Metabolic visualisation of Recon3Map with Minerva

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INTRODUCTION

The visualisation of metabolic pathways is an essential tool to understand the biological meaning underlying COBRA metabolic models. This would allow the user to visualise what can not be appreciated at first sight by directly looking to the model outputs. Here we present a visualisation through Recon3Map [1] a visualisation of human metabolism, derived from Recon3 [2], using the Minerva platform [3]. Other human models and maps can be found at the Virtual Metabolic Human [4] website: http://www.vmh.life.

EQUIPMENT SETUP

In order to access remotely to Recon3Map, the user has to be registered. To obtain your credentials, you must access the ADMIN area and request an account. To access ReconMap follow the link: http://www.vmh.life .

Then, use your credentials to remotely access to http://www.vmh.life

```
load('minerva.mat')
minerva.minervaURL = 'http://www.vmh.life/minerva/index.xhtml';
minerva.map = 'ReconMap-3';
if 1
    %minerva.login = 'user_name';
    %minerva.password = 'user_password';
    minerva.login = 'cobratoolbox-test';
    minerva.password = 'test';
else
    minerva.login = 'ronan.fleming';
    minerva.password = 'ronan.fleming1';
    %minerva.login = 'cobratoolbox-test';
    %minerva.password = 'test';
```

```
    end
minerva.googleLicenseConsent = 'true';
```

Initialise the Cobra Toolbox.

A specific solver might be required (depending on the analysis you want to realise in the COBRA model).

```
changeCobraSolver('ibm_cplex','QP');
```

```
 > changeCobraSolver: IBM ILOG CPLEX interface added to MATLAB path.
```

```
changeCobraSolver('ibm_cplex', 'LP');
```

```
 > changeCobraSolver: IBM ILOG CPLEX interface added to MATLAB path.
```

Load your generic metabolic model. Recon's most recent version "Recon2.04" can be freely downloaded from http://www.vmh.life

```
model = load('Recon3DModel_301.mat')
```

```
model = struct with fields:

    model: [1×1 struct]
```

```
if isfield(model,'model')
    model=model.model;
end
```

PROCEDURE

1. Overlay a flux distribution

As an example of layout, we would like to see the the fluxes when maximizing ATP production through complex V (ATP synthase) in the Electron Transport Chain. To do so, we use Flux Balance Analysis (FBA) and set as an objective function the reaction responsible of this process (`'ATPS4m'`).

`ChangeObjective` function, changes the objective function of a constraint-based model

`optimizeCbModel` function solves a flux balance analysis problem.

```
formula = printRxnFormula(model, 'ATPS4mi')
```

```
ATPS4mi     adp[m] + pi[m] + 4 h[i]     ->    h2o[m] + 3 h[m] + atp[m]
formula = 1×1 cell array

    {'adp[m] + pi[m] + 4 h[i]  -> h2o[m] + 3 h[m] + atp[m] '}
```

```
model_atp_production = model  % re-name the model to do not modify the
original one.
```

```
model_atp_production = struct with fields:
                    S: [5835×10600 double]
                 mets: {5835×1 cell}
                    b: [5835×1 double]
```

```
                csense: [5835×1 char]
                  rxns: {10600×1 cell}
                    lb: [10600×1 double]
                    ub: [10600×1 double]
                     c: [10600×1 double]
                osense: -1
                 genes: {2248×1 cell}
                 rules: {10600×1 cell}
            metCharges: [5835×1 int64]
           metFormulas: {5835×1 cell}
             metSmiles: {5835×1 cell}
              metNames: {5835×1 cell}
             metHMDBID: {5835×1 cell}
         metInChIString: {5835×1 cell}
             metKEGGID: {5835×1 cell}
           metPubChemID: {5835×1 cell}
            description: 'Recon3DModel.mat'
                grRules: {10600×1 cell}
             rxnGeneMat: [10600×2248 double]
      rxnConfidenceScores: [10600×1 double]
              rxnNames: {10600×1 cell}
              rxnNotes: {10600×1 cell}
           rxnECNumbers: {10600×1 cell}
          rxnReferences: {10600×1 cell}
              rxnKEGGID: {10600×1 cell}
             subSystems: {10600×1 cell}
             metCHEBIID: {5835×1 cell}
               metPdMap: {5835×1 cell}
             metReconMap: {5835×1 cell}
                modelID: 'Recon3DModel'
                 rxnCOG: {10600×1 cell}
       rxnKeggOrthology: {10600×1 cell}
             rxnReconMap: {10600×1 cell}
                version: 'Recon3D_01'
             PleaseCite: 'Brunk et al, Nat Biotech, 2018; doi:10.1038/nbt.4072'
```

```
model_atp_production = changeObjective(model_atp_production, 'ATPS4mi');
```

