



From **physiological maps** to **disease ontology maps** using a systems biology approach

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University of Liège
Liège - Belgium



ONTOX is part of the **ASPIS cluster** that represents Europe's effort towards the sustainable, animal-free and reliable chemical risk assessment of tomorrow.



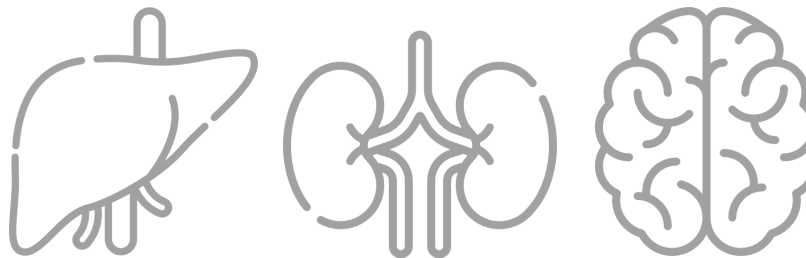
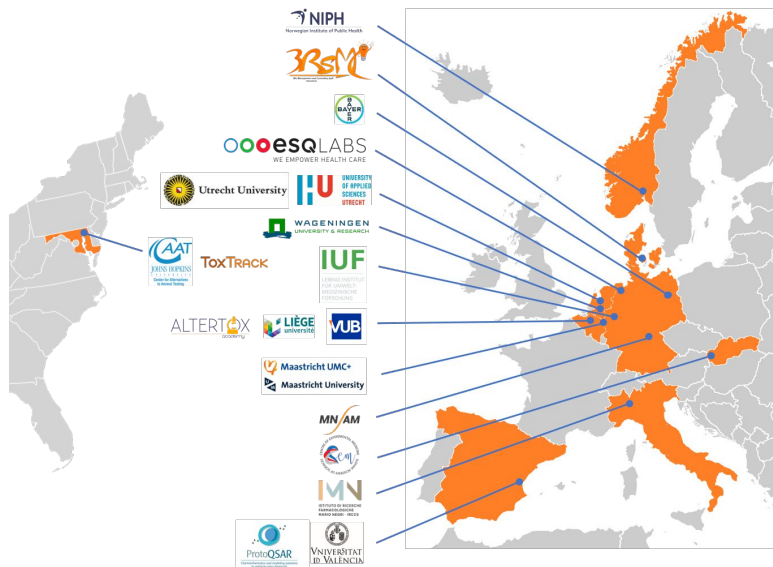
This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 963845

The ONTOX project

Ontology-driven and artificial intelligence-based repeated dose toxicity testing of chemicals for next generation risk assessment

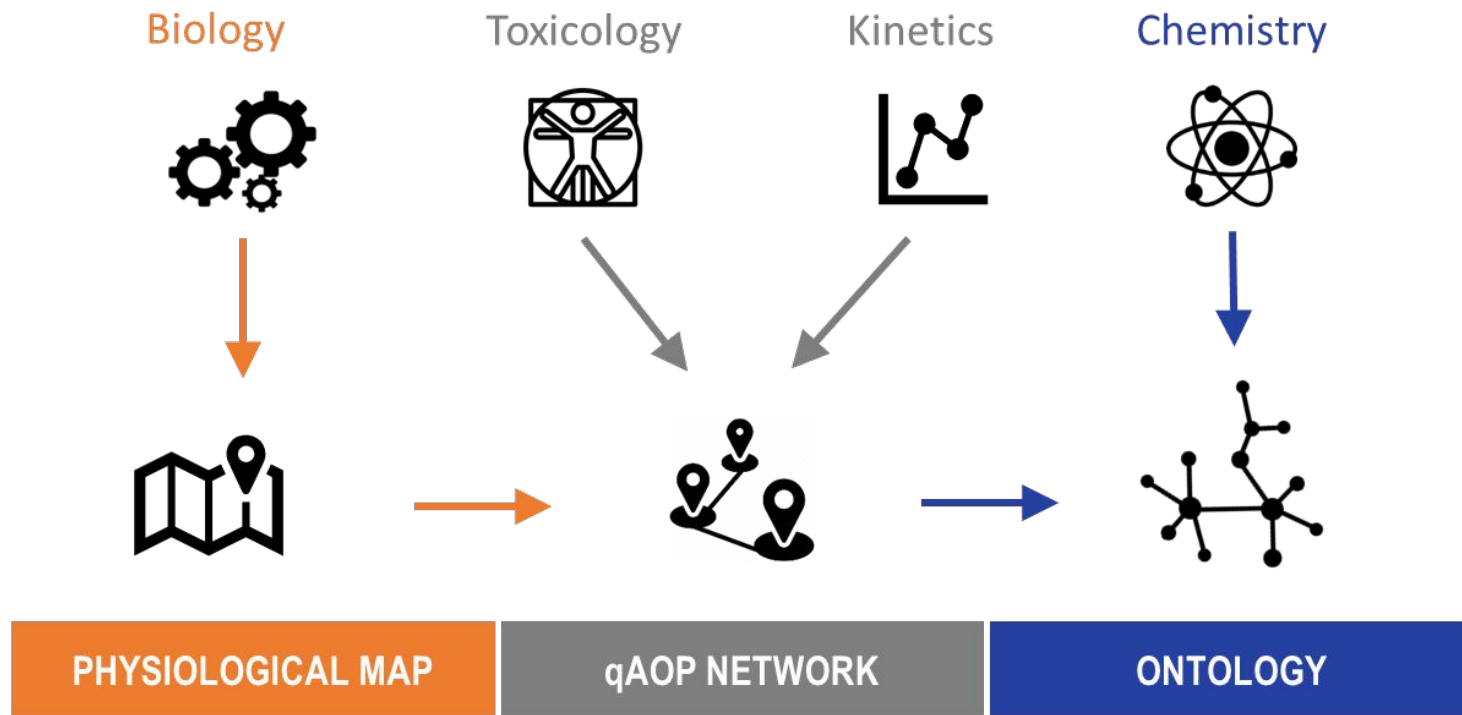
Goal: development of an animal-free and human-relevant strategy for the prediction of chemical-induced toxicity

Focus: liver, kidneys and developing brain

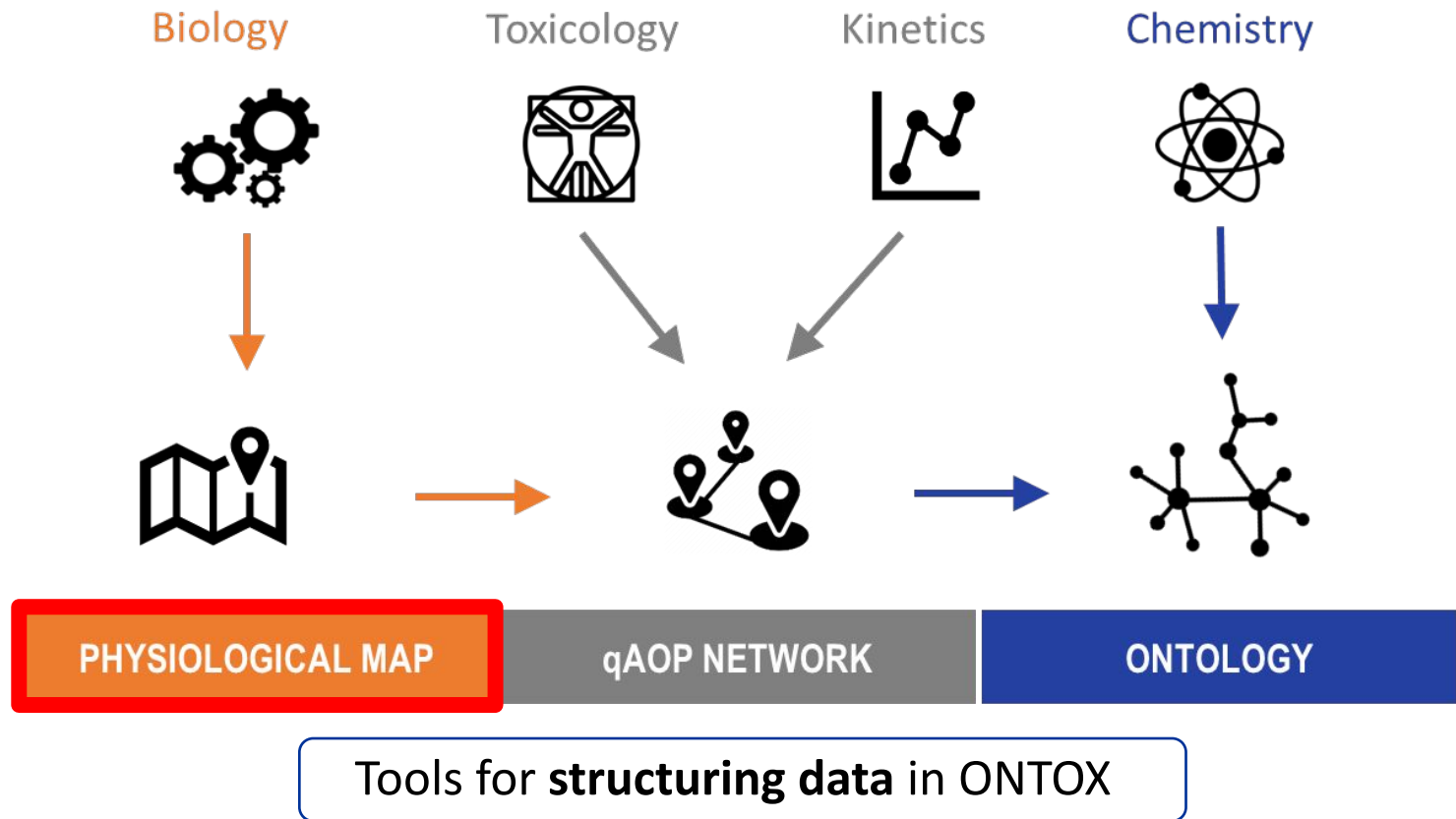


Rationale: Rely as much as possible on available data, models and methods

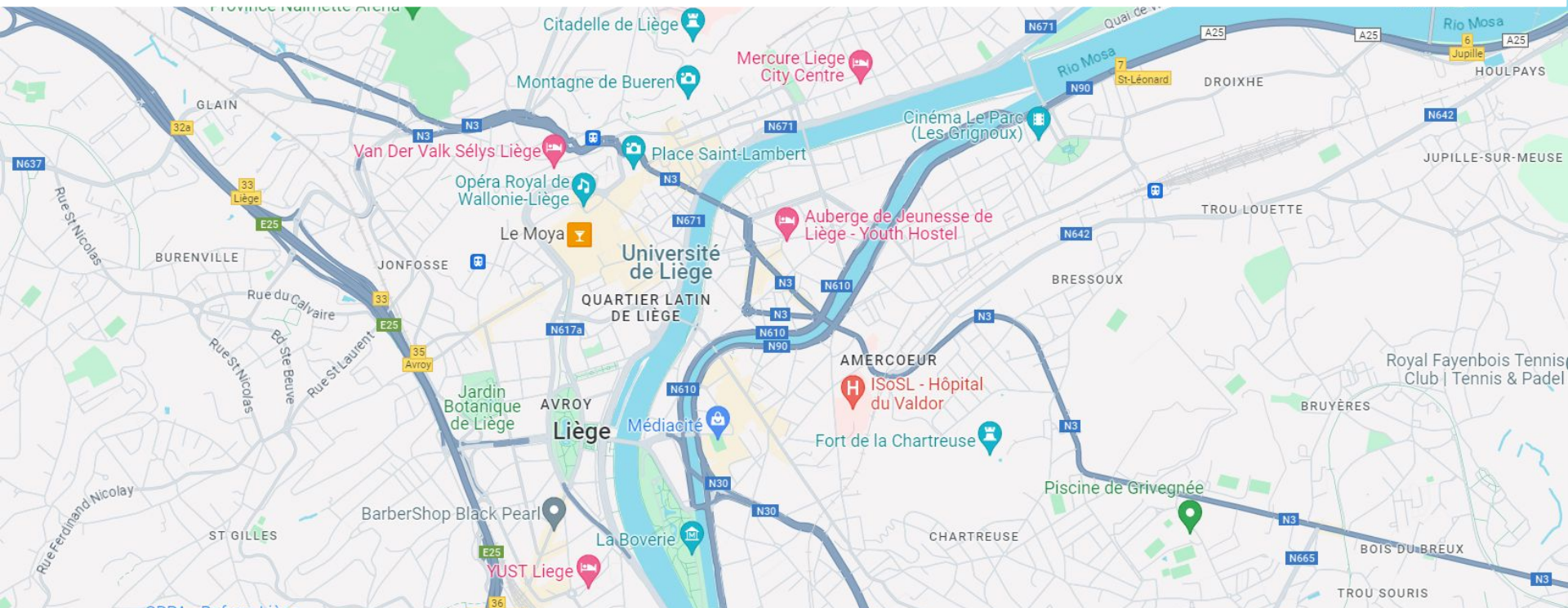
Toolbox: *in vitro* & *in silico* methods



