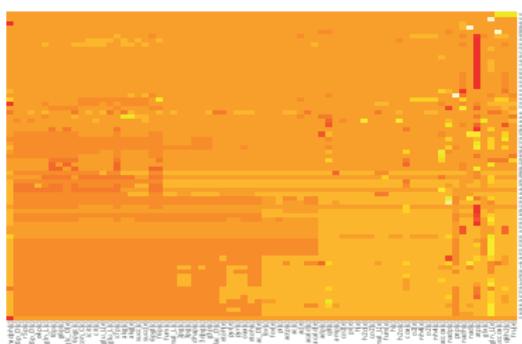
# Metabolic Pathways Activation Pattern Changes



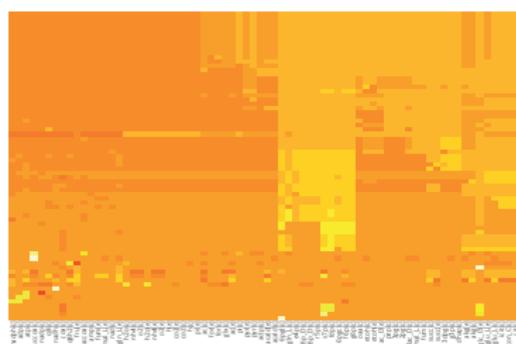
# Changes in Metabolites Production



$$\text{foldChange} = \frac{\text{Scenario2} - \text{Scenario1}}{|\text{Scenario1}|}$$



**Inflammation Related**  
Metabolic Changes



**Tibolone Related**  
Metabolic Changes

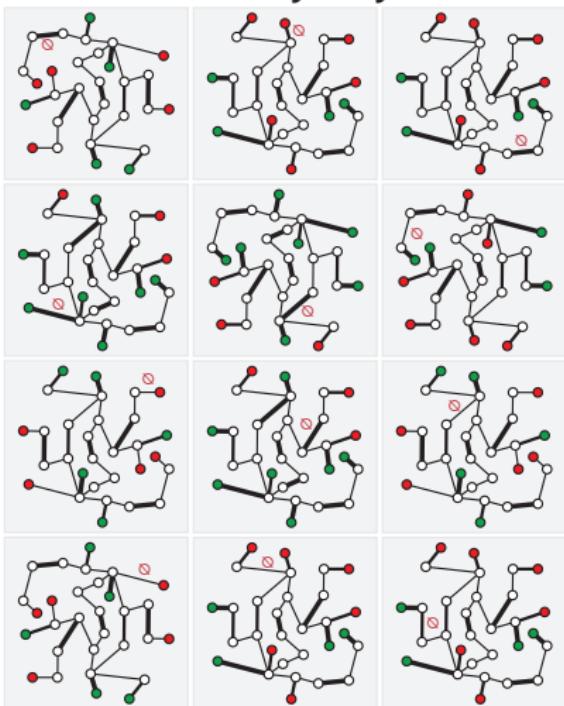
Now running at:



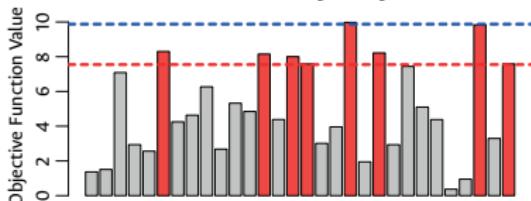
## **OBJECTIVE 4:**

Evaluate the importance of proteins and metabolic pathways previously identified on the dynamics of the metabolic model.

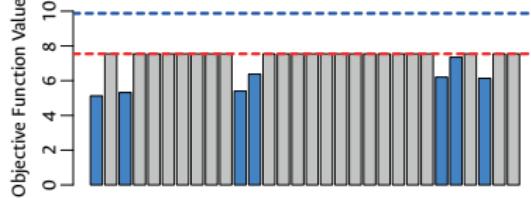
## Essentiality Analysis



### Pro-Inflammatory Enzymes



### Anti-Inflammatory Enzymes



# Software Packages



## 'g2f' Package

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Language: R  
Stable: CRAN  
Development: gibbslab/g2f  
License: GPL-2  
Binaries: Windows - Linux - Mac



## 'minval' Package

An R Package for MINimal VALidation of stoichiometric reactions  
Daniel Osorio, Janneth Gonzalez and Andres Pinzón-Velasco.

