

# Convert a reconstruction into a flux balance analysis model

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**Reviewers:**

## INTRODUCTION

Even with quality control during the reconstruction process, it is not appropriate to assume that any reconstruction can be converted directly into a model and used to make predictions. A model must satisfy certain assumptions before it can be used to make reliable predictions. Depending on the type of model, these assumptions will be different. Each assumption should be chemically or biologically motivated and expressed in an unambiguous manner and preferably both intuitively and mathematically. Flux balance analysis is a mathematical method widely used for studying genome-scale biochemical network. Here one aims to predict steady-state reaction fluxes, where there is a balance between production and consumption of each molecular species that is not exchanged across the specified boundary of a system. In this situation, one might obtain erroneous predictions if the system boundary is incorrectly specified. If a reconstruction contains one or more supposedly mass balanced reactions, but which are actually not mass balanced, such reactions in a model can lead to inadvertent leakage of a metabolite from the model, in violation of mass balance. Similarly, when generating a model for flux balance analysis, it is important to ensure that the network is flux consistent, that is, each reaction can carry a non-zero steady state flux.

Given a reconstruction with  $\hat{m}$  reactants involved in  $\hat{n}$  reactions, this tutorial demonstrates a method to identify and extract the largest subset of the reconstruction whose internal reactions are both stoichiometrically and flux consistent and whose external reactions are flux consistent. This model is then mathematically consistent with the basic requirements for generation of predictions using flux balance analysis. The identification of the component of the reconstruction that does not satisfy the aforementioned modelling conditions is also useful for targeting reconstruction effort towards resolving stoichiometric inconsistency or resolving flux inconsistency. The example used in this tutorial illustrates the process of extracting a model consistent with flux balance analysis, from a ReconX reconstruction.

## PROCEDURE

### Select reconstruction to convert into a model and enter parameters

Load the ReconX reconstruction, and save the original reconstruction in the workspace, unless it is already loaded into the workspace.

```
clear model
if ~exist('modelOrig','var')
    %select your own model, or use Recon2.0model instead
    if 1
        filename='Recon3D_301.mat'
        load(filename);
        model=Recon3D;
    else
        filename='Recon2.0model.mat';
```

```

        if exist('Recon2.0model.mat','file')==2
            model = readCbModel(filename);
        end
    end
    model.csense(1:size(model.S,1),1)='E';
    modelOrig = model;
else
    model=modelOrig;
end

```

```

filename =
'Recon3D_301.mat'

```

Set the level of printing, zero for silent, higher for more output.

```

printLevel=2;

```

Choose the directory to place the results

```

basePath='~/work/sbgCloud/';
%resultsPath=[basePath '/programReconstruction/projects/recon2models/results/
reconXs/' model.modelID];
resultsPath=[basePath '/courses/2019_Leiden_COBRA/practicalsDemo/Day4/'
model.modelID];
resultsFileName=[resultsPath filesep model.modelID];

```

Create and enter the folder for the results if it does not already exist

```

if ~exist(resultsPath,'dir')
    mkdir(resultsPath)
end
cd(resultsPath)

```

Optionally create a diary to save the output in case it is very long, this makes it easier to search, especially when debugging the process during the early stages.

```

if 0
    diary([resultsFileName '_diary.txt'])
end

```

## Overview some of the key properties of the reconstruction

Noting the initial size of the reconstruction is useful for comparisons later with subsets derived according to mathematical specifications.

```

[nMet,nRxn]=size(model.S);
fprintf('%6s\t%6s\n','#mets','#rxns')

```

```

#mets      #rxns

```

```

fprintf('%6u\t%6u\t%s\n',nMet,nRxn,' totals.')

```

Make sure the stoichiometric matrix is stored in a sparse format as this accelerates computations with large networks

```
model.S=sparse(model.S);
```

## Check in case the reconstruction is a model that is already ready for flux balance analysis

There is no need to run this live script any further if the reconstruction already satisfies the conditions necessary for flux balance analysis. That is if all internal reactants and reactions are stoichiometrically consistent, and all reactions are flux consistent, then the reconstruction satisfies the criteria to designate it a model ready for flux balance analysis.

SIntMetBool                m x 1 Boolean of metabolites heuristically though to be involved in mass balanced reactions.

SIntRxnBool                n x 1 Boolean of reactions heuristically though to be mass balanced.