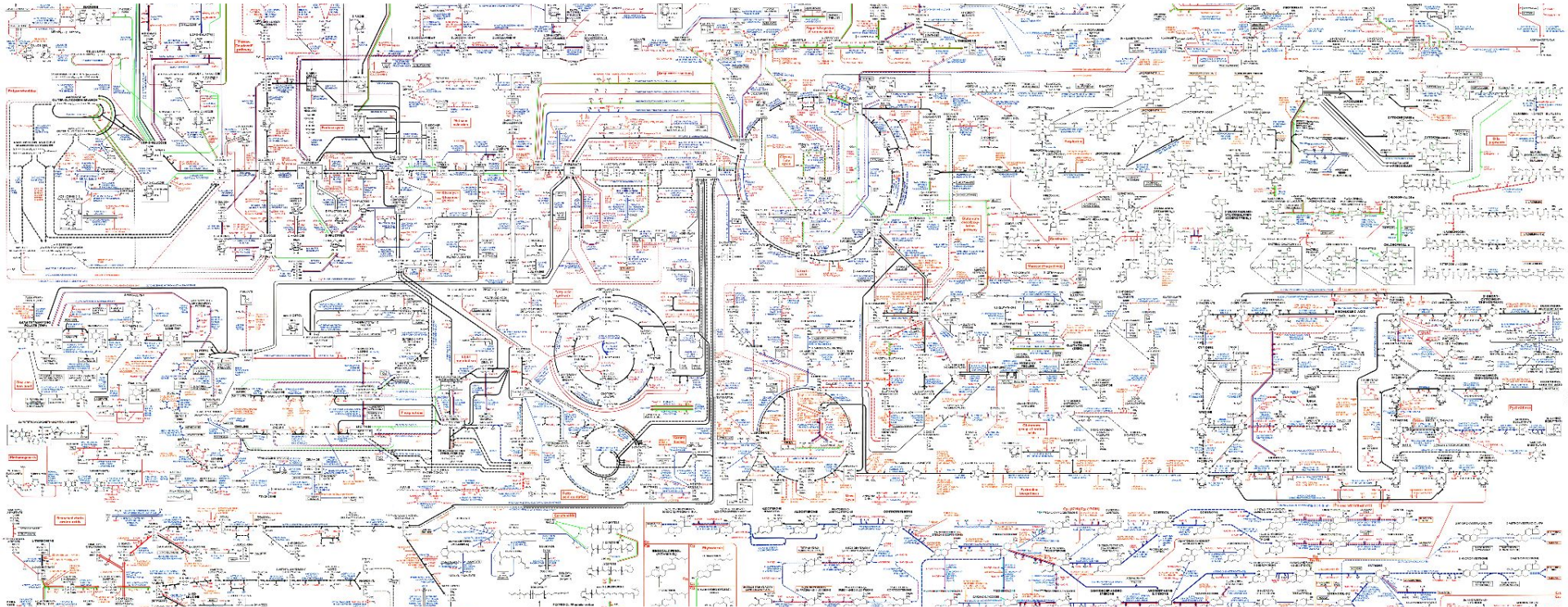
Tools for **structuring data** in ONTOX



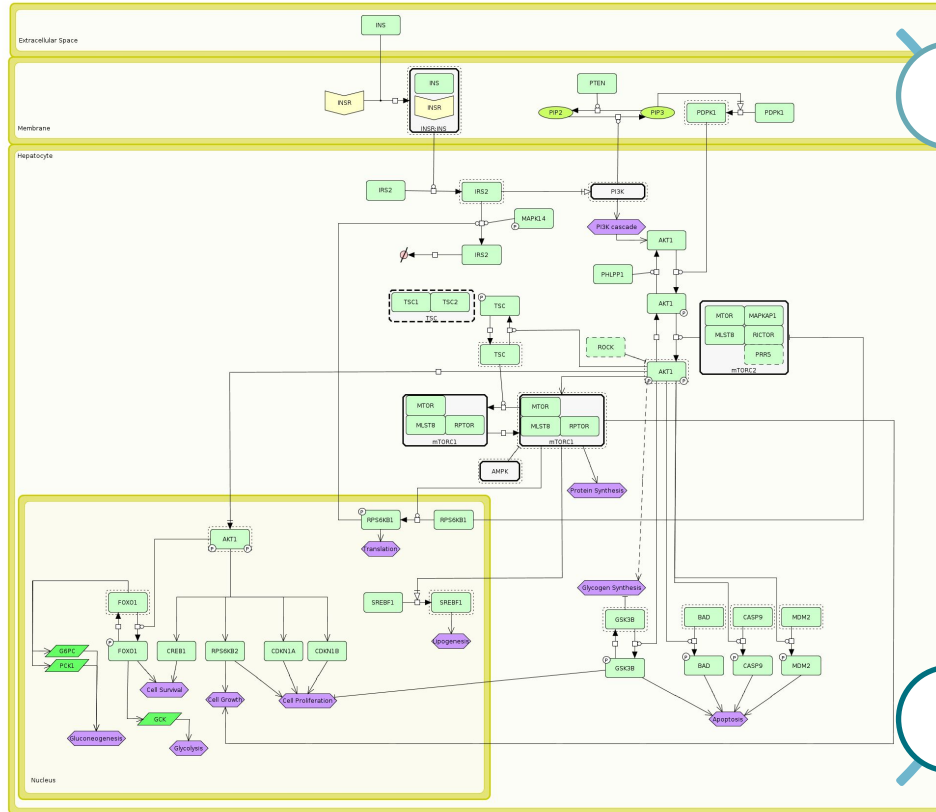
The concept of physiological maps



The concept of physiological maps



The concept of physiological maps



Graphical representation

Biological processes and interactions

Physiological knowledge

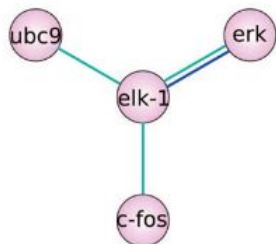
Standardized format

Data integration and annotation

Human and machine readable

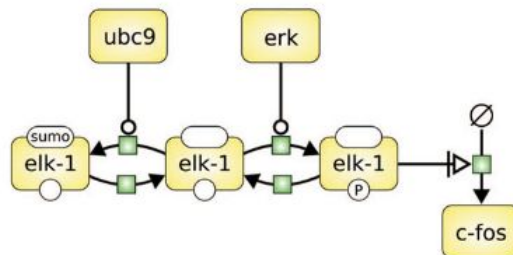


a interaction network



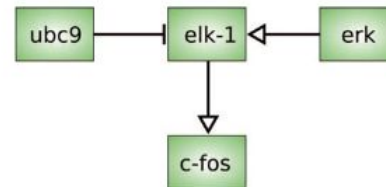
- directed
- sequential
- mechanistic

b process descriptions



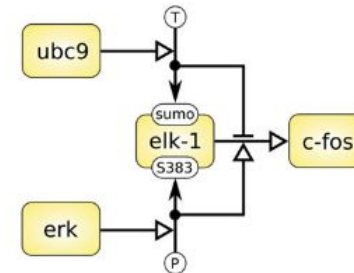
- directed
- sequential
- mechanistic

c activity flows



- directed
- sequential
- mechanistic

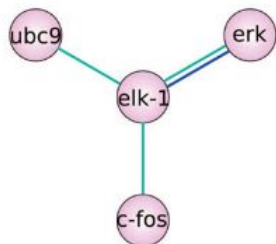
d entity relationships



- directed
- sequential
- mechanistic

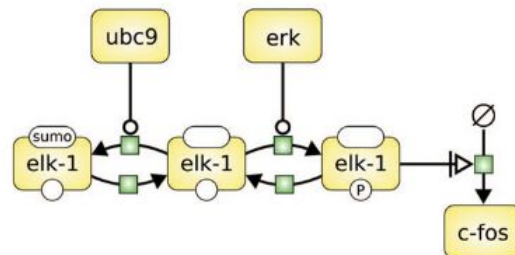


a interaction network



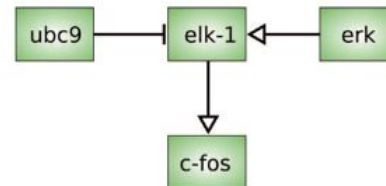
- directed
- sequential
- mechanistic

b process descriptions



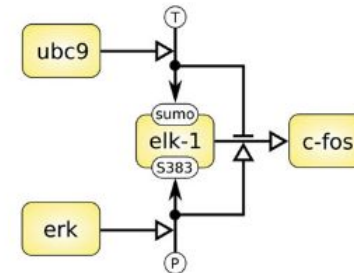
- directed
- sequential
- mechanistic

c activity flows



- directed
- sequential
- mechanistic

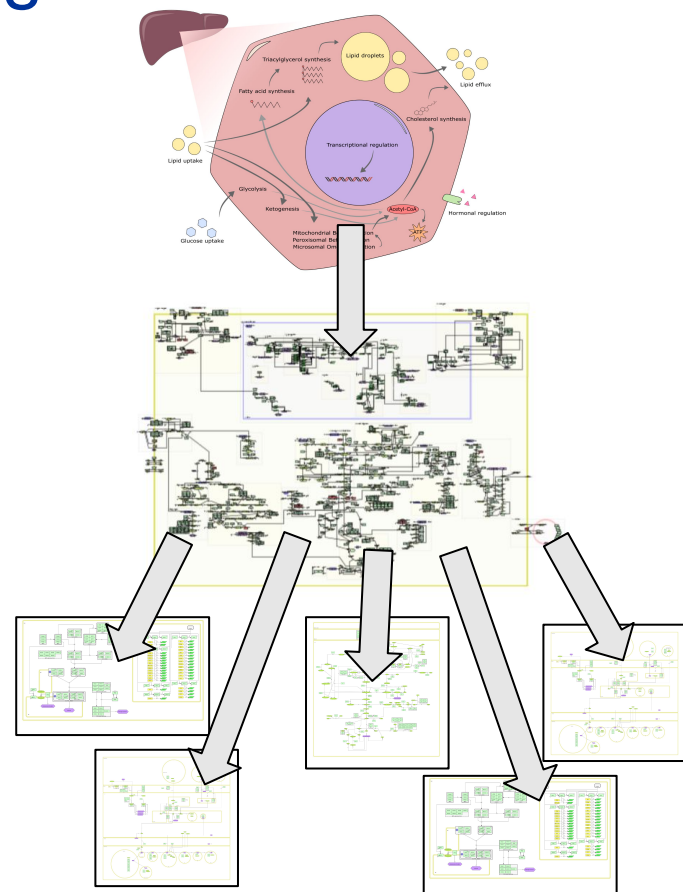
d entity relationships



- directed
- sequential
- mechanistic

The concept of physiological maps

- **Conceptual model:** used to navigate on the main map and submaps.
- **Main map:** different PMs presents different requirements for the main map (ex: Brain map has a cell-cell interaction using Activity Flow SBGN and graphical representation; Liver maps have submaps integrated in a big Process Description SBGN map).
- **Submaps:** they include detailed representation of undisturbed pathways and AOPs.



Our current maps



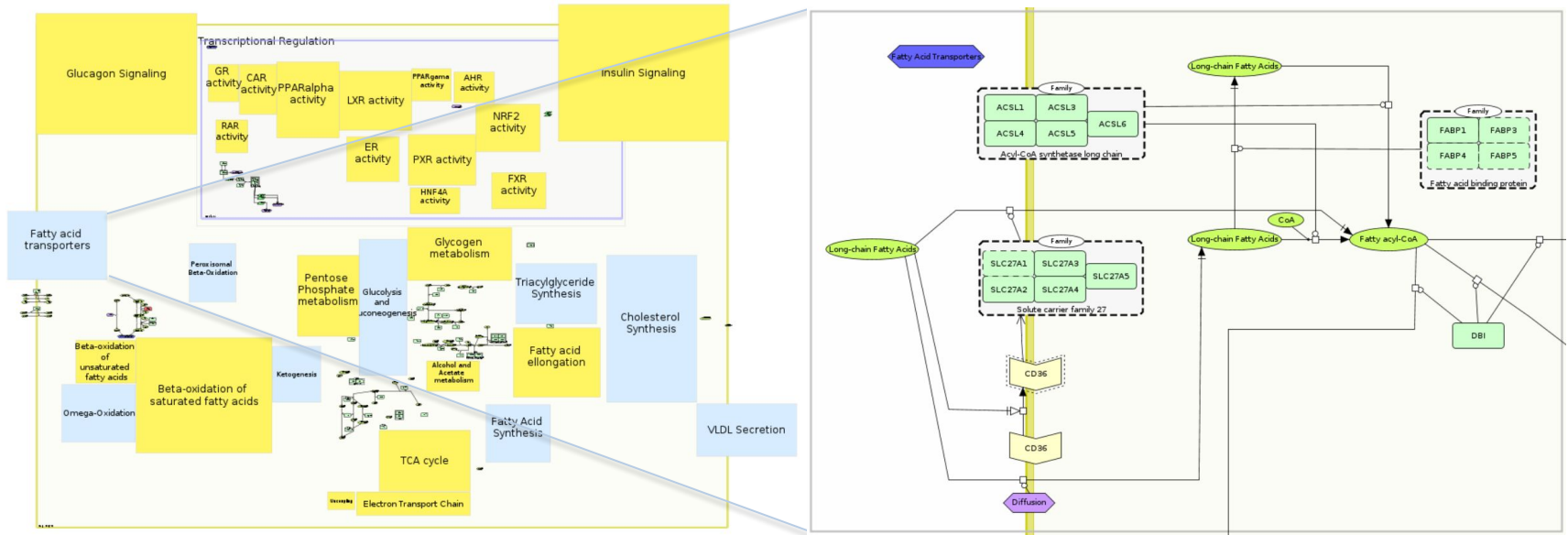
✓ bile secretion
✓ lipid metabolism



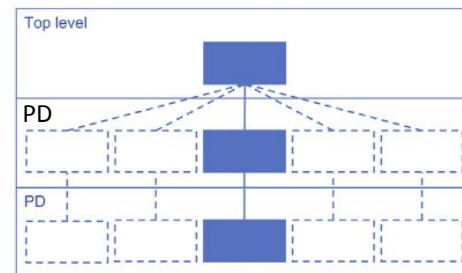
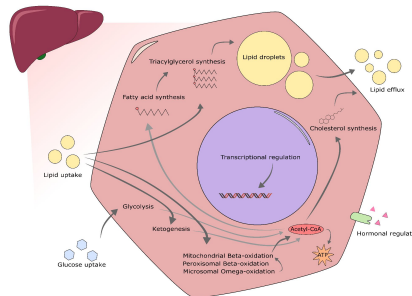
✓ nephron physiology



✓ neural tube closure
✓ cognitive function development



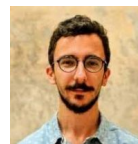
Liver maps



Liver Lipid Metabolism Physiological Map

Pathway	Location on the map
Cholesterol Biosynthesis Pathway	Submap and main map
Fatty Acid and Cholesterol Transporters	Submap and main map
Glucose metabolism (hepatocytes)	Main map
Fatty acid omega-oxidation	Main map
Peroxisomal beta-oxidation	Main map
Triacylglyceride Synthesis	Main map
Mitochondrial Metabolism Pathways	Submap
Gene regulatory network	Main map
Glucagon signaling	Submap and main map
Insulin signaling	Submap and main map

Main curator and domain expert



Luiz Ladeira

University of Liège - Belgium



Anouk Verhoeven

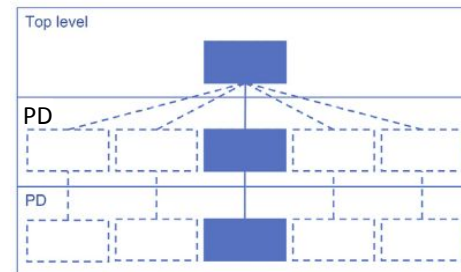
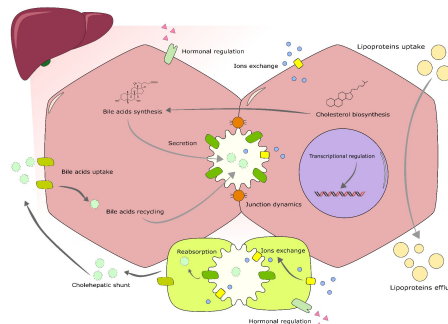
Vrije Universiteit Brussel (VUB) - Belgium



Julen Sanz Serrano

Vrije Universiteit Brussel (VUB) - Belgium

Liver maps



Liver Bile Secretion Physiological Map

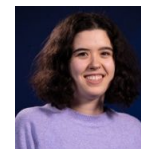
Pathway	Location on the map
Cholesterol Biosynthesis Pathway	Submap and main map
Fatty Acid and Cholesterol Transporters	Submap and main map
Mitochondrial Metabolism Pathways	Submap
Apoptosis Pathway	Submap
Autophagy Pathway	Submap
Bile acids biosynthesis	Main Map
Canaliculi dynamics pathways	Main Map
Bile salts circulation	Main Map
Bile salts uptake	Main Map
Cholehepatic shunt	Main Map
Glucagon signaling	Submap and main map
Insulin signaling	Submap and main map
Gene regulatory network	Main Map