Language: R  
Stable: CRAN  
Development: gibbslab/minval  
License: GPL-2  
Binaries: Windows - Linux - Mac



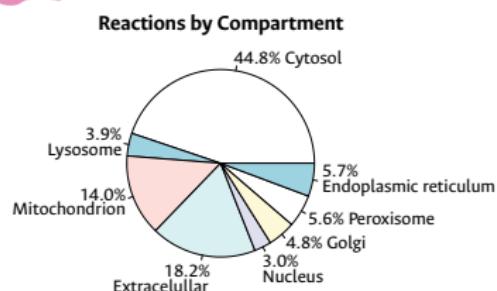
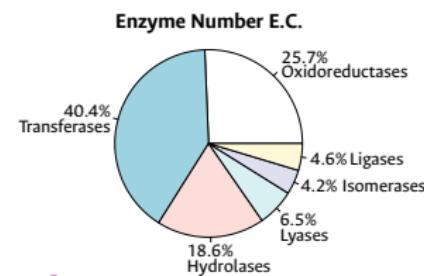
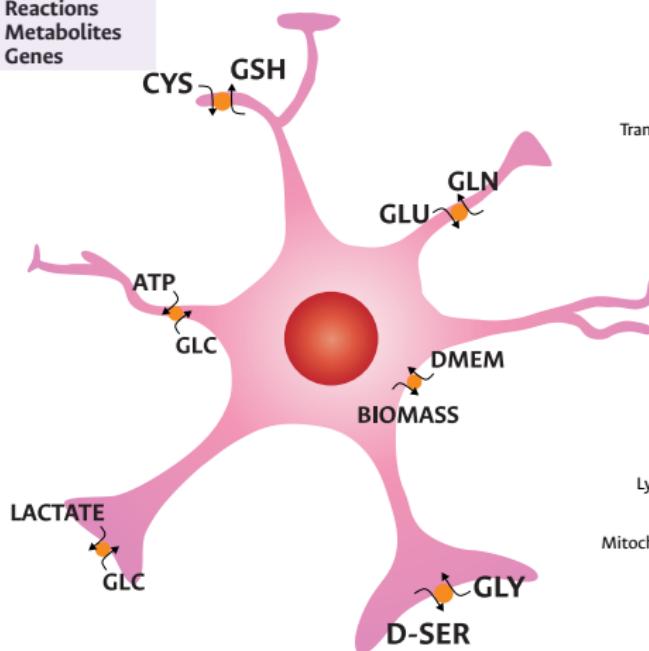
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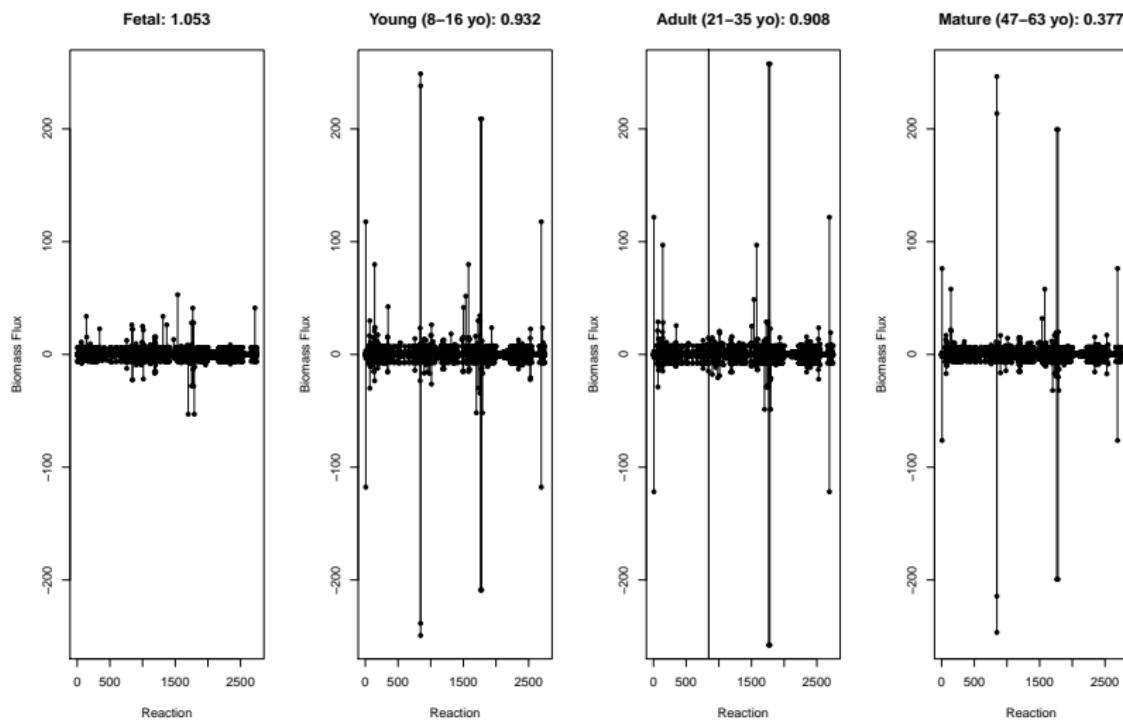
Language: R  
Stable: CRAN  
Development: gibbslab/exp2flux  
License: GPL-2  
Binaries: Windows - Linux - Mac