SConsistentMetBool        m x 1 Boolean vector indicating consistent mets

SConsistentRxnBool        n x 1 Boolean vector indicating consistent rxns

fluxConsistentMetBool     m x 1 Boolean vector indicating flux consistent mets

fluxConsistentRxnBool     n x 1 Boolean vector indicating flux consistent rxns

```
if all(isfield(model,{ 'SIntMetBool', 'SIntRxnBool', 'SConsistentMetBool', ...
'SConsistentRxnBool', 'fluxConsistentMetBool', 'fluxConsistentRxnBool' })))
    if all(model.SIntMetBool & model.SConsistentMetBool)...
        && nnz(model.SIntRxnBool &
model.SConsistentRxnBool)==nnz(model.SIntRxnBool)...
        && all(model.fluxConsistentMetBool)...
        && all(model.fluxConsistentRxnBool)
        fullyStoichAndFluxConsistent=1;
        fprintf('%s\n', 'Reconstruction is a model that is already ready for
flux balance analysis')
    end
    return
else
    fullyStoichAndFluxConsistent=0;
    fprintf('%s\n', 'Reconstruction must be tested to check if it is ready
for flux balance analysis')
end
```

Reconstruction must be tested to check if it is ready for flux balance analysis

## Manually remove certain reactions from the reconstruction

Before attempting to algorithmically remove stoichiometrically or flux inconsistent supposed internal reactions from a reconstruction to generate a model, there is an option to review the content of the reconstruction and manually identify reactions for removal. That is, there are two options:

A. Skip manual review of reconstruction content. Move to the next step.

B. Review the content of the reconstruction and omit any reactions that are assumed to be stoichiometrically or flux inconsistent. With respect to stoichiometric inconsistency, such reactions may be obviously mass imbalanced and not satisfy the heuristic conditions for identification as an external reaction. Alternatively, such reactions may be identified by a previous pass through of this tutorial as being of unknown stoichiometric consistency (`model.unknownSConsistencyRxnBool(j)==1`), after the largest stoichiometrically consistent subset of the network has been identified. This is an iterative process where multiple rounds of identification of the largest stoichiometrically consistent set and manual curation of the remainder that is of unknown stoichiometric consistency is necessary.

```
if strcmp(filename, 'Recon3.0model')
    modelOrig=model;
    if 0
        if 1
            %Rename some of the biomass reactions to make them more
            obviously exchange
            %reactions
            model.rxns{strcmp(model.rxns, 'biomass_reaction')}=
            'EX_biomass_reaction';
            model.rxns{strcmp(model.rxns, 'biomass_maintenance')}=
            'EX_biomass_maintenance';
            model.rxns{strcmp(model.rxns, 'biomass_maintenance_noTrTr')}=
            'EX_biomass_maintenance_noTrTr';

            %ATP hydrolysis is not imbalanced like all the other demand
            reactions so
            %give it a different accronym ATPM = ATP Maintenance
            bool=strcmp('DM_atp_c_', model.rxns);
            model.rxns{bool}='ATPM';
        end
        [model, removeMetBool, removeRxnBool] =
        manuallyAdaptRecon3(model, printLevel);
    else
        [model, removeMetBool, removeRxnBool] =
        manuallyAdaptRecon3Ines(model, printLevel);
    end
    [nMet0, nRxn0]=size(modelOrig.S);
    [nMet, nRxn]=size(model.S);
    if nMet0==nMet && nRxn0==nRxn && printLevel>0
        fprintf('%s\n', '--- Manually removing rows and columns of the
        stoichiometric matrix---')
        fprintf('%6s\t%6s\n', '#mets', '#rxns')
        fprintf('%6u\t%6u\t%6s\n', nMet0, nRxn0, ' totals.')
        fprintf('%6u\t%6u\t%6s\n', nMet0-nMet, nRxn0-nRxn, ' manually removed.')
```

```

        fprintf('%6u\t%6u\t%s\n',nMet,nRxn,' remaining.')
    end
end