Main curator and domain expert



Luiz Ladeira

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Annika Drees

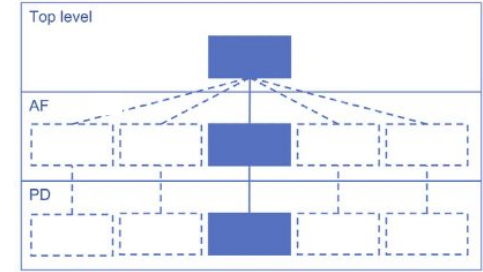
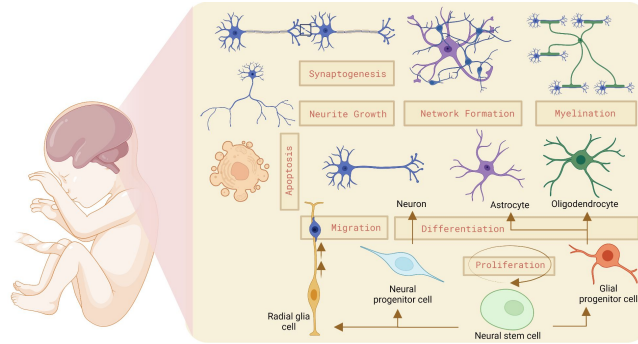
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Brain map



Brain Development Physiological Map

Pathway	Location on the map
Cell-cell interaction map	Main map
Neuron development	Submap
Oligodendrocyte development	Submap
Astrocyte development	Submap
Radial glia development	Submap

Main curator and domain expert



Luiz Ladeira

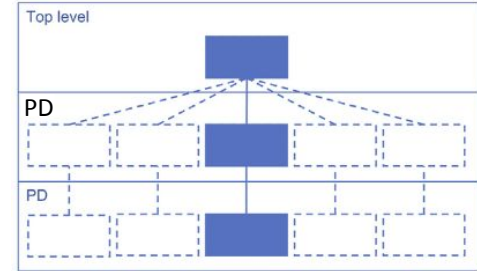
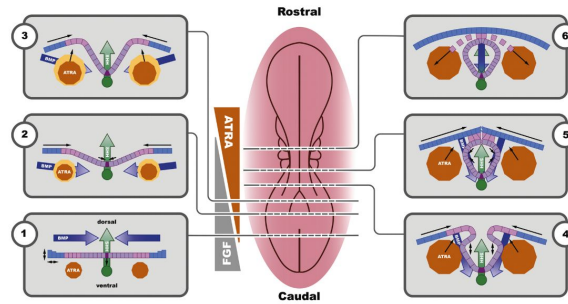
University of Liège - Belgium



Eliška Kuchovská

IUF – Leibniz-Institut für umweltmedizinische Forschung GmbH - Germany

NTC map



Neural Tube Closure Physiological Map

Pathway	Location on the map
BMP signaling pathway	Submap
SHH signaling pathway	Submap
FGF signaling pathway	Submap
Wnt signaling pathway	Submap
Gene regulatory network	Submap
ATRA metabolism	Submap
Folate metabolism	Submap

Main curator and domain expert



Alessio Gamba

University of Liège - Belgium

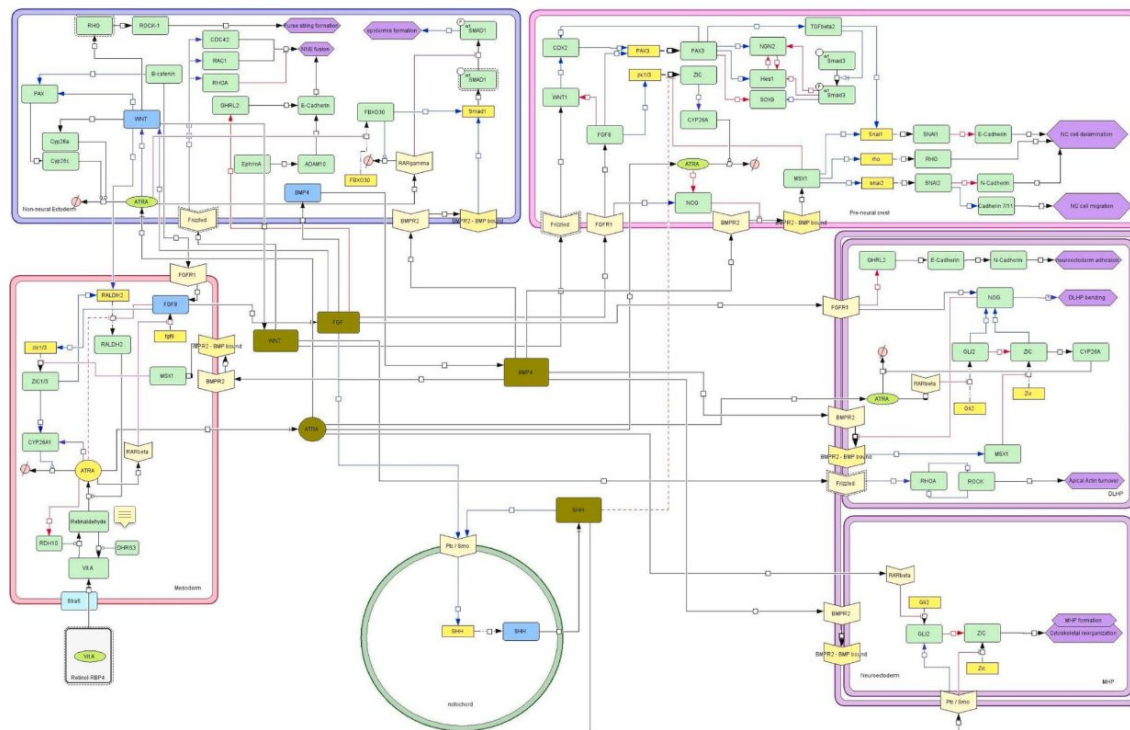


Job Berkhout

Utrecht University (UU) - The Netherlands
The Centre for Health Protection of the Dutch
National Institute for Public Health and the
Environment (RIVM)



NTC map



Harm Heusinkveld

Utrecht University (UU) - The Netherlands
The Centre for Health Protection of the Dutch National Institute for Public Health and the Environment (RIVM)

Contents lists available at [ScienceDirect](https://www.sciencedirect.com)

Reproductive Toxicology

journal homepage: www.elsevier.com/locate/reprotox

An ontology for developmental processes and toxicities of neural tube closure

Harm J. Heusinkveld^{1,2,*}, Yvonne C.M. Staal¹, Nancy C. Baker³, George Daston⁴, Thomas B. Knudsen⁴, Aldert Piersma¹

¹ Centre for Health Protection, National Institute for Public Health and the Environment (RIVM), Bilthoven, the Netherlands

² Leiden, Research Triangle Park NC 27711, USA

³ Global Product Stewardship, The Procter & Gamble Company, Cincinnati, OH USA

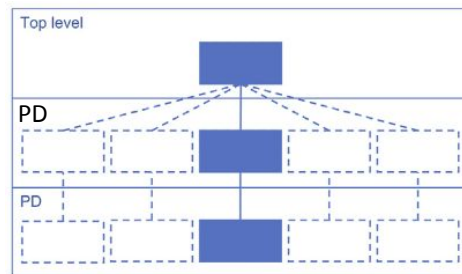
⁴ Center for Computational Toxicology and Exposure, U.S. Environmental Protection Agency, Research Triangle Park NC 27711, USA

https://ontox.elixir-luxembourg.org/minerva/index.xhtml?id=Neural_Tube_Closure_PM_v1

Kidney map

Nephron Physiological Map

Pathway	Location on the map
Renin-Angiotensin pathway	Submap and main map
Mitochondrial metabolism pathways	Submap
Transporters dynamics pathways	Main map
Endocytosis pathway	Submap
Post translational modifications	Submap and main map
Thyroid hormones signaling pathway	Submap and main map
TLR signaling pathway	Submap and main map
HTR2C signaling pathway	Submap and main map
Apoptosis	Submap
NLRP3 inflammasome	Submap

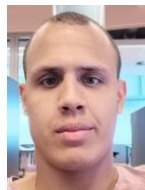


Main curator and domain expert



Alessio Gamba

University of Liège - Belgium

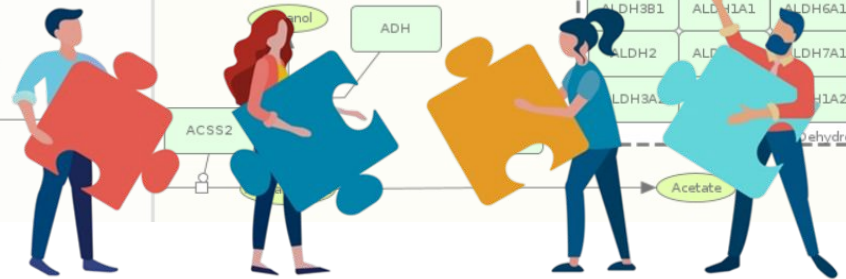
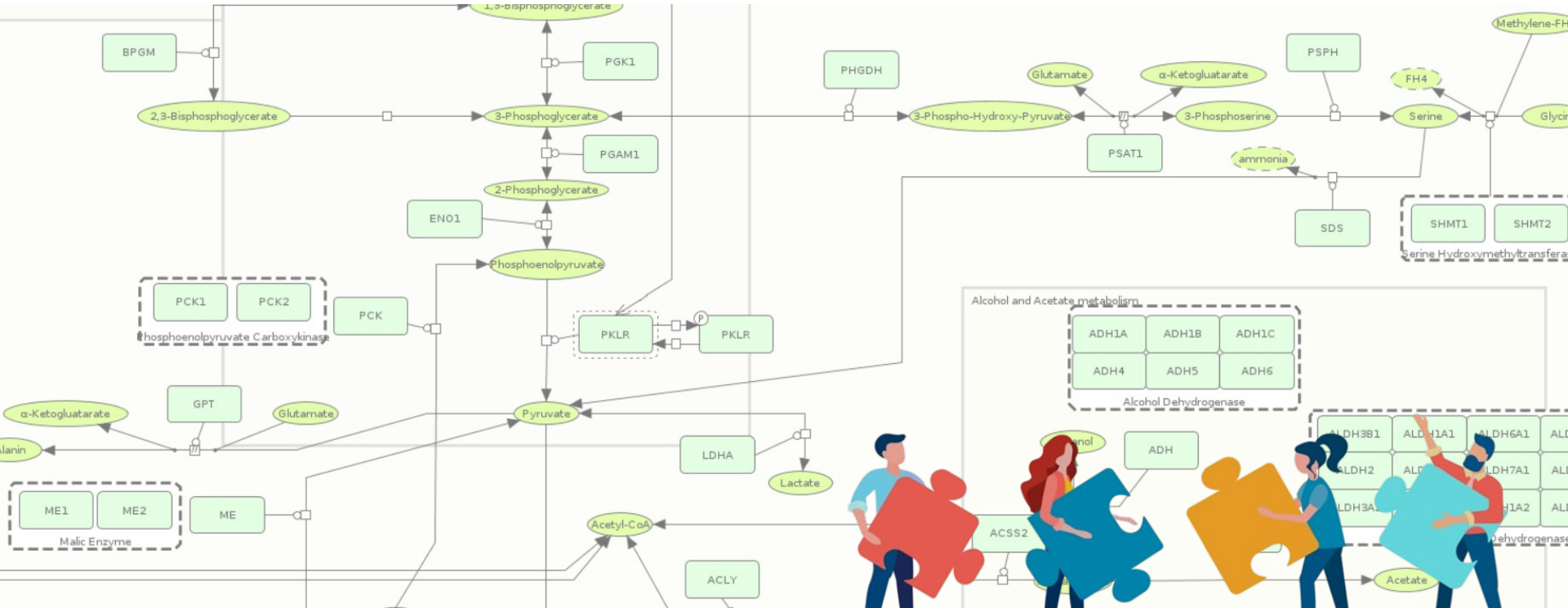


Devon Barnes

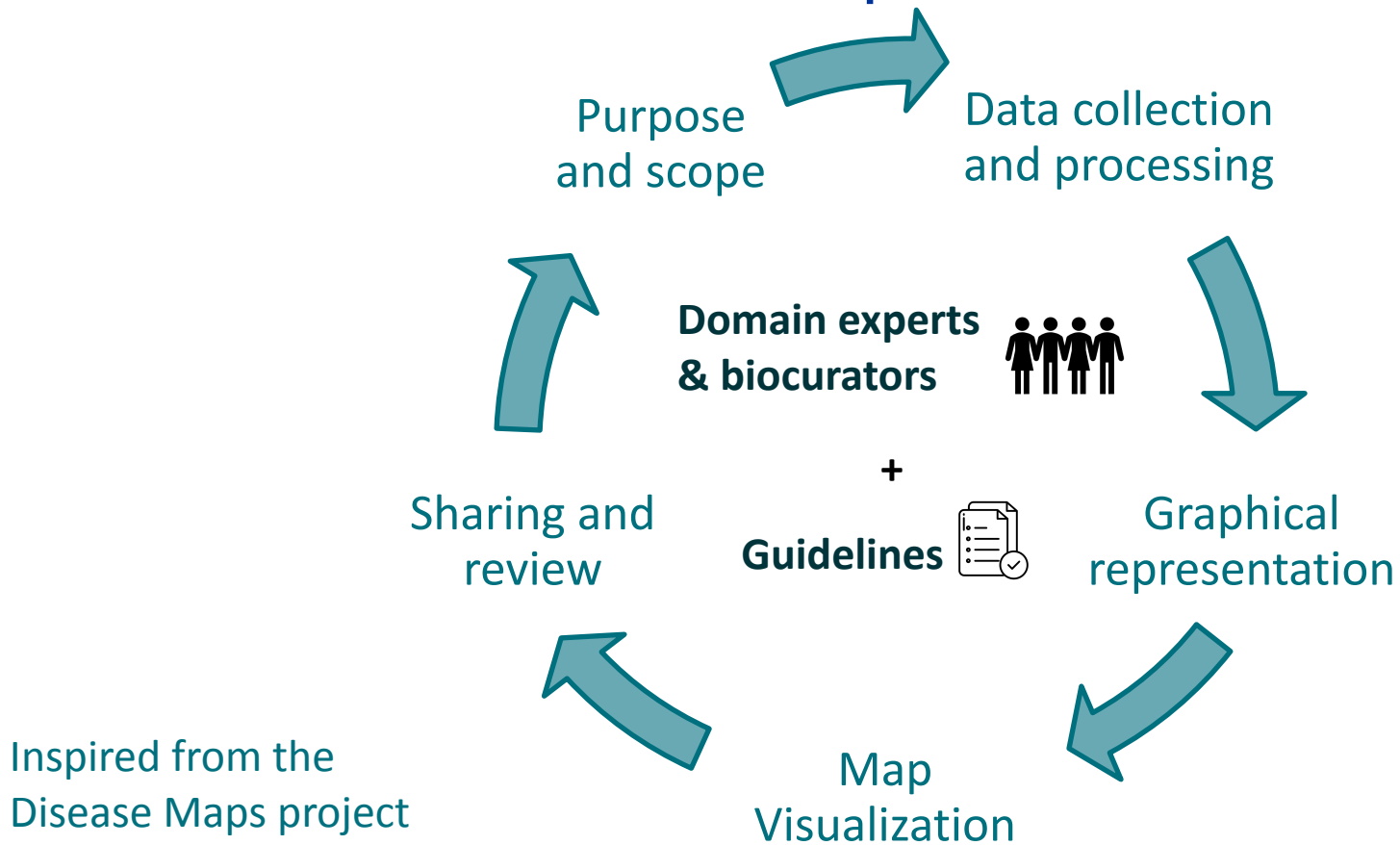
Utrecht University (UU) - The Netherlands