# Astrocyte Model

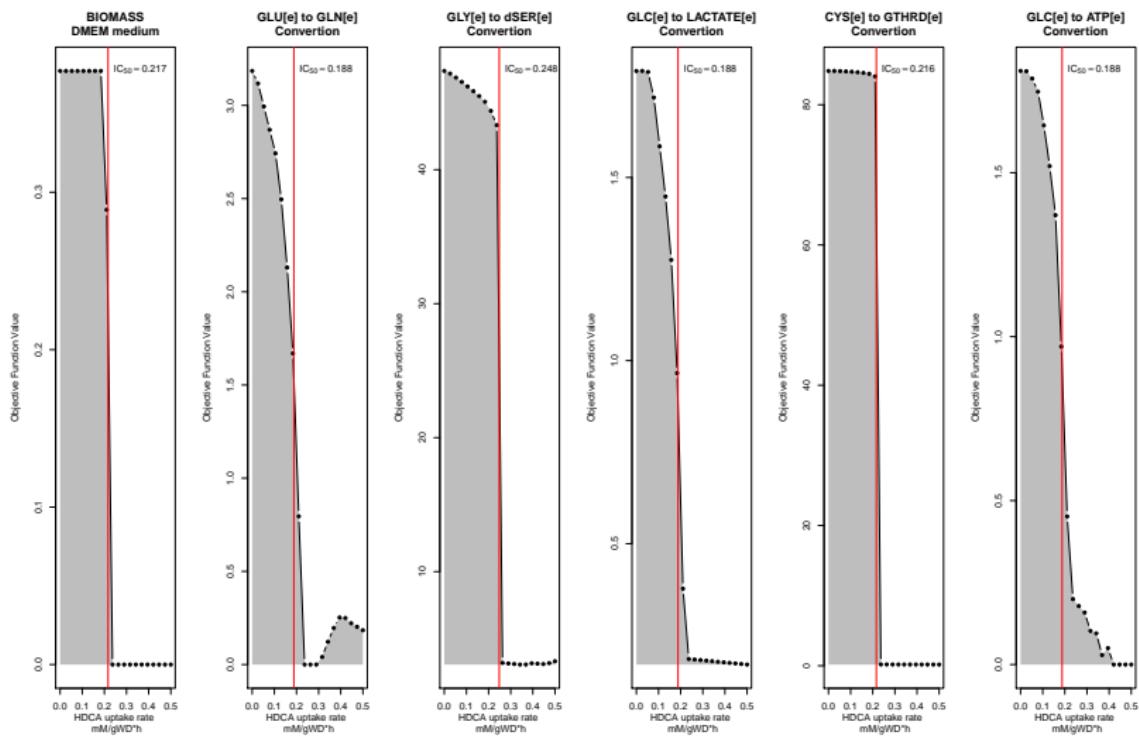
8 Compartments  
2747 Reactions  
1956 Metabolites  
1262 Genes