Regularised flux balance analysis

```
solution_atp_prod_max_regularised = optimizeCbModel(model_atp_production,
'max',1e-6);
```

Sparse flux balance anaysis

```
solution_atp_prod_max_sparse = optimizeCbModel(model_atp_production,
'max','zero');
```

This reaction, (S)-2-Methylbutanoyl Coenzyme A:Acceptor 2, 3-Oxidoreductase Valine, Leucine And Isoleucine Degradation ('r0603') is expressed in the regularised flux balance analysis solution, but not in the sparse flux balance analysis solution.

```
formula = printRxnFormula(model, 'r0603');
```

```
r0603    2mbcoa[m] + q10[m]    ->    2mb2coa[m] + q10h2[m]
```

```
nnz(solution_atp_prod_max_regularised.v)
```

```
ans = 10600
```

```
solution_atp_prod_max_regularised.v(strcmp(model.rxns,'r0603'))
```

```
ans = 2.3688
```

```
nnz(solution_atp_prod_max_sparse.v)
```

```
ans = 41
```

```
solution_atp_prod_max_sparse.v(strcmp(model.rxns,'r0603'))
```

```
ans = 0
```

ANTICIPATED RESULTS

The `buildFluxDistLayout` function creates a layout that is automatically sent to the ReconMap website. After this, you can visualise your layout in http://www.vmh.life. Use your credentials to log in as it is previously explained. Select your input map (minerva.model) and go to "overlays" section to find your layout.
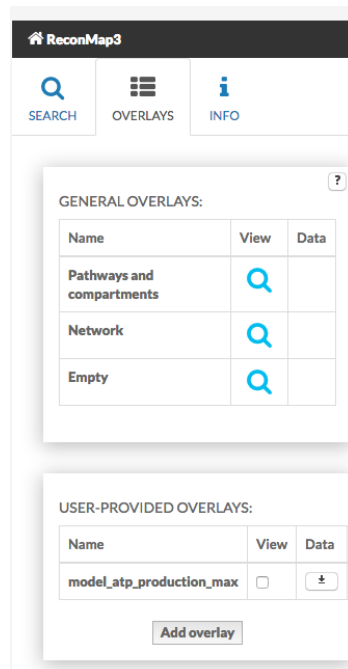

```
serverResponse = buildFluxDistLayout(minerva, model,
solution_atp_prod_max_sparse, 'atp_prod_max_sparse_2020','#6617B5')
```

```
Overlay generated successfully.
serverResponse =
'Overlay generated successfully.'
```

```
%TODO
% why does this does not work at present?
% perhaps because the size of the curl post is too large?
if 0
```

```
    serverResponse = buildFluxDistLayout(minerva, model,
solution_atp_prod_max_regularised, 'atp_prod_max_2020','#6617B5')
end
```

If everything is correctly defined you should get a structure with 2 values. If everything works fine, the output of this function should be:

`[1] 'Overlay generated successfully.'`

If there is any error, the message obtained will display:

`[0] 'Overlay NOT generated successfully.'`
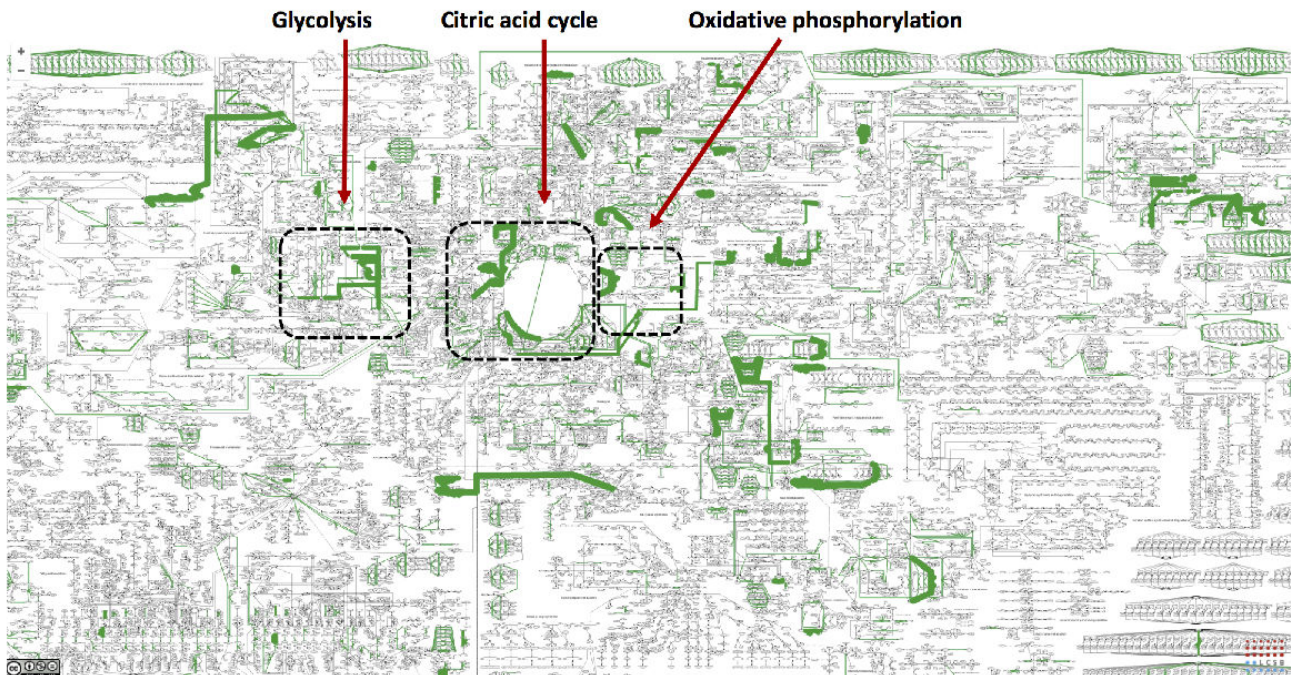
Note: If the "layout name" selected has been already given, an error might appear. Please, try to give a new layout name each time you run the code.