Assembling the puzzle

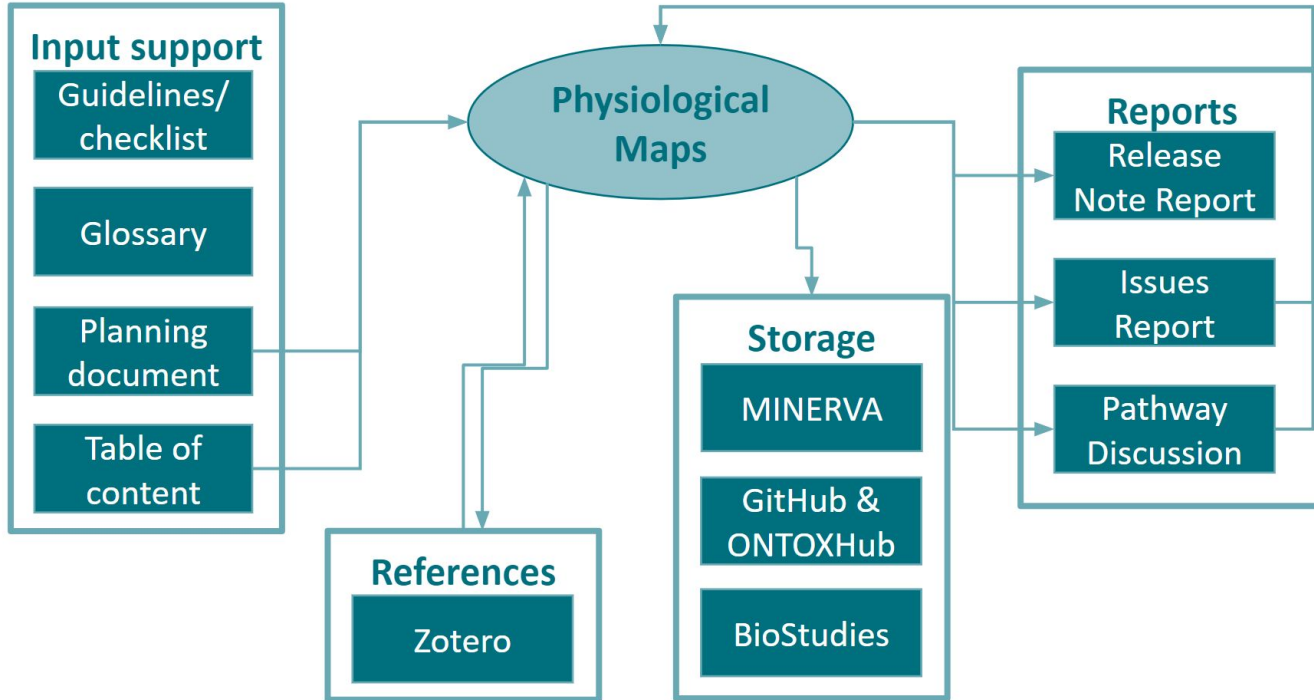


How do we build these maps?



Inspired from the
Disease Maps project

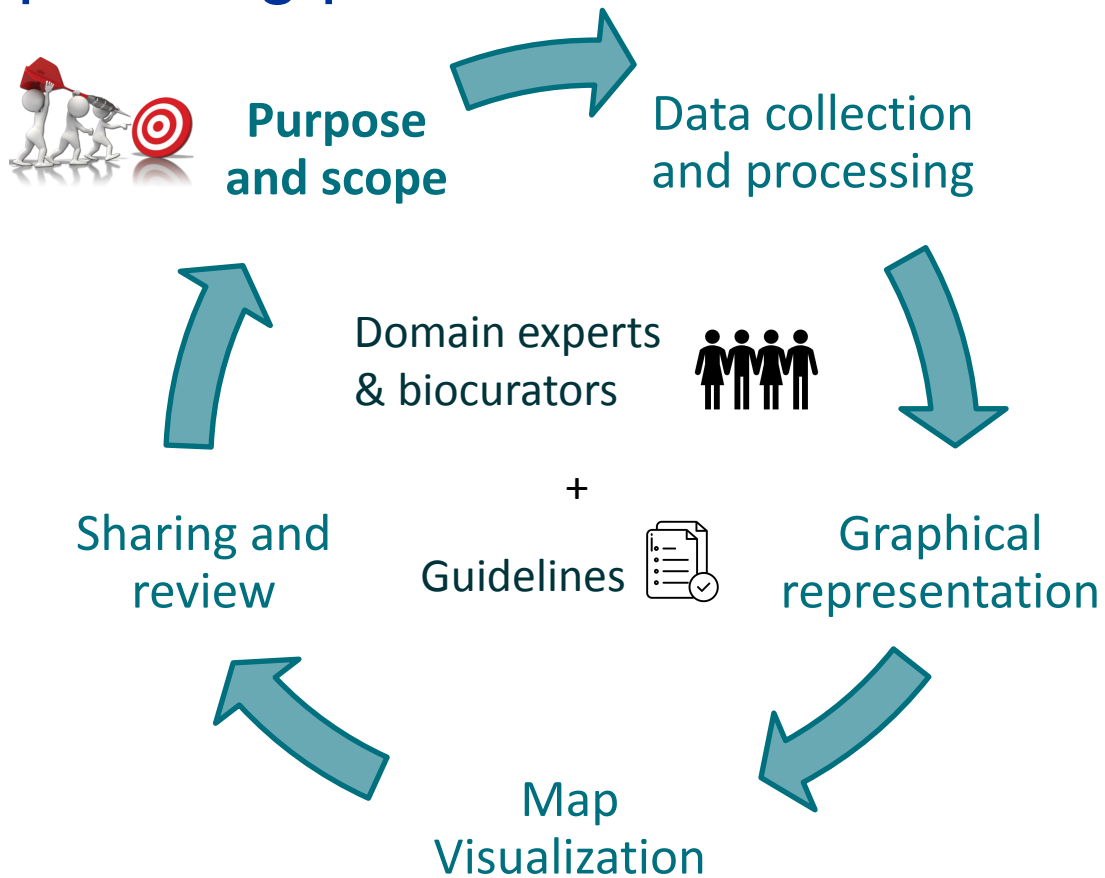
Documentation structure



Planning phase

- **Modular structure:**
 - ✓ Use of **submaps (= smaller modules)**
 - ✓ Improve data visualization
 - ✓ Simplify review and curation processes
 - ✓ Modules can be submitted to WikiPathways: accelerate curation
- **Annotation:**
 - ✓ Manual & **automated annotation** (guidelines, MINERVA)
- **Documentation:**
 - ✓ Curation **guidelines**
 - ✓ Quality control checklist
- **Storage:**
 - ✓ MINERVA, GitHub, BioStudies

Workflow: planning phase



Purpose and scope

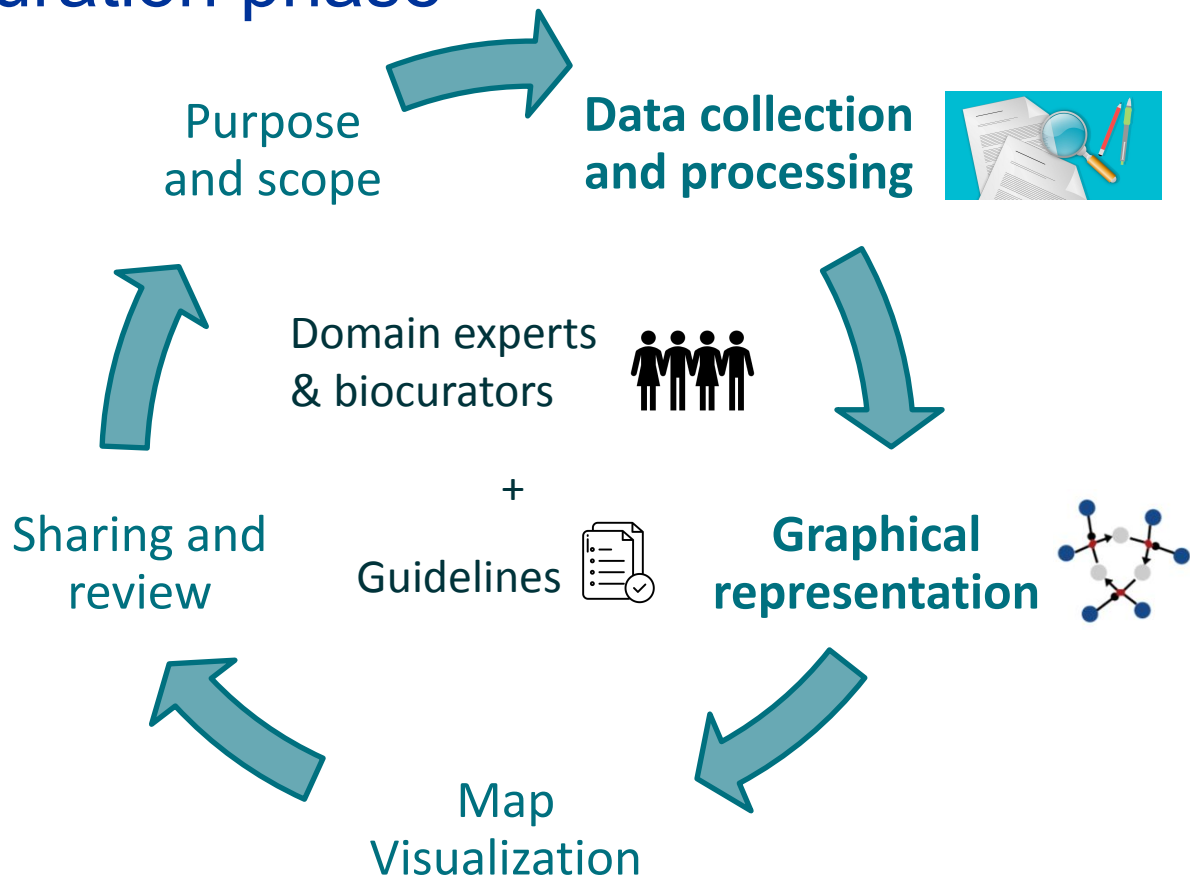
Plan for the map:

- **Who?**
List of curators and domain experts
- **Why?**
Intended use of the map
- **What?**
 - ✓ Map content (pathways, molecules)
 - ✓ First references
- **How?**
 - ✓ Level of granularity
 - ✓ Sustainability/storage

Table of content:

Pathway	Diagrams		Comments
	Resource	Access	
Cholesterol Biosynthesis Pathway	Reactome, WP, and literature	10.3180/R-HSA-191273.7 , WP4718	Pathway revised. Stable version. Comments are in the Discussion file .
Fatty Acid and Cholesterol Transporters	Wikipathways Wikipathways KEGG	WP5061 WP5304 map04979	Pathway revised. Stable version. Comments are in the Discussion file . Lipoprotein uptake and secretion are included.
Mitochondrial Metabolism Pathways	PDMap, COVID19Map, and literature	PDMap COVID19Map	Pathway revised. Stable version. Comments are in the Discussion file . Oxidative stress and fatty acid biosynthesis are included.

Workflow: curation phase



Curation phase

- **Data resources:**

- ✓ Books, research papers, review articles, databases   
- ✓ Other maps (e.g. from Disease Maps project)
- ✓ Tools for systematic review **sysrev**

- **Graphical representation:**

- ✓ Graphical standard - **Systems Biology Graphical Notation (SBGN)**:
 - Visual language for biochemical interaction networks
 - Standardized representation
- ✓ Diagram editor: **CellDesigner**
 - SBGN-compatible networks
 - Data format: **Systems Biology Markup Language (SBML)**
 - Free and open
 - Manual annotation

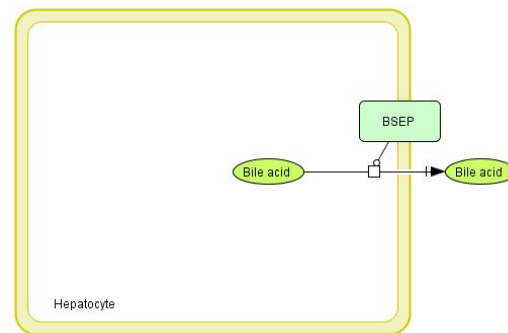
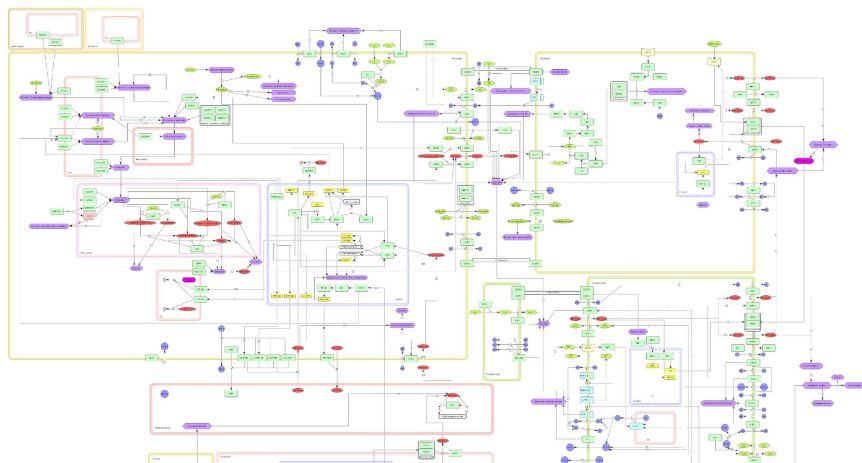
Curating literature

Location

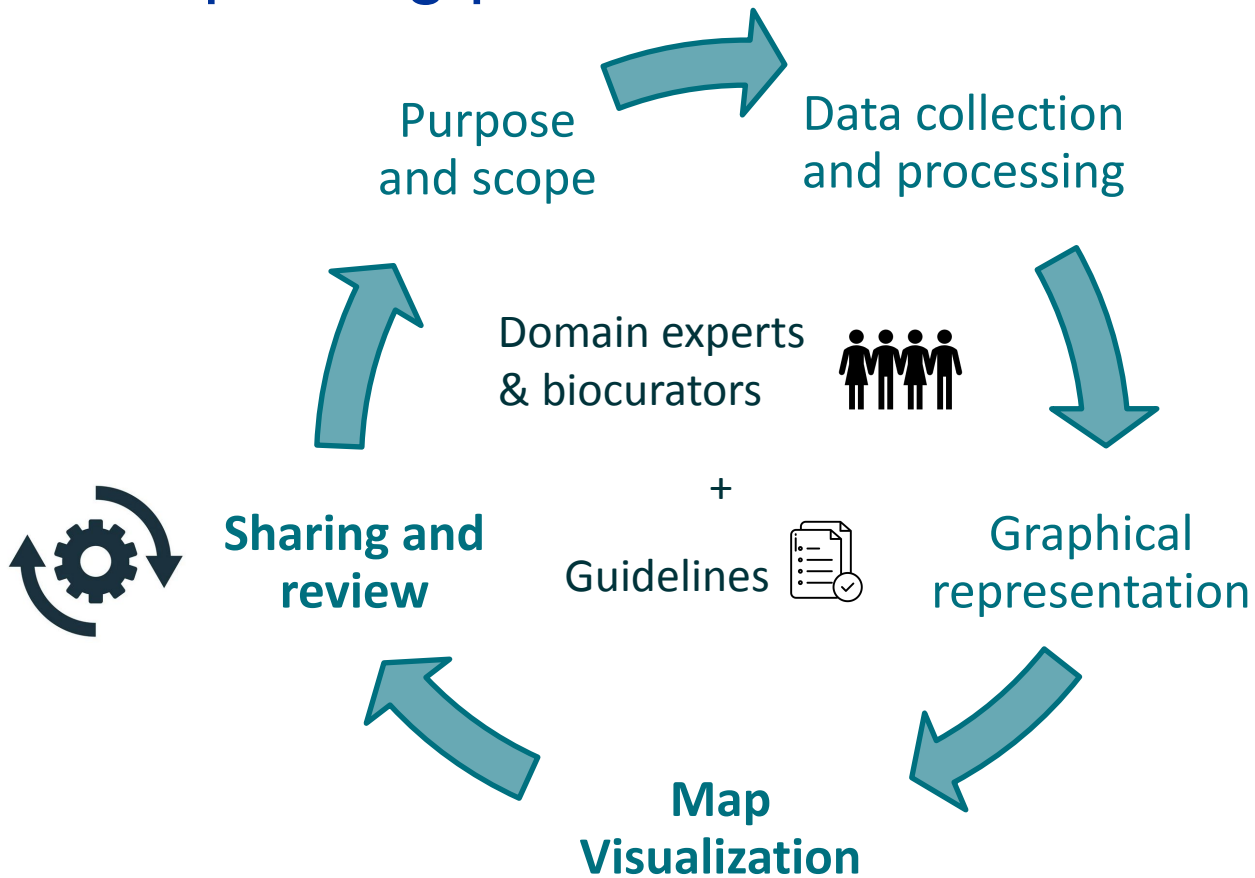
Subject **BSEP** an apical canalicular domain ATP binding cassette (ABC) export pump – is the primary transporter of bile acids from the hepatocyte into

Function bile and the rate limiting factor in the formation of bile salt-dependent bile flow. BSEP exists within lipid rich microdomains in canalicular membrane

Boyer and Soroka. 2021. <https://doi.org/10.1016/j.jhep.2021.02.011>.