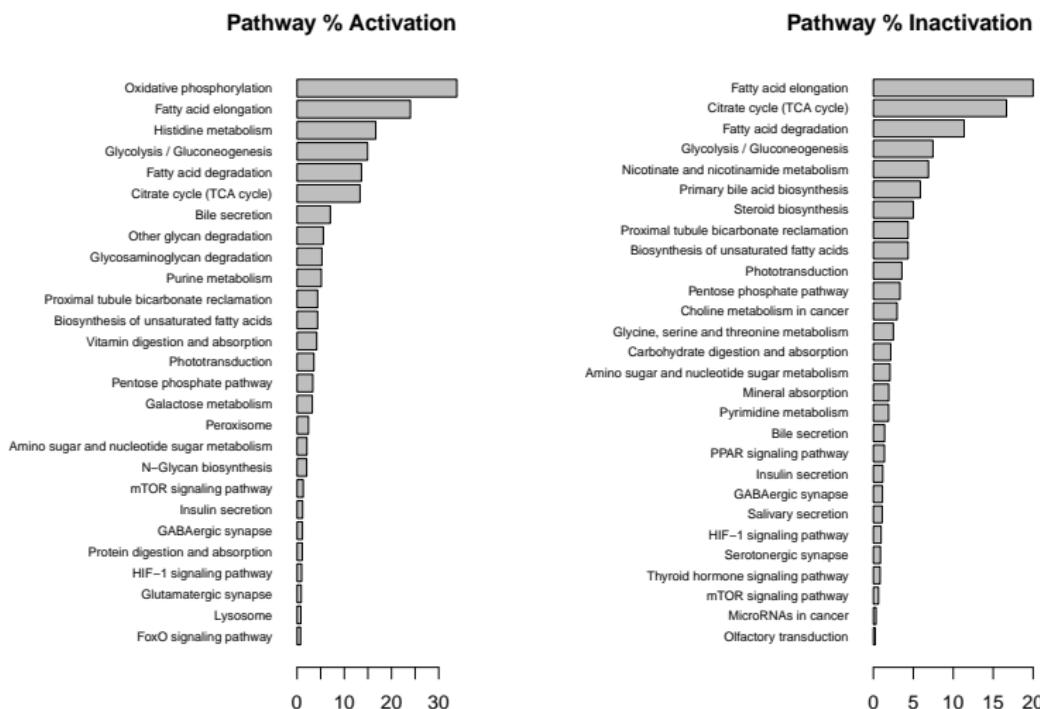
# Age Related Metabolic Changes in Astrocytes