`[0]    'ERROR. Layout with given identifier ("atp_productio…'`

2. Overlay a SubSystem

There is also the possibility to highlight a specific subSystems by using the function `generateSubsytemsLayout`. A subSystem is a group of metabolic reactions involved in the same metabolic pathway, such as glycolysis, Oxidative phosphorylation, citric acid cycle, etc. Add the name of a specific subSystem you want to highlight from the COBRA model (see the example, TCA cycle), and the color reference.

```
generateSubsytemsLayout(minerva, model, 'Citric acid cycle', '#6617B5');

Overlay generated successfully.
```
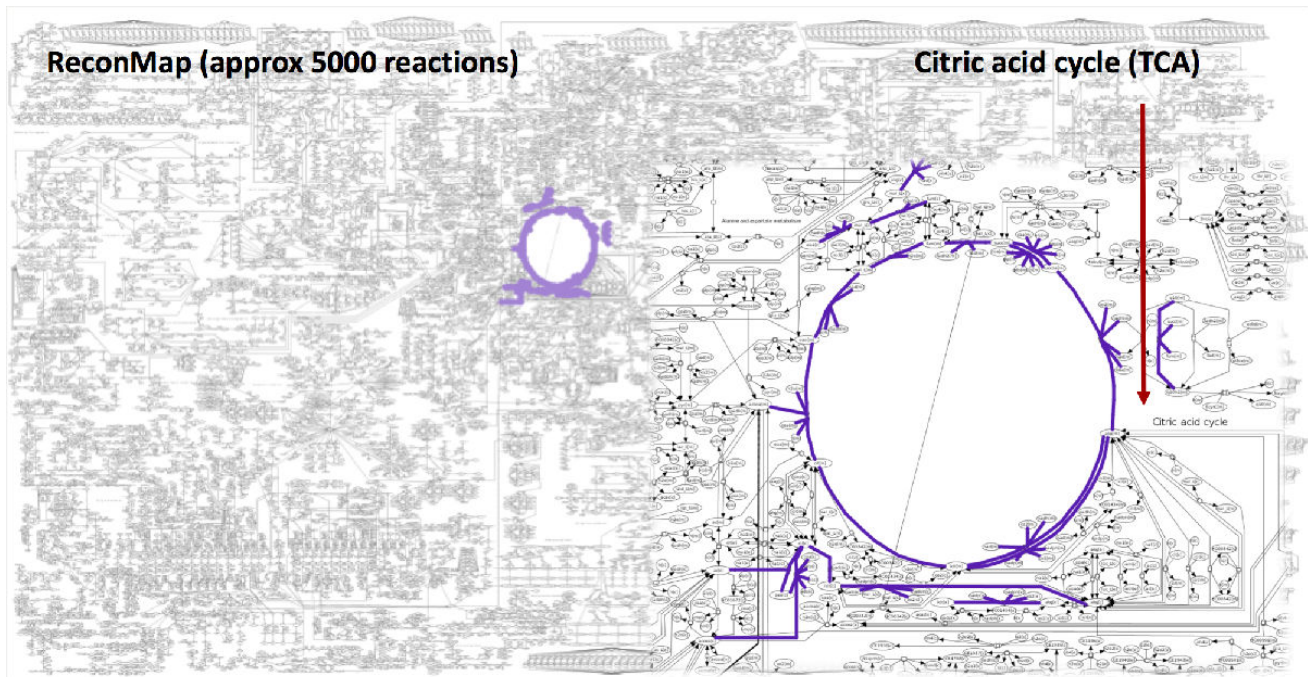
Alternatively, the user can generate a layout of all common subSystems between model and map using the function `generateSubsystemLayouts`.

Note: every single layout can be observed individually, or merged with other layouts. Therefore, making possible the visualization of several layouts at the same time.

RECONMAPS

There is an additional set of maps available in VMH consistent with the content of Recon 3D [3]. ReconMap3 is the general map that follows the same approach as the previous iteration, but we have also included 6 organelle-specific maps. You can see each of these by clicking on the corresponding button on the interface as shown below.

ReconMaps

To navigate ReconMap3 click the button below. You can also navigate any of the 6 organelle maps by clicking the corresponding button in the image!
Tip: when switching between different organelle maps a page refresh is sometimes necessary.