```

## Remove any trivial rows and columns of the stoichiometric matrix

Remove any zero rows or columns of the stoichiometric matrix

```

modelOrig=model;
model=removeTrivialStoichiometry(model);
[nMet0,nRxn0]=size(modelOrig.S);
[nMet,nRxn]=size(model.S);
if nMet0==nMet && nRxn0==nRxn && printLevel>0
    fprintf('%s\n','---Checking for Remove any trivial rows and columns of
the stoichiometric matrix----')
    fprintf('%6s\t%6s\n','#mets','#rxns')
    fprintf('%6u\t%6u\t%s\n',nMet0,nRxn0,' totals.')
    fprintf('%6u\t%6u\t%s\n',nMet0-nMet,nRxn0-nRxn,' duplicates removed.')
    fprintf('%6u\t%6u\t%s\n',nMet,nRxn,' remaining.')
end

```

```

---Checking for Remove any trivial rows and columns of the stoichiometric matrix----
#mets      #rxns
 8399      13543      totals.
    0         0      duplicates removed.
 8399      13543      remaining.

```

Check for duplicate columns by detecting the columns of the S matrix that are identical upto scalar multiplication.

```

modelOrig=model;
dupDetectMethod='FR';
dupDetectMethod='S';
removeFlag=0;
[modelOut,removedRxnInd, keptRxnInd] =
checkDuplicateRxn(model,dupDetectMethod,removeFlag,printLevel-2);

```

Remove any duplicate reactions, and uniquely involved reactants, from the stoichiometric matrix.

```

if length(removedRxnInd)>0
    irrevFlag=0;
    metFlag=1;
    %set all reactions reversible that are duplicates
    model.lb(removedRxnInd)=-model.ub(removedRxnInd);
    %remove duplicates
    model = removeRxns(model,model.rxns(removedRxnInd),irrevFlag,metFlag);
end

```

Display the statistics on the duplicate reactions,

```

[nMet0,nRxn0]=size(modelOrig.S);

```

```
[nMet,nRxn]=size(model.S);
if nMet0==nMet && nRxn0==nRxn && printLevel>0
    fprintf('%s\n','---Remove any duplicate reactions---')
    [nMet0,nRxn0]=size(modelOrig.S);
    [nMet,nRxn]=size(model.S);
    fprintf('%6s\t%6s\n','#mets','#rxns')
    fprintf('%6u\t%6u\t%s\n',nMet0,nRxn0,' totals.')
    fprintf('%6u\t%6u\t%s\n',nMet0-nMet,nRxn0-nRxn,' duplicates removed.')
    fprintf('%6u\t%6u\t%s\n',nMet,nRxn,' remaining.')
end
```

```
---Remove any duplicate reactions---
#mets      #rxns
 8399      13543      totals.
      0         0      duplicates removed.
 8399      13543      remaining.
```

## Remove any duplicate reactions upto protons

Remove reactions reactions that differ only in the number of protons involved as substrates or products. Also remove exclusively involved reactants.

Save a temporary model for testing, before making any changes.

```
modelH=model;
```

Find the proton indicies in different compartments. A proton, with index i, is asumed to be represented by an abbreviation within model.mets{*i*} like h[\*], where \* denotes the compartment symbol.

```
nMetChars=zeros(length(modelH.mets),1);
for m=1:length(modelH.mets)
    nMetChars(m,1)=length(modelH.mets{m});
end
protonMetBool=strncmp(modelH.mets,'h',1) & nMetChars==length('h[*]');
if printLevel>2
    disp(modelH.mets(protonMetBool))
end
```

Zero out the proton stoichiometric coefficients from the temporary model for testing

```
modelH.S(protonMetBool,:)=0;
```