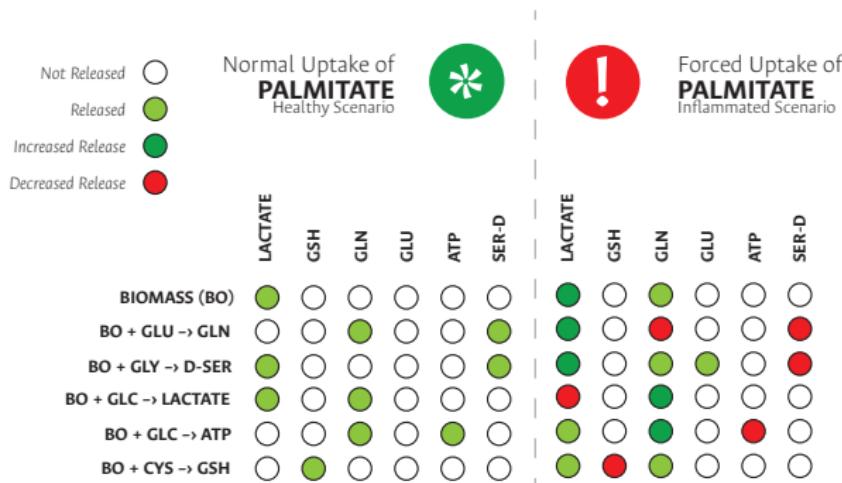
$$IC_{50} = 0.208 \pm 0.024 \text{ mMgDW}^{-1}\text{h}^{-1}$$



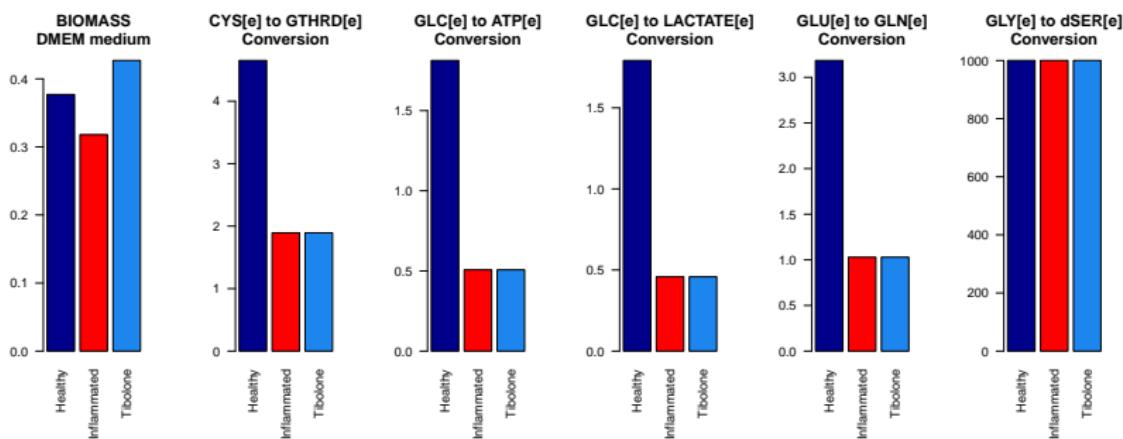
# Inflammation Related Metabolic Changes in Astrocytes