Workflow: updating phase



Updating phase

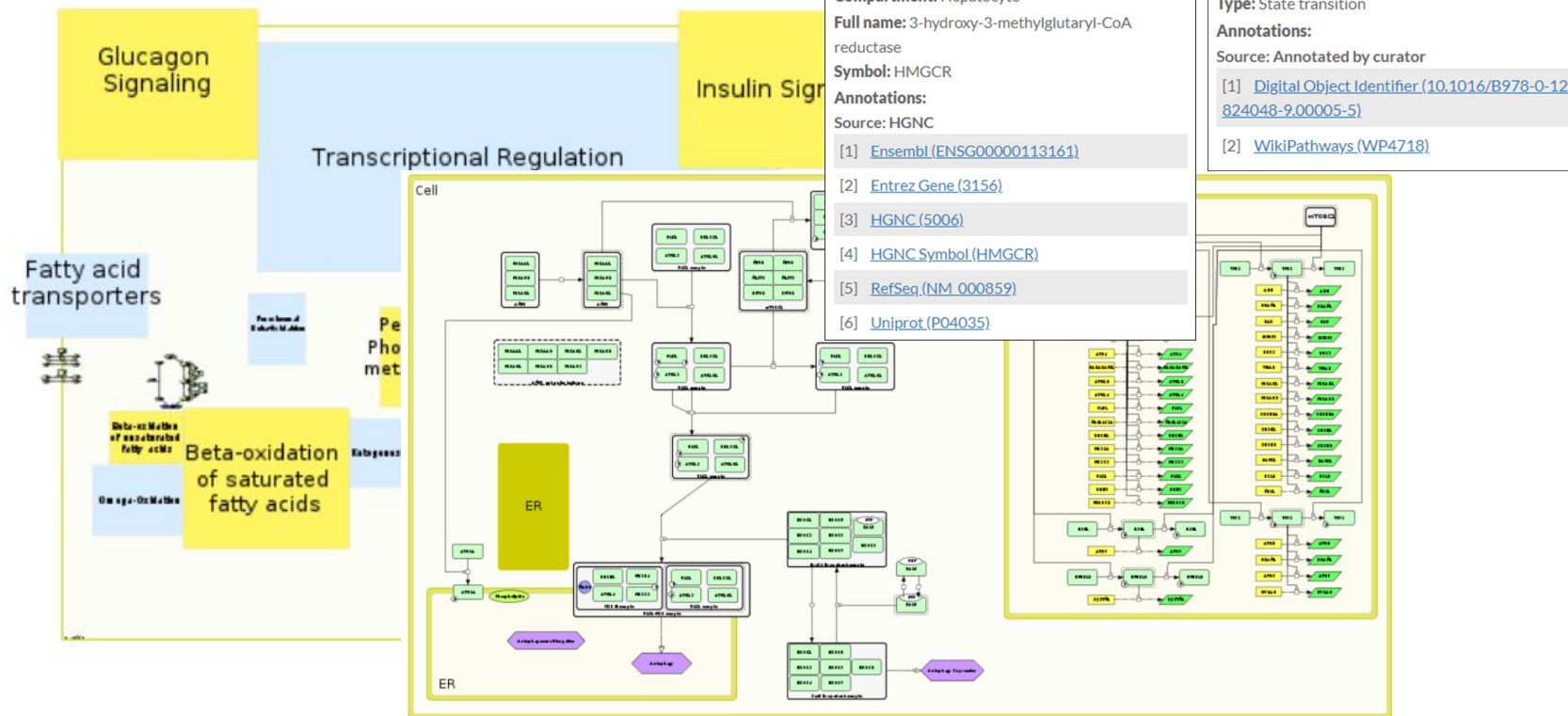
- **Visualization and annotation tool – MINERVA platform:**

- ✓ **Molecular Interaction Networks Visualization**
- ✓ **User-friendly interface**
- ✓ **Content exploration (e.g. searching for drug targets), overlay of experimental datasets**
- ✓ **Automated annotation** using identifiers (e.g. ChEBI, Ensembl, Uniprot, Gene Ontology)
- ✓ **Conversion in various formats (SBML, GPML, etc.)**



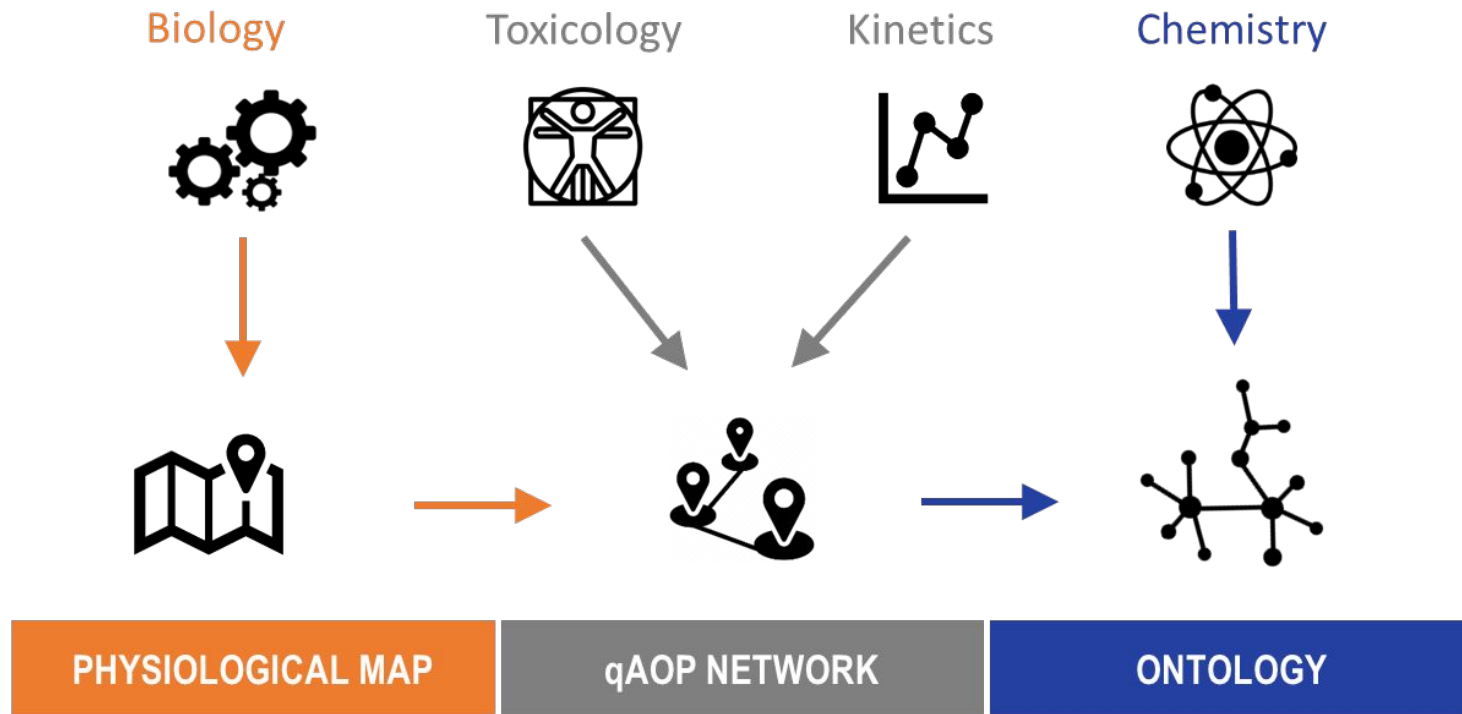
Updating phase

Example from MINERVA

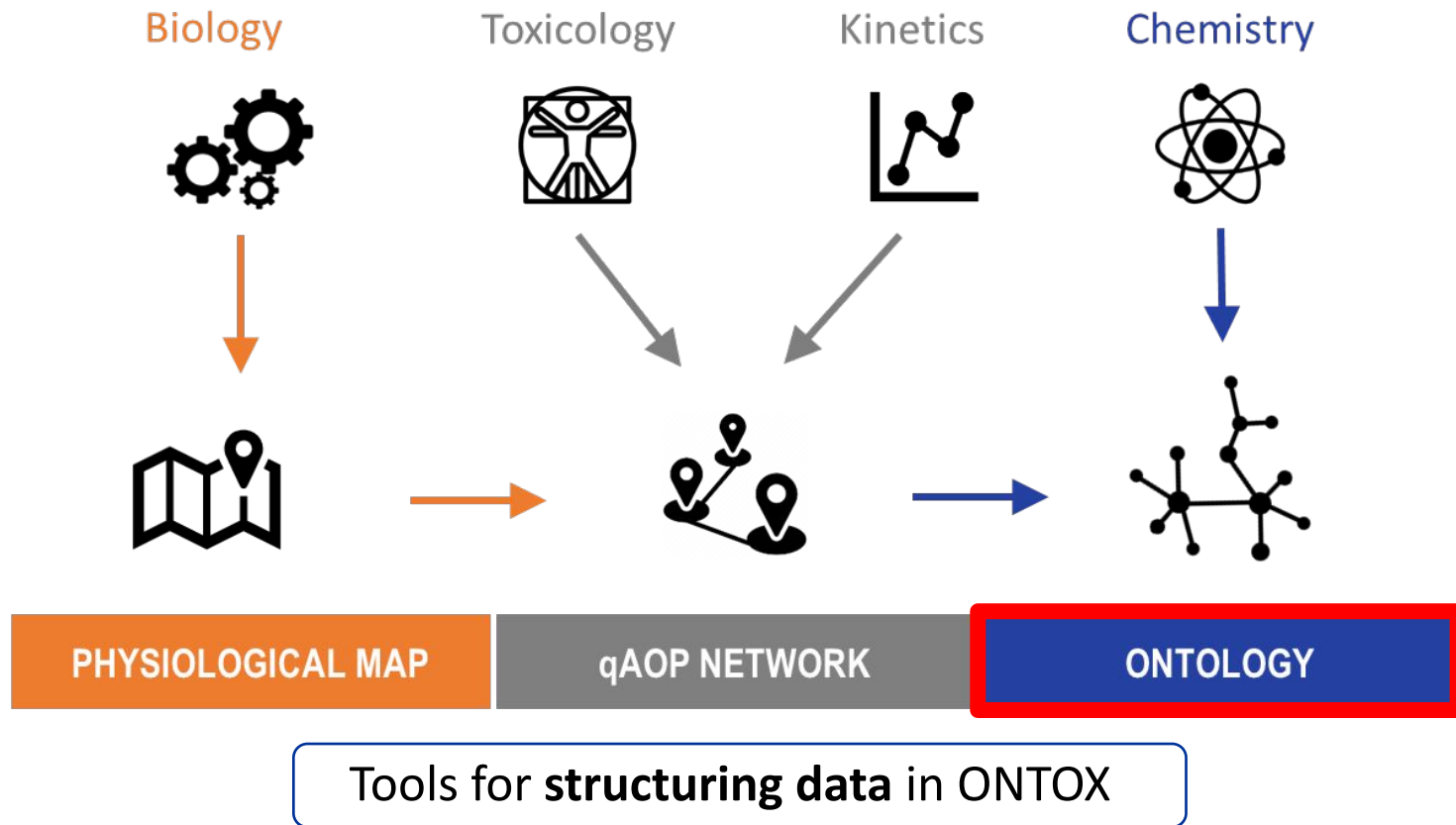


Updating phase

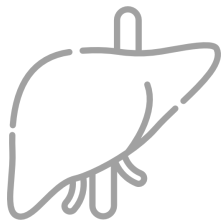
- **Sharing and review:**
 - ✓ Maps need a **continuous update**
 - ✓ Commenting facilitated by the MINERVA Platform
 - ✓ Storage on GitHub and BioStudies
 - ✓ Collaboration between domain experts and curators
 - Create a **bridge between systems biology and toxicology communities**