Check for duplicate columns, upto protons, by detecting the columns of the S matrix that are identical upto scalar multiplication.

```
dupDetectMethod='FR';
removeFlag=0;
[modelOut,removedRxnInd, keptRxnInd] =
checkDuplicateRxn(modelH,dupDetectMethod,removeFlag,printLevel-1);
```

```
Checking for reaction duplicates by stoichiometry (up to orientation) ...
Keep:      25HVITD2t      25hvitd2[c]      ->      25hvitd2[e]
```

```

Duplicate: 25HVITD2tin 25hvitd2[e] -> 25hvitd2[c]
Keep: 25HVITD2tin_m 25hvitd2[c] -> 25hvitd2[m]
Duplicate: 25HVITD2tm 25hvitd2[m] -> 25hvitd2[c]
Keep: 25HVITD3t 25hvitd3[c] -> 25hvitd3[e]
Duplicate: 25HVITD3tin 25hvitd3[e] -> 25hvitd3[c]
Keep: 25HVITD3tin_m 25hvitd3[c] -> 25hvitd3[m]
Duplicate: 25HVITD3tm 25hvitd3[m] -> 25hvitd3[c]
Keep: 3MOBT2im 3mob[c] -> 3mob[m]
Duplicate: HMR_3746 3mob[c] <=> 3mob[m]
Keep: 5MTHFt 5mthf[e] <=> 5mthf[c]
Duplicate: MTHFte 5mthf[c] -> 5mthf[e]
Keep: ADNt adn[e] <=> adn[c]
Duplicate: ADNCNT3tc adn[e] <=> adn[c]
Keep: ADPRIBt adprib[e] -> adprib[c]
Duplicate: ADPRIBte adprib[c] <=> adprib[e]
Keep: ALAt4 nal[e] + ala_L[e] -> nal[c] + ala_L[c]
Duplicate: HMR_9605 nal[e] + ala_L[e] -> nal[c] + ala_L[c]
Keep: ALCD21_D nad[c] + 12ppd_R[c] -> nadh[c] + lald_D[c]
Duplicate: PPDOx nadh[c] + lald_D[c] -> nad[c] + 12ppd_R[c]
Keep: ALCD22_D nad[c] + lald_D[c] -> nadh[c] + mthgxl[c]
Duplicate: LALDO2x nadh[c] + mthgxl[c] -> nad[c] + lald_D[c]
Keep: ATPasel h2o[c] + atp[c] -> adp[c] + pi[c]
Duplicate: DM_atp_c_ h2o[c] + atp[c] -> adp[c] + pi[c]
Warning: BTNT2 has more than one replicate
Keep: BTNT2 btn[e] <=> btn[c]
Duplicate: BTNte btn[c] -> btn[e]
Keep: C14STRr nadph[r] + 44mctr[r] -> nadp[r] + 44mzym[r]
Duplicate: r0780 nadp[r] + 44mzym[r] <=> nadph[r] + 44mctr[r]
Keep: C160CPT1 crn[c] + pmtcoa[c] <=> coa[c] + pmtcrn[c]
Duplicate: C160CPT2rbc coa[c] + pmtcrn[c] <=> crn[c] + pmtcoa[c]
Keep: C161CPT2 coa[m] + hdcecrn[m] <=> crn[m] + hdcoa[m]
Duplicate: r0446 crn[m] + hdcoa[m] <=> coa[m] + hdcecrn[m]
Keep: C181CPT1 crn[c] + odecoa[c] <=> coa[c] + odecrn[c]
Duplicate: C181CPT2rbc coa[c] + odecrn[c] <=> crn[c] + odecoa[c]
Keep: CITtam cit[c] + mal_L[m] <=> cit[m] + mal_L[c]
Duplicate: HMR_4964 cit[c] + mal_L[m] -> cit[m] + mal_L[c]
Keep: CRNt crn[e] <=> crn[c]
Duplicate: CRNtHa crn[c] -> crn[e]
Keep: CRNtNa nal[e] + crn[e] -> nal[c] + crn[c]
Duplicate: CRNCT2te nal[c] + crn[c] <=> nal[e] + crn[e]
Keep: CRVNCtr crvnc[e] <=> crvnc[c]
Duplicate: CE0328te crvnc[c] <=> crvnc[e]
Keep: CYSt4 nal[e] + cys_L[e] -> nal[c] + cys_L[c]
Duplicate: CYSSNAT5tc nal[e] + cys_L[e] <=> nal[c] + cys_L[c]
Keep: CYTDt cytd[e] <=> cytd[c]
Duplicate: CYTDt2r cytd[e] <=> cytd[c]
Keep: DALAt2r ala_D[e] <=> ala_D[c]
Duplicate: ALA-DTDe ala_D[c] -> ala_D[e]
Keep: DMHPTCRNte dmhptcrn[c] <=> dmhptcrn[e]
Duplicate: DMHPTCRNtr dmhptcrn[e] <=> dmhptcrn[c]
Keep: DNDPt10m dadp[c] + dcdp[m] -> dcdp[c] + dadp[m]
Duplicate: DNDPt29m dcdp[c] + dadp[m] -> dadp[c] + dcdp[m]
Keep: DNDPt11m dadp[c] + dgdp[m] -> dgdp[c] + dadp[m]
Duplicate: DNDPt35m dgdp[c] + dadp[m] -> dadp[c] + dgdp[m]
Keep: DNDPt14m dtdp[m] + dudp[c] -> dtdp[c] + dudp[m]
Duplicate: DNDPt22m dtdp[c] + dudp[m] -> dtdp[m] + dudp[c]
Keep: DNDPt15m dgdp[m] + dudp[c] -> dgdp[c] + dudp[m]
Duplicate: DNDPt33m dgdp[c] + dudp[m] -> dgdp[m] + dudp[c]
Keep: DNDPt16m dadp[m] + dudp[c] -> dadp[c] + dudp[m]
Duplicate: DNDPt8m dadp[c] + dudp[m] -> dadp[m] + dudp[c]
Keep: DNDPt17m dcdp[m] + dudp[c] -> dcdp[c] + dudp[m]
Duplicate: DNDPt26m dcdp[c] + dudp[m] -> dcdp[m] + dudp[c]
Keep: DNDPt23m dgdp[m] + dtdp[c] -> dgdp[c] + dtdp[m]
Duplicate: DNDPt34m dgdp[c] + dtdp[m] -> dgdp[m] + dtdp[c]