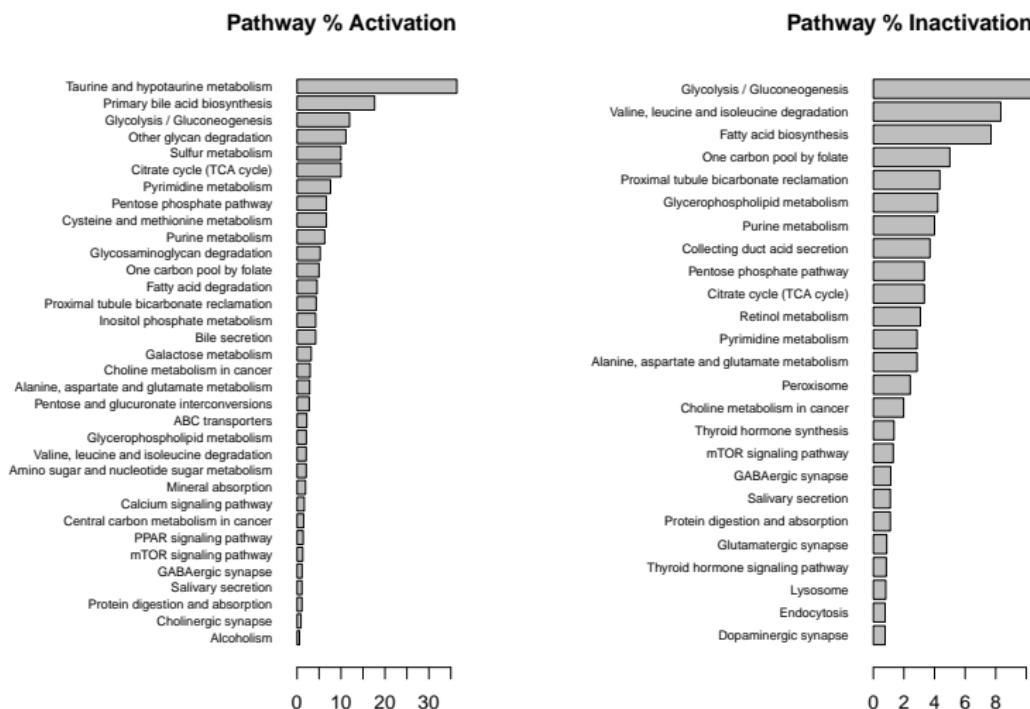
# Gliotransmitters Release Rate



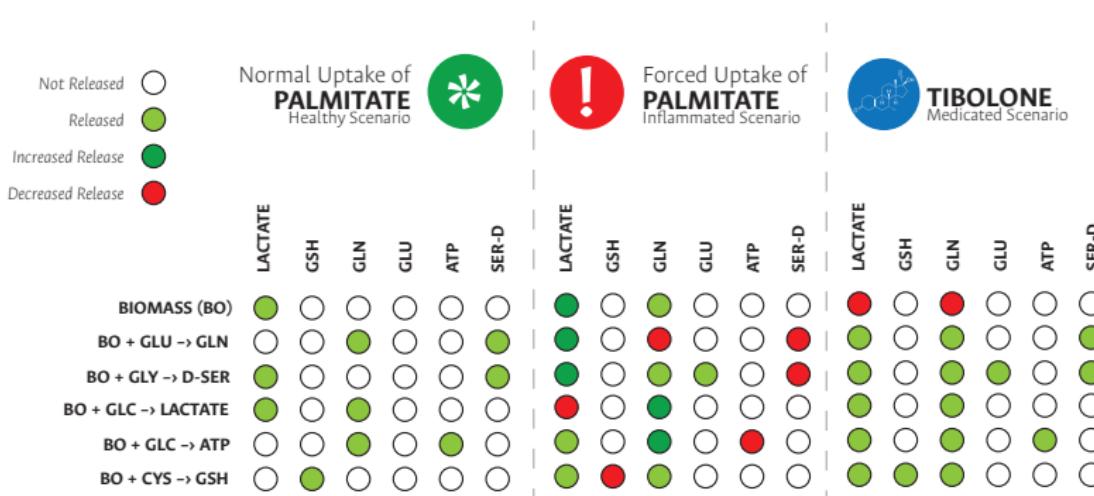
# Tibolone Effects in Inflamed Astrocytes