Tools for **structuring data** in ONTOX



Ontology case-studies



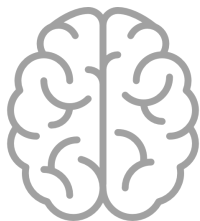
- ✓ Lipid metabolism
- ✓ Bile secretion

- ❖ Steatosis
- ❖ Cholestasis



- ✓ Nephron

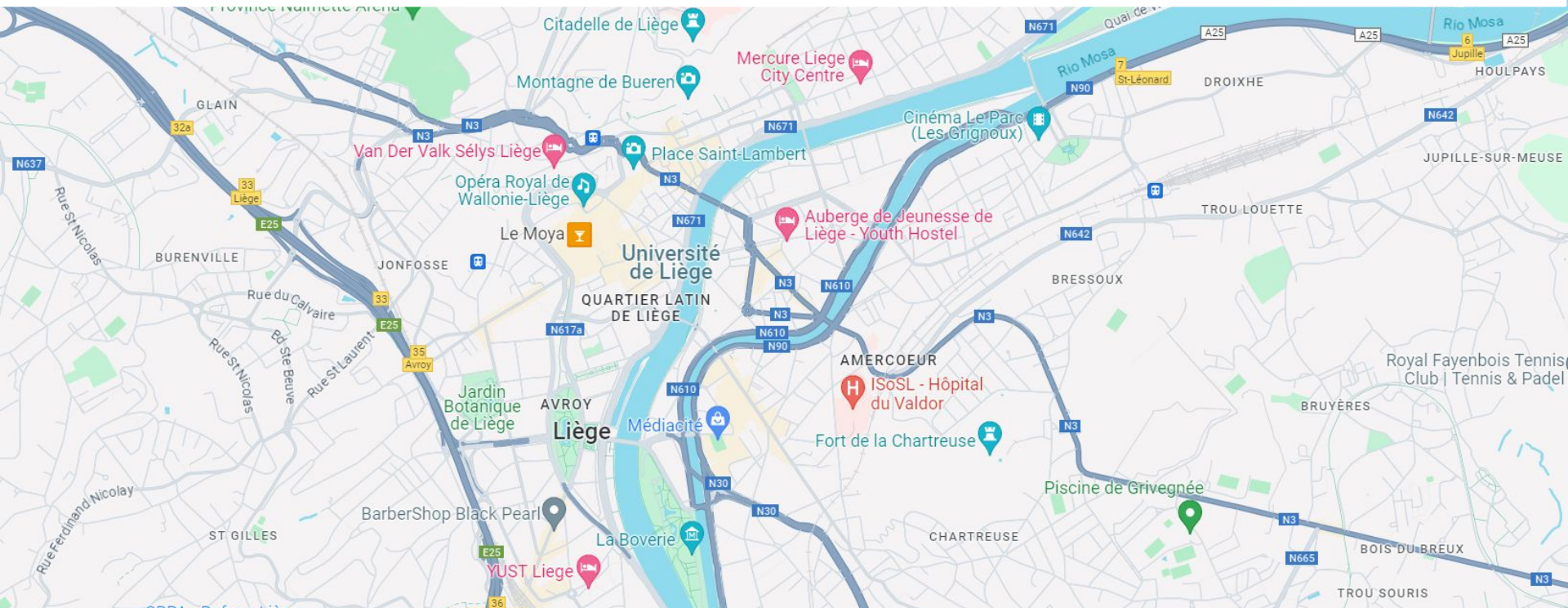
- ❖ Tubular necrosis
- ❖ Cholestasis



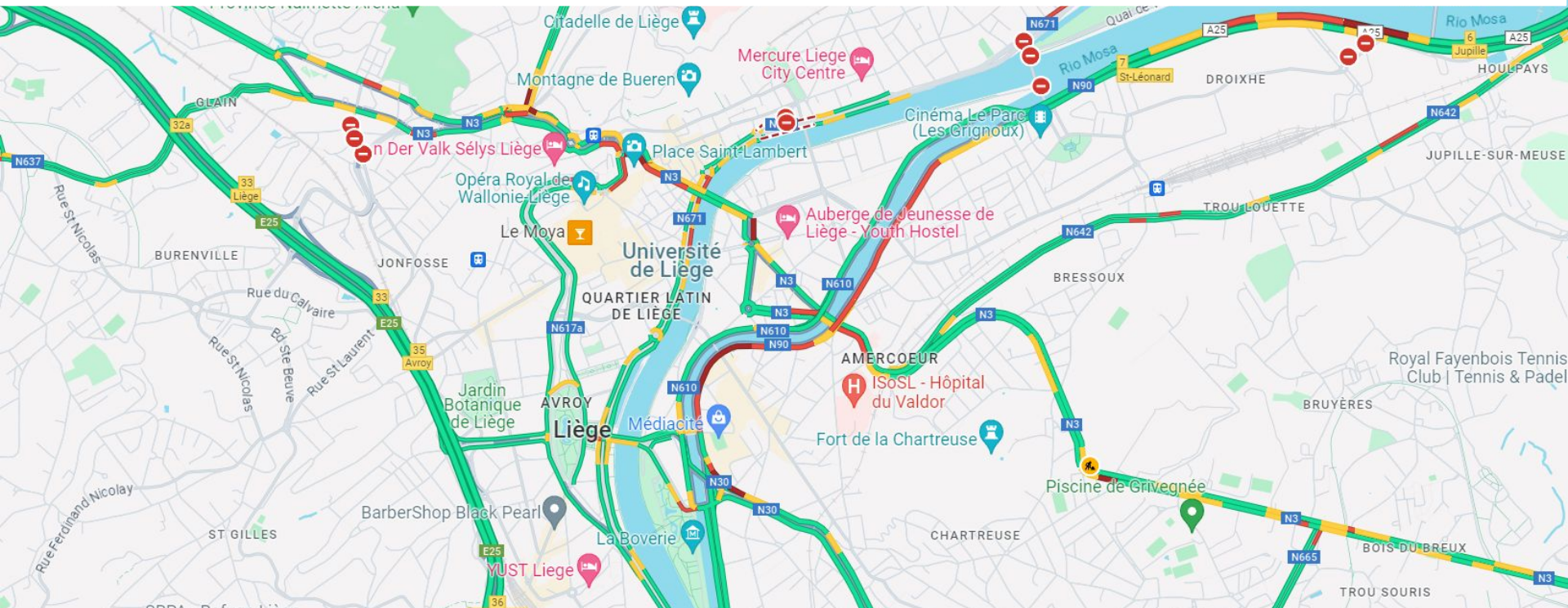
- ✓ Neural tube closure
- ✓ Brain development

- ❖ Neural tube closure defects
- ❖ Cognitive function defects

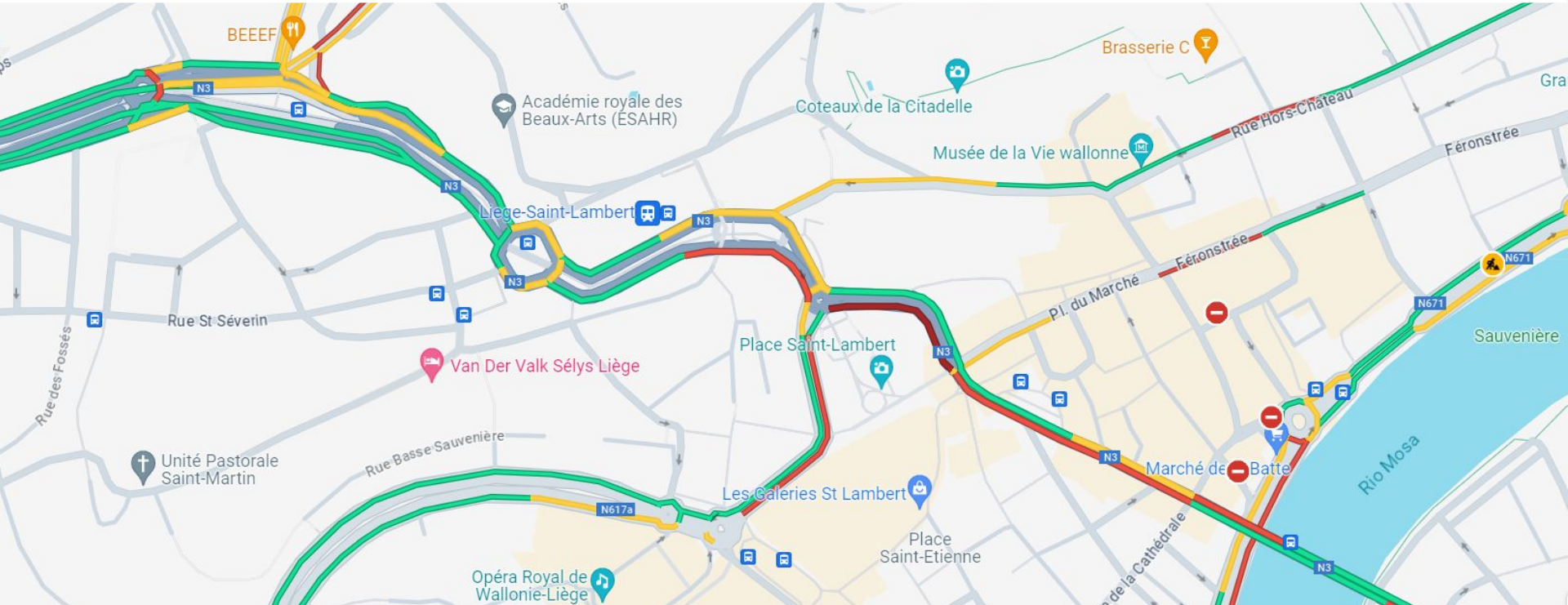
The concept of ontology maps



The concept of ontology maps



The concept of ontology maps



The concept of ontology maps



Manège de la Caserne Fonck

4,2 ★★★★★ (658) ⓘ

Local para eventos

Visão geral

Avaliações

Sobre



Rotas



Salvar



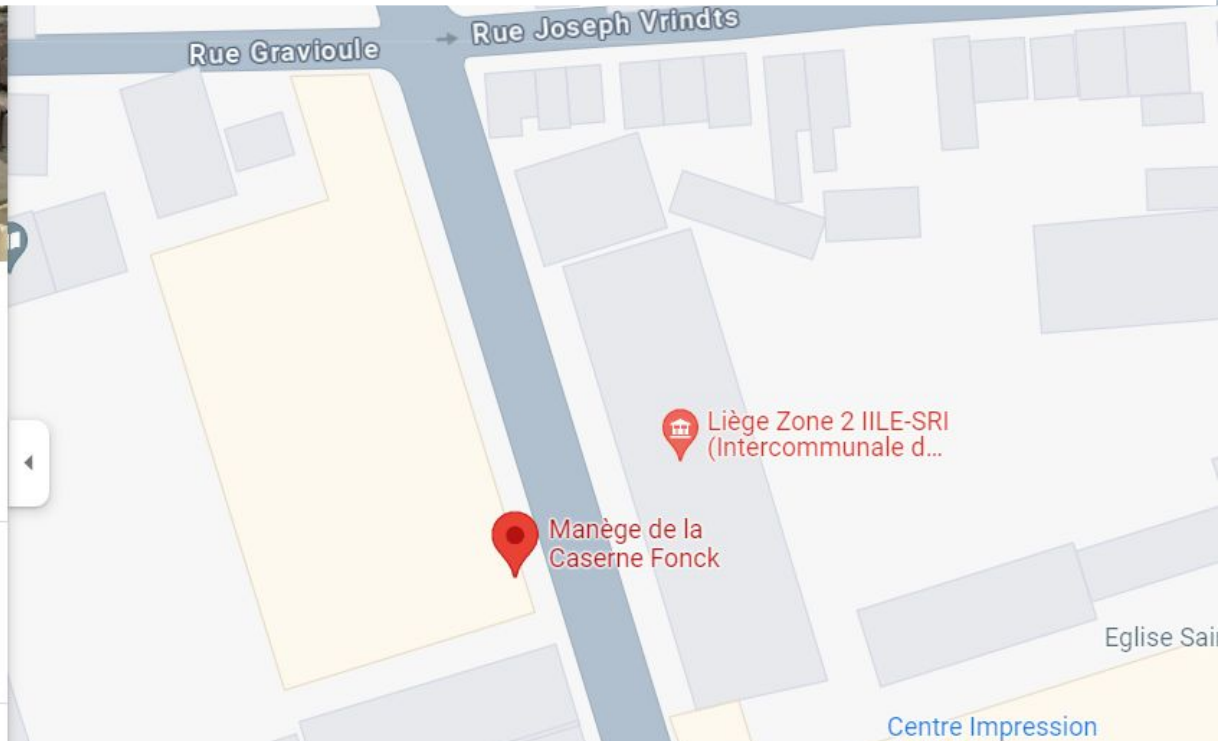
Próximo



Enviar para o
smartphone



Compartilhar



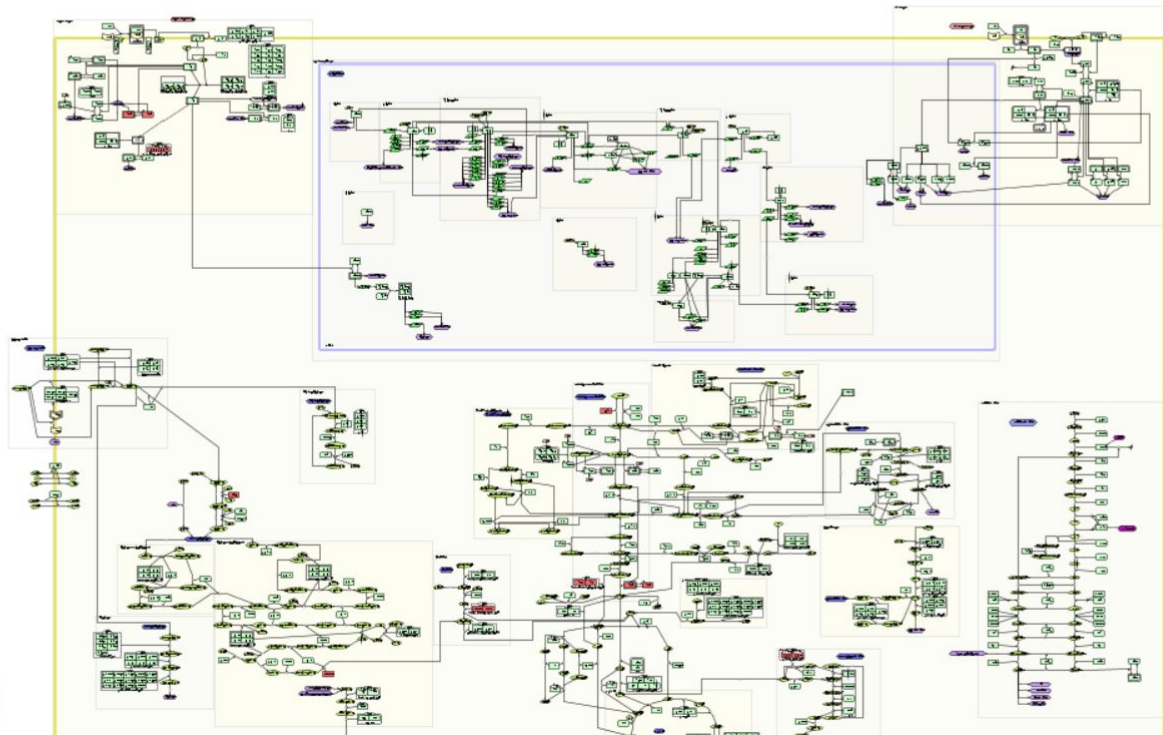
ONTOX ontology maps

SEARCH OVERLAYS SUBMAPS INFO

CONTENT DRUG CHEMICAL MIRNA

SEARCH IN CONTENT: ?