ReconMap3
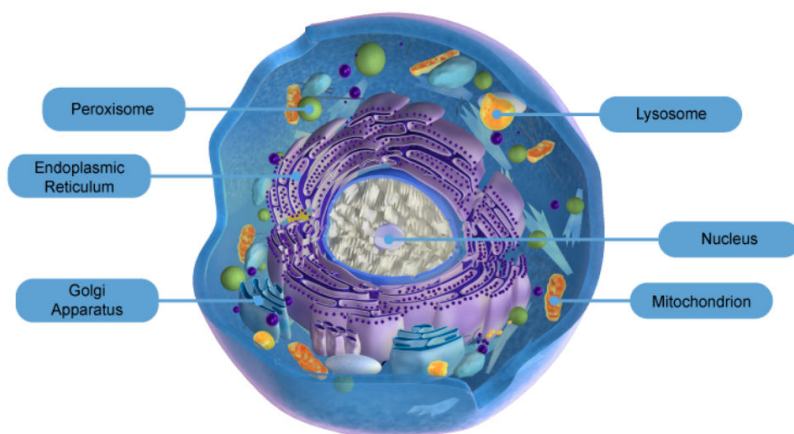
To submit flux distributions to specific maps, users just need to change the map variable from the minerva struct with the identifier of the desired map using one of the lines of code displayed below:

```
minerva.map = 'ReconMap-3'; % ReconMap 3
minerva.map = 'reticulum'; % Endoplasmic reticulim map
minerva.map = 'peroxisome'; % Peroxisome map
minerva.map = 'nucleus'; % Nucleus map
minerva.map = 'mitochondrion'; % Mitochondrion map
minerva.map = 'lysosome'; % Lysosome map
minerva.map = 'golgi'; % Golgi apparatus map
```

3. Manual upload of overlays

It is also possible to manually submit your overlays. For this purpose, you can generate a tab delimited file with the necessary information using the `printInRecon3Dmap` function.

```
%function printInRecon3Dmap(rxnList, colorValues, outputDir)
% This function generates a TXT file that can be integrated in the VMH
% database (vmh.life) to overlay certain reactions
```

Overlay File format:

To upload reaction fluxes in Recon2Map use the following tab delimited format:

name   reactionIdentifier   lineWidth   color

HEX1   2   #57c657

If you are uploading a file to Recon3Map3 and newer versions, it is necessary to add a prefix "R_" to each reaction identifier.

name   reactionIdentifier   lineWidth   color

R_HEX1   2   #57c657

Steps to upload an Overlay File:

1. **Login** to https://www.vmh.life/#reconmap with the same credentials as used above.
2. Select **Overlays > User-provided overlays** section
3. Select **Add overlay**
4. Select **Choose File** to navigate to a local file to uploa.

Note: To see uploaded overlay agree to the Google Maps terms.

For more information on MINERVA's functionalities check MINERVA' manual at: https://minerva.pages.uni.lu/doc/

REFERENCES:

[1] Alberto Noronha, Anna Dröfn Daníelsdóttir, Piotr Gawron, Freyr Jóhannsson, Sofía Jónsdóttir, Sindri Jarlsson, Jón Pétur Gunnarsson, Sigurður Brynjólfsson, Reinhard Schneider, Ines Thiele, and Ronan M. T. Fleming. ReconMap: an interactive visualization of human metabolism. Bioinformatics , 33(4):605607, February 2017.

[2] Elizabeth Brunk, Swagatika Sahoo, Daniel C Zielinski, Ali Altunkaya, Andreas Dräger, Nathan Mih, Francesco Gatto, Avlant Nilsson, German Andres Preciat Gonzalez, Maike Kathrin Aurich, Andreas Prli , Anand Sastry, Anna D Danielsdottir, Almut Heinken, Alberto Noronha, Peter W Rose, Stephen K Burley, Ronan M T Fleming, Jens Nielsen, Ines Thiele & Bernhard O Palsson. Recon3D enables a three-dimensional view of gene variation in human metabolism. *Nature Biotechnology* volume 36, pages 272–281 (2018)

[3] P. Gawron, M. Ostaszewski, V. Satagopam, S. Gebel, A. Mazein, M. Kuzma, S. Zorzan, F. McGee, B. Otjacques, R. Balling, and R. Schneider "MINERVA—a platform for visualization and curation of molecular interaction networks" in npj Systems Biology and Applications, vol. 2, p. 16020, Sep. 2016, doi:10.1038/npjsba.2016.20.

[4] Noronha et al., "The Virtual Metabolic Human database: integrating human and gut microbiome metabolism with nutrition and disease", Nucleic Acids Research (2018); https://academic.oup.com/nar/advance-article/doi/10.1093/nar/gky992/5146204