# Tibolone Metabolic Changes in Inflamed Astrocytes



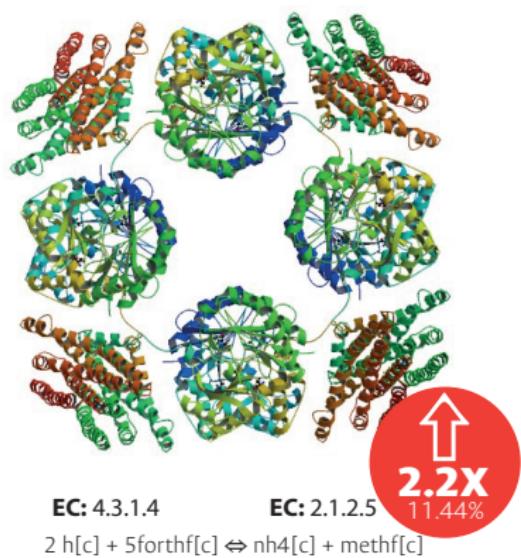
# Gliotransmitters Release Rate



# ProInflammatory Enzymes

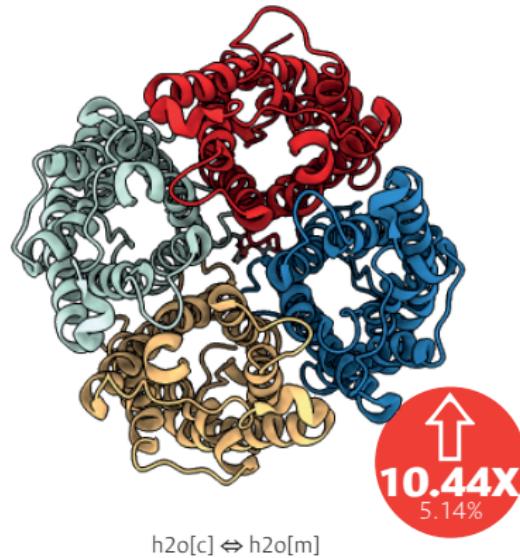
## FTCD

FormimidoylTransferase CycloDeaminase



## H2Otm

H2O Transport Mitochondrial



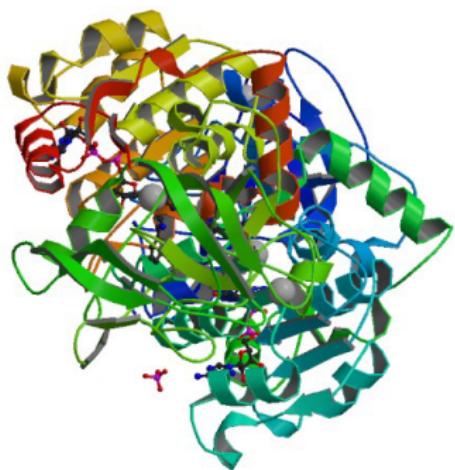
# 74 Anti inflammatory Enzymes



# Tibolone Related Enzymes

**r0739**

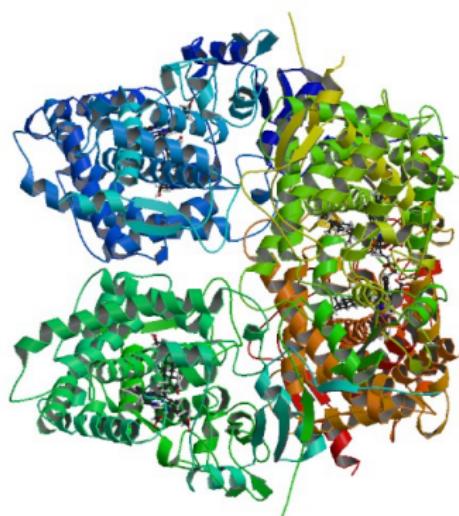
Alcohol Dehydrogenase 1 - 7



EC: 1.1.1.1

**REM1804m | REM1807m**

Cytochrome P450 Family 27 Subfamily A Member 1



EC: 1.14.15.15





# Advances of this work were presented as:

## Metabolic inflammation effects over the gliotransmitters release in mature astrocytes: a network-based approach.

Daniel Osorio MSc., Janneth Gonzalez PhD., Andrés Pinzón-Velasco PhD.  
Bioinformatics and Computational Systems Biology Lab, Universidad Nacional de Colombia.



at: \_\_\_\_\_



CDMX, México  
Short Talk



Barcelona, España  
Poster

ICGEB Course on Bioinformatics and Computational Neuroscience



Bogotá, Colombia  
Short Talk

# Acknowledgements

This study was developed at the:



## **Bioinformatics and Computational Systems Biology Lab**

Institute for Genetics - Universidad Nacional de Colombia

**CONTACT:**

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**Andrés Pinzón PhD**  
[ampinzonv@unal.edu.co](mailto:ampinzonv@unal.edu.co)