PERFECT MATCH



ONTOX ontology maps

SEARCH OVERLAYS SUBMAPS INFO

CONTENT DRUG CHEMICAL MiRNA

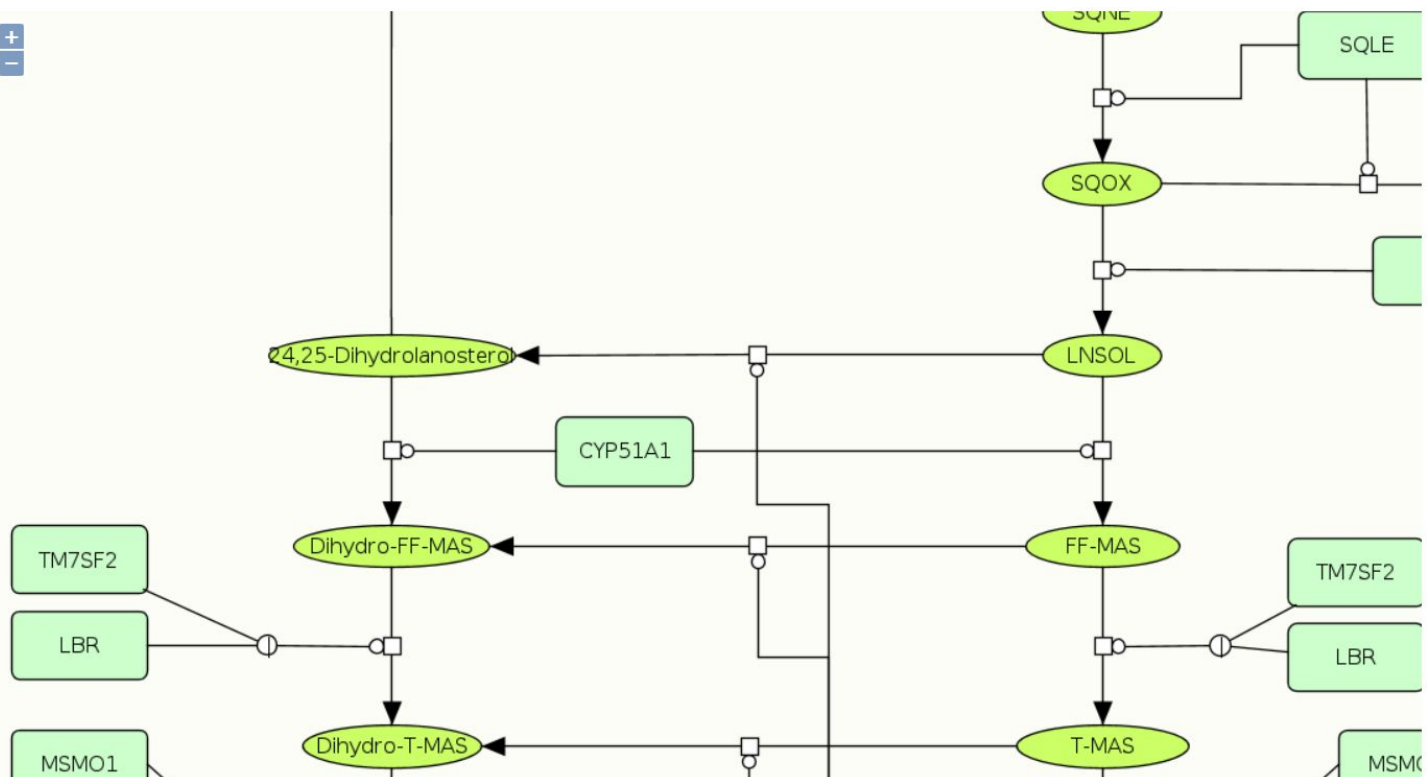
SEARCH IN CONTENT: ?

PERFECT MATCH

TM7SF2

LBR

MSMO1



ONTOX ontology maps

SEARCH OVERLAYS SUBMAPS INFO

CONTENT DRUG CHEMICAL MiRNA

SEARCH IN CONTENT: ?

PERFECT MATCH

Reaction: re27

Type: State transition

Annotations:

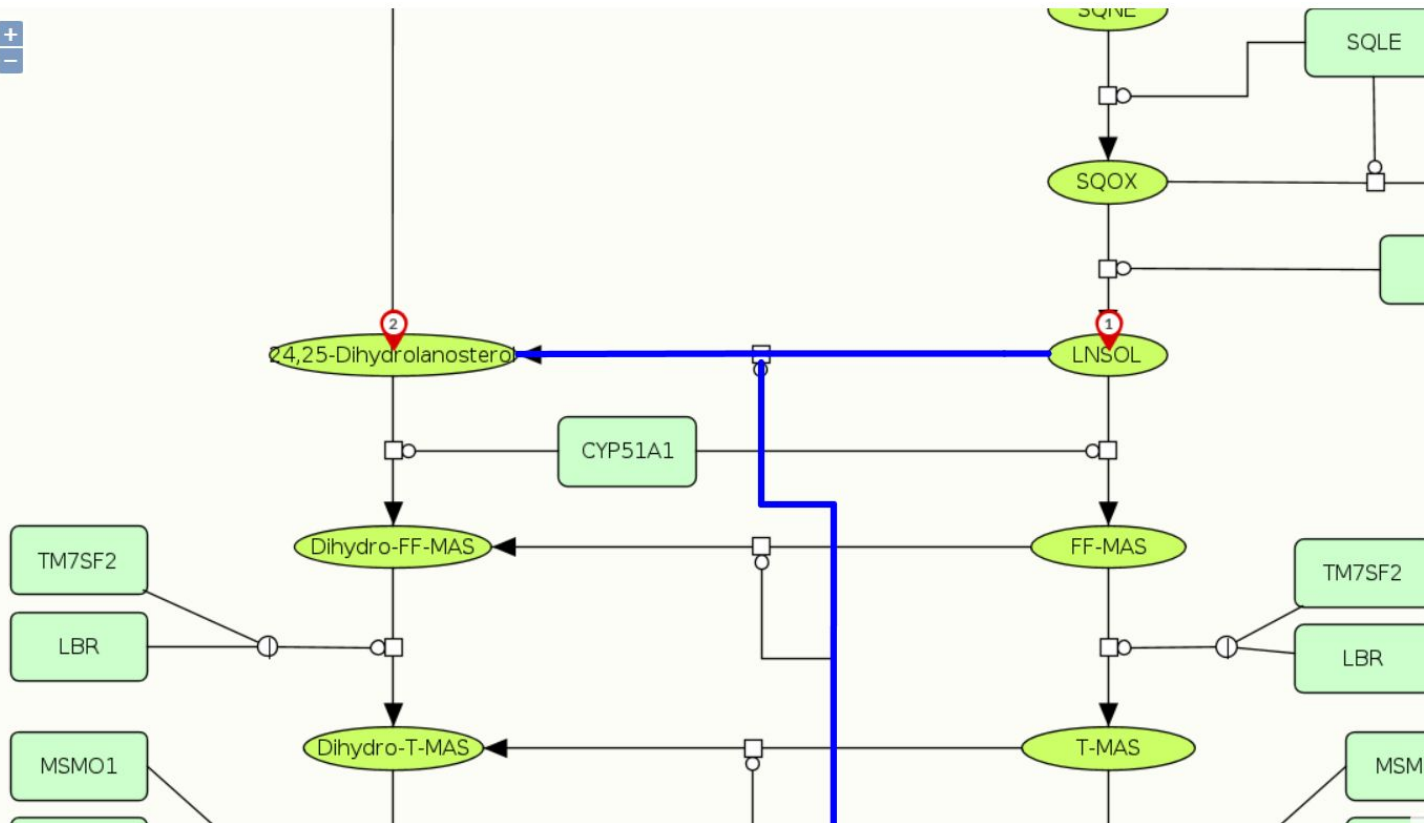
Source: Annotated by curator

[1] [Digital Object Identifier \(10.1016/B978-0-12-824048-9.00005-5\)](https://doi.org/10.1016/B978-0-12-824048-9.00005-5)

1 Simple molecule: LNSOL

Compartment: Hepatocyte

Full name: lanosterol



ONTOX ontology maps

SEARCH OVERLAYS SUBMAPS INFO

CONTENT DRUG CHEMICAL MiRNA

SEARCH IN CONTENT: ?

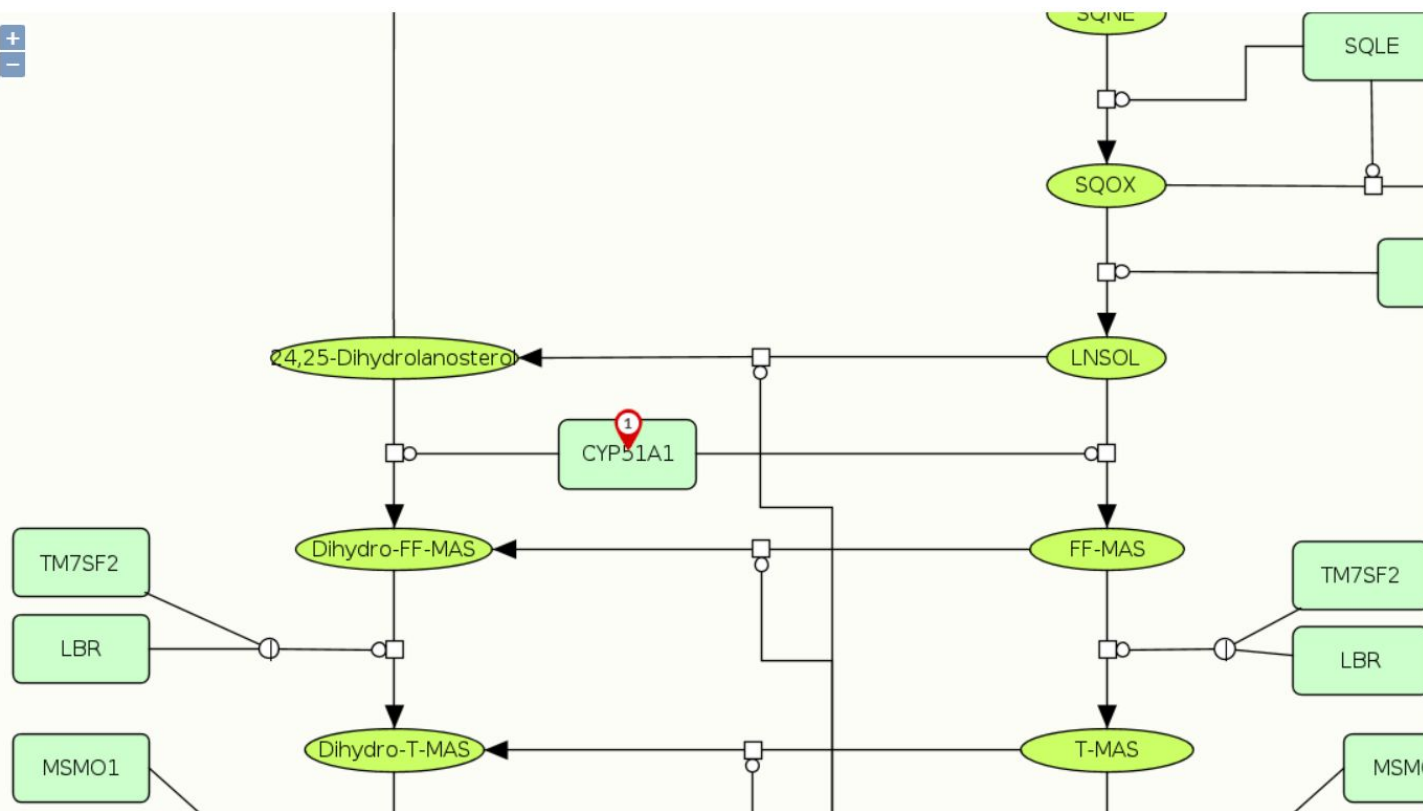
PERFECT MATCH

Synonyms: CP51, CYPL1, LDM, P450-14DM, P450L1

Annotations:

Source: HGNC

- [1] [Ensembl \(ENSG00000001630\)](#)
- [2] [Entrez Gene \(1595\)](#)
- [3] [HGNC \(2649\)](#)
- [4] [HGNC Symbol \(CYP51A1\)](#)
- [5] [RefSeq \(NM_000786\)](#)
- [6] [Uniprot \(Q16850\)](#)



ONTOX ontology maps

SEARCH OVERLAYS SUBMAPS INFO

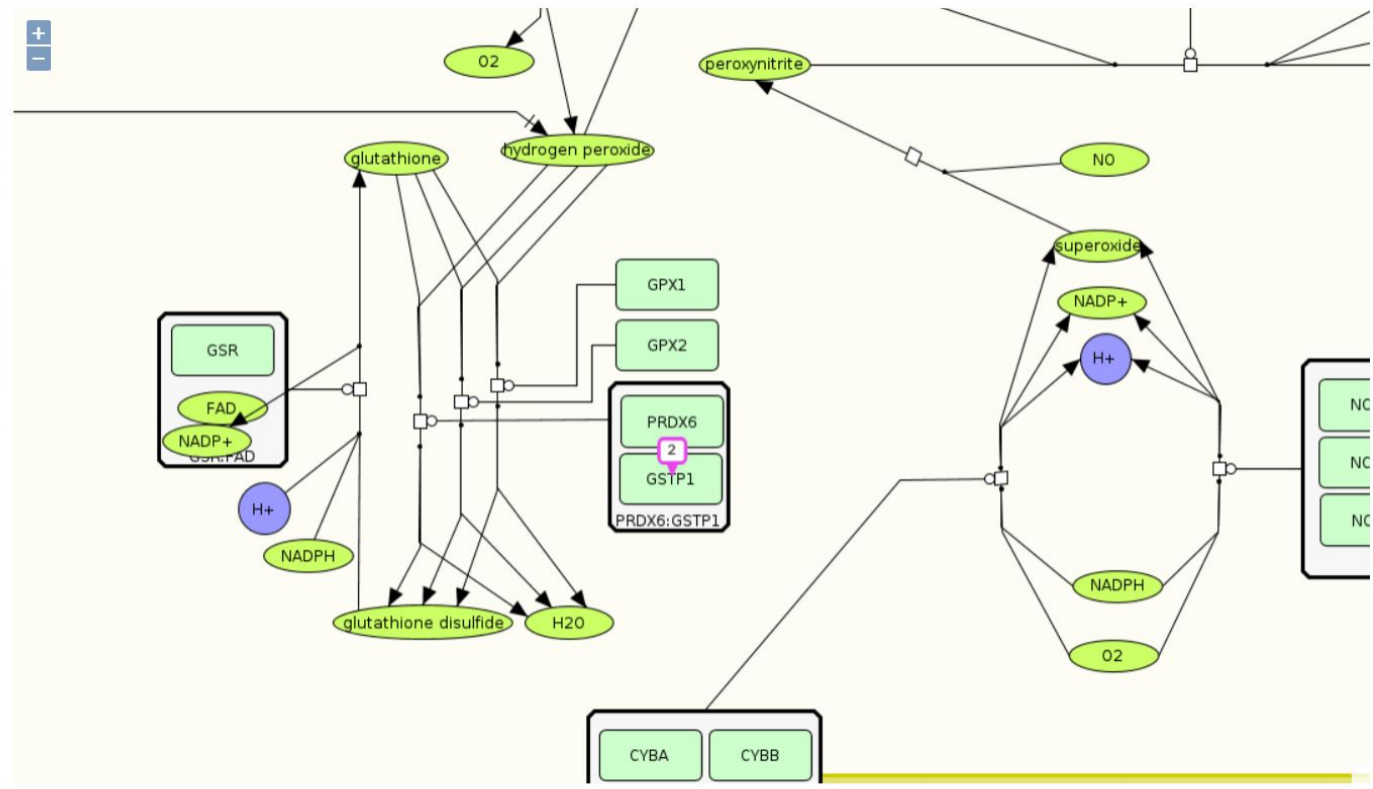
CONTENT DRUG CHEMICAL MiRNA

SEARCH FOR TARGETS OF:

paracetamol

Drug: Acetaminophen

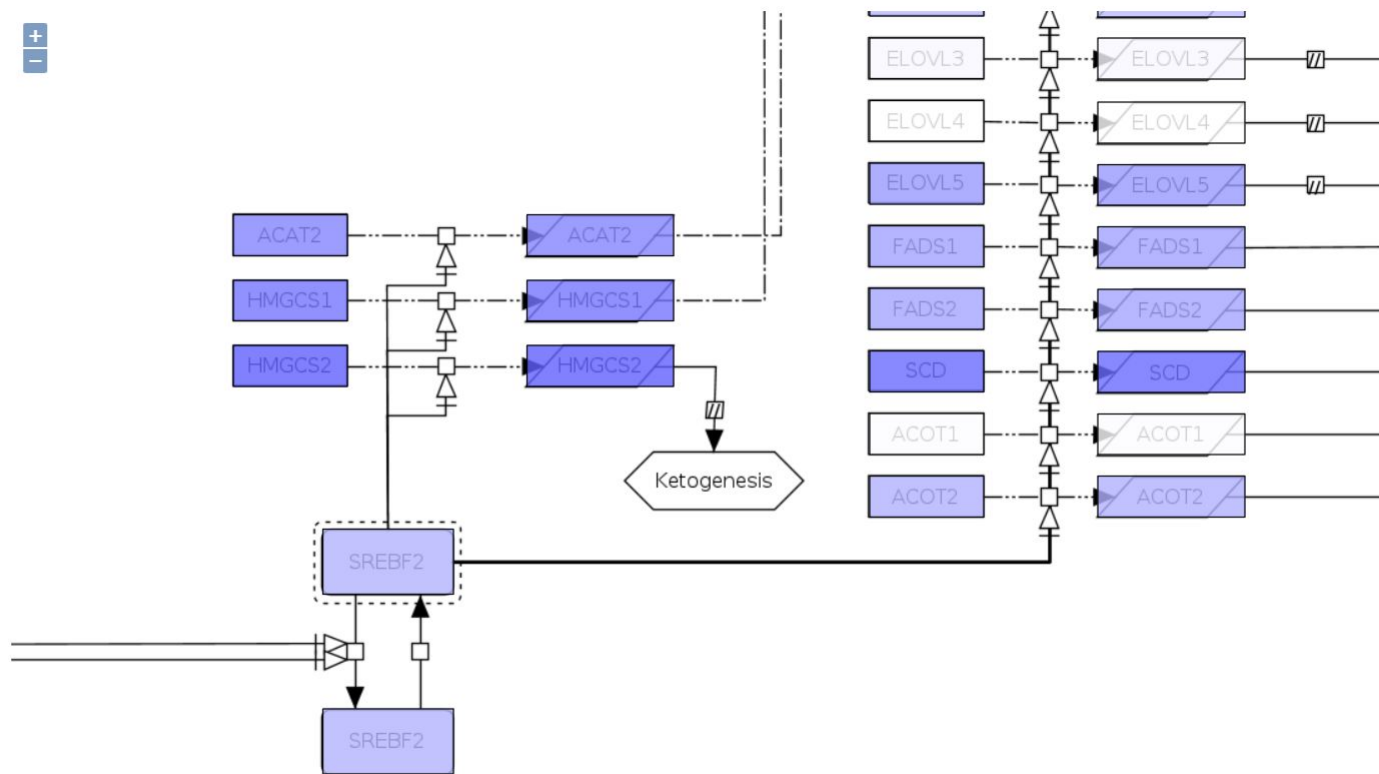
Description: Acetaminophen (paracetamol), also commonly known as *Tylenol*, is the most commonly taken analgesic worldwide and is recommended as first-line therapy in pain conditions by the World Health Organization (WHO) [A176318]. It is also used for its antipyretic effects, helping to reduce fever [F4124]. This drug was initially approved by the U.S. FDA in 1951 [FDA label] and is available in a variety of forms including syrup form, regular tablets, effervescent tablets, injection, suppository, and other forms [L5756, L5774, F4124], [FDA label]. Acetaminophen is often found combined with other drugs in more than 600 over the counter (OTC) allergy medications, cold medications, sleep



ONTOX ontology maps

SEARCH OVERLAYS SUBMAPS INFO

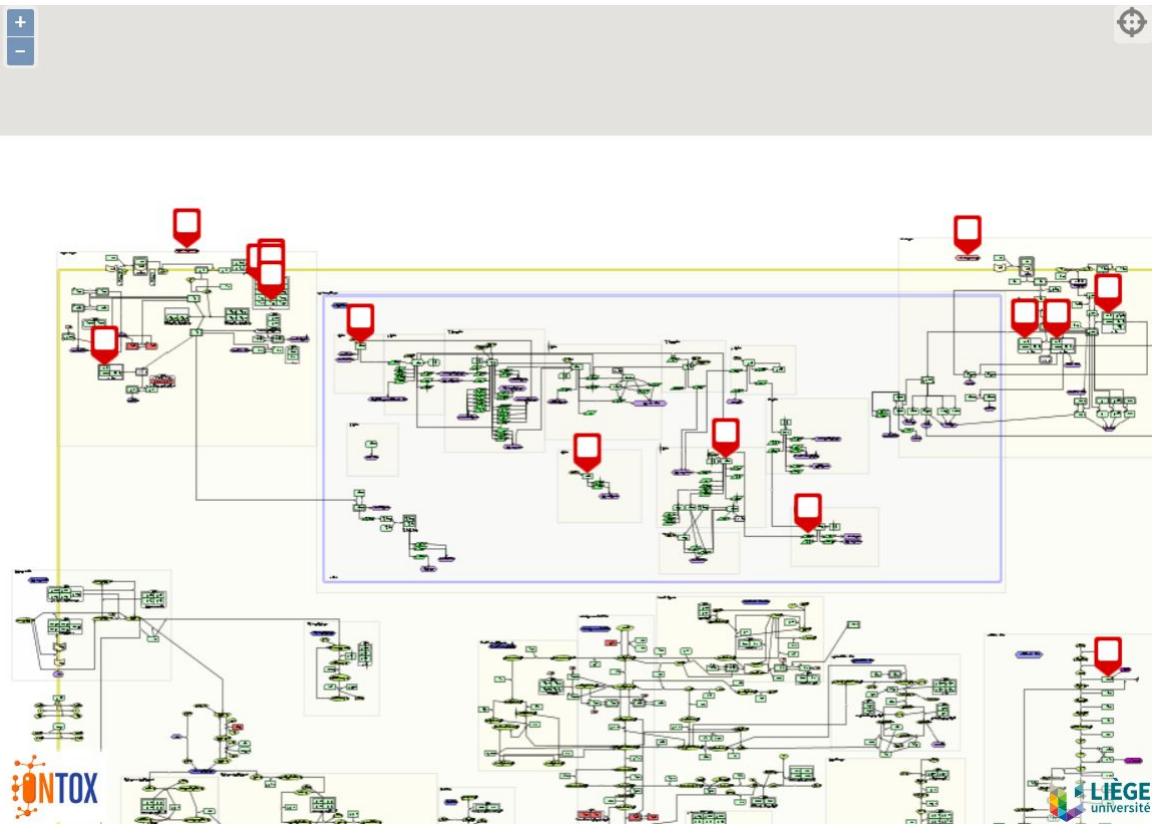
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1		<input type="checkbox"/>	
2	hepatocytes_rna_singlecell_hpa	<input checked="" type="checkbox"/>	
3		<input type="checkbox"/>	
4		<input type="checkbox"/>	
5		<input type="checkbox"/>	
6		<input type="checkbox"/>	
7		<input type="checkbox"/>	



USER PROVIDED OVERLAYS:

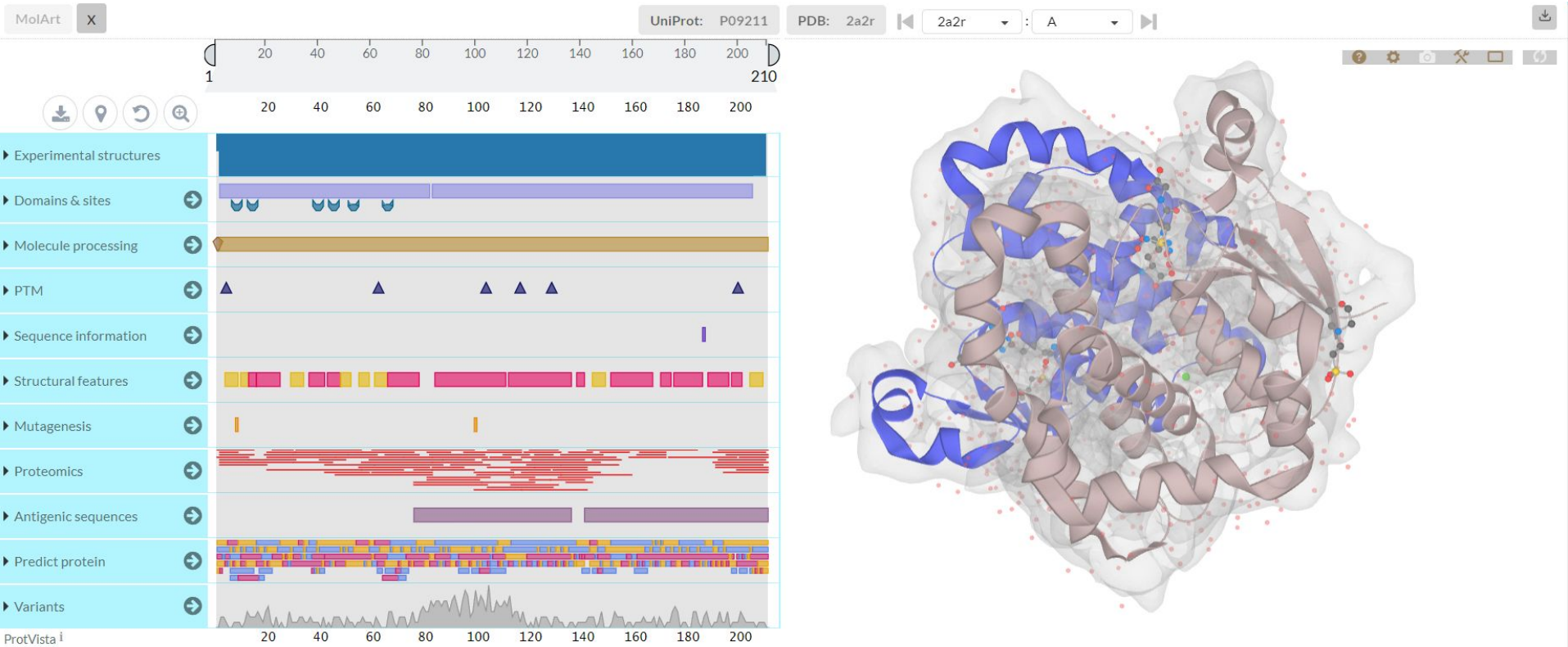
Powered by MINERVA Platform (v17.0.2)

ONTOX ontology maps



		complication, wound deiscence, wound infection	
Daliresp	ADVERSE REACTIONS	Abdominal pain, Acute kidney injury, Angioedema, Anxiety, Atrial fibrillation, Back pain, Decreased appetite, Depression, Diarrhoea, Dizziness, Dyspepsia, Gastritis, Headache, Hypersensitivity, Influenza, Insomnia, Lung neoplasm malignant, Muscle spasms, Nausea, Pancreatitis acute, Prostate cancer, Psychiatric symptom, Rash, Rhinitis, Sinusitis, Suicidal behaviour, Tremor, Urinary tract infection, Urticaria, Vomiting, Weight decreased, Weight decreased	PDE4A, PDE4C, PDE4D
Daliresp	WARNINGS AND PRECAUTIONS	Anxiety, Completed suicide, Depression, Insomnia, Psychiatric symptom, Suicidal behaviour, Suicidal ideation, Suicide attempt, Weight decreased, Weight decreased	PDE4A, PDE4C, PDE4D
DUREZOL	ADVERSE	Anterior chamber cell, Anterior chamber flare, Application site discomfort, Application site irritation, Blepharitis, Cataract subcapsular, Ciliary hyperaemia, Ciliary hyperaemia, Conjunctival hyperaemia, Conjunctival oedema, Corneal oedema, Corneal pigmentation, Corneal striae, Dry eye, Episcleritis, Eye infection, Eye inflammation, Eye irritation, Eye pain, Eye pruritus, Eye symptom, Eyelid irritation,	NR3C1

ONTOX ontology maps



Multi-layered ontology maps

- **Integration with adverse outcome pathways (AOPs)**
 - ✓ Use PMs as benchmarks to **fill gaps** in AOPs
 - ✓ **AOP networks** designed using **SBGN representation**
 - Data integration between physiological maps and AOPs
 - Standardization of AOPs and interoperability
- **Integration with other data (chemical, kinetic, omics, etc.)**
 - ✓ Plug-ins displaying tables with annotated information
 - ✓ Overlay of data (e.g. omics, drug databases)

Conclusions

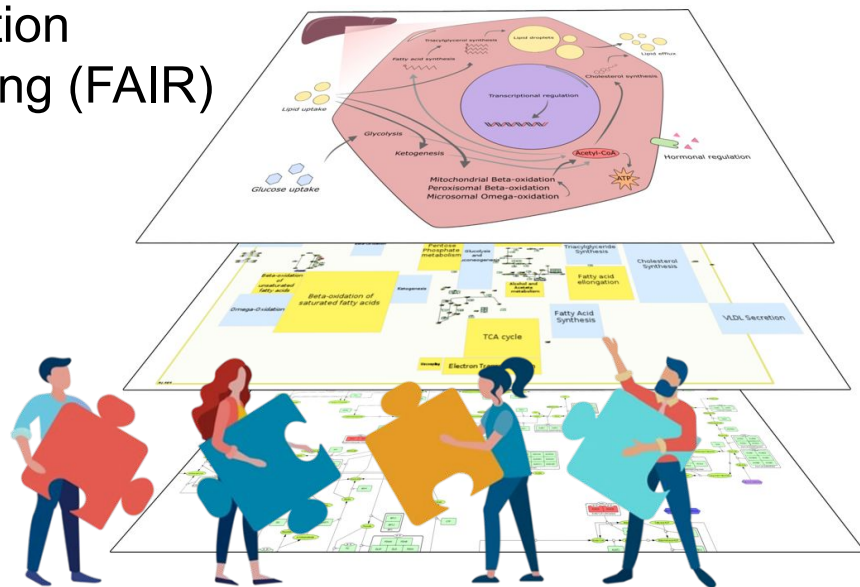
From physiological maps...

- Graphical representation of biological processes
- Standardized notation
- Guidelines for data curation and annotation
- Tool for knowledge integration and sharing (FAIR)

... to ontology maps:

- Founded on PMs
- AOPs, chemical, kinetic, omics data
- Mechanistic risk assessment

Large-scale community effort



Resources

systems medicine disease maps

Disease maps for specific diseases

Liver steatosis
★★★★☆

Resource: Liver Lipid Metabolism Physiological Map
Contact: Luiz Ladeira, University of Liège, Liège, Belgium

Cholestasis
★★★★☆

Resource: Liver Bile Secretion Physiological Map
Contact: Luiz Ladeira, University of Liège, Liège, Belgium

Cross-disease projects

Neural tube closure
defects ★★★★★

Resource: Neural Tube Closure Physiological Map
Contact: Alessio Gamba, University of Liège, Liège, Belgium
Publications: PubMed

Heusinkveld, et al. 2021. [Doi: 10.1016/j.reprotox.2020.09.002](https://doi.org/10.1016/j.reprotox.2020.09.002)

Kidney crystallopathy
and tubular necrosis
★★★★☆

Resource: Nephron Physiological Map
Diseases: kidney crystallopathy, tubular necrosis
Contact: Alessio Gamba, University of Liège, Liège, Belgium

Cognitive function
defects ★★★★★

Resource: Brain Development Physiological Map
Contact: Luiz Ladeira, University of Liège, Liège, Belgium

Resources



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Thank you for your attention

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