```

```

Keep:      DNDPt24m      dadp[m] + dtdp[c]      ->      dadp[c] + dtdp[m]
Duplicate:  DNDPt9m      dadp[c] + dtdp[m]      ->      dadp[m] + dtdp[c]
Keep:      DNDPt25m      dcdp[m] + dtdp[c]      ->      dcdp[c] + dtdp[m]
Duplicate:  DNDPt27m      dcdp[c] + dtdp[m]      ->      dcdp[m] + dtdp[c]
Keep:      DNDPt28m      dcdp[c] + dgdpm[m]      ->      dgdpm[c] + dcdp[m]
Duplicate:  DNDPt36m      dgdpm[c] + dcdp[m]      ->      dcdp[c] + dgdpm[m]
Keep:      DOPAtu      dopa[e]      <=>      dopa[c]
Duplicate:  DOPAENT4tc      dopa[e]      <=>      dopa[c]
Keep:      EBP2r      zymstnl[r]      ->      lthstrl[r]
Duplicate:  r1381      lthstrl[r]      <=>      zymstnl[r]
Keep:      FE2t      fe2[e]      ->      fe2[c]
Duplicate:  FE2DMT1      fe2[e]      ->      fe2[c]
Keep:      FE2tm      fe2[c]      ->      fe2[m]
Duplicate:  HMR_5420      fe2[c]      ->      fe2[m]
Keep:      FUCFUCFUCGALACGLC13GALACGLCGAL14ACGLCGALGLUSIDete      fucfucfucgalacglc13galacglcgall14acglcg
Duplicate:  HMR_9651      fucfucfucgalacglc13galacglcgall14acglcggluside_hs[c]      <=>      fucfucfucgalac
Keep:      FUCFUCFUCGALACGLCGAL14ACGLCGALGLUSIDete      fucfucfucgalacglcgall14acglcggluside_hs[e]
Duplicate:  HMR_9645      fucfucfucgalacglcgall14acglcggluside_hs[c]      <=>      fucfucfucgalacglcgall14ac
Keep:      FUCGALFUCGALACGLCGALGLUSIDete      fucgalfucgalacglcggluside_hs[e]      <=>      fucgalfucgala
Duplicate:  HMR_9643      fucgalfucgalacglcggluside_hs[c]      <=>      fucgalfucgalacglcggluside_hs[e]
Keep:      GALFUCGALACGLCGAL14ACGLCGALGLUSIDete      galfucgalacglcgall14acglcggluside_hs[e]      <=>
Duplicate:  HMR_9646      galfucgalacglcgall14acglcggluside_hs[c]      <=>      galfucgalacglcgall14acglcg
Keep:      GALt1r      gal[e]      <=>      gal[c]
Duplicate:  GALt2_2      gal[e]      <=>      gal[c]
Keep:      GDPTg      gdp[c]      <=>      gdp[g]
Duplicate:  HMR_7743      gdp[c]      <=>      gdp[g]
Warning: GLCt1r has more than one replicate
Keep:      GLCt1r      glc_D[e]      <=>      glc_D[c]
Duplicate:  GLCGLUT2      glc_D[c]      ->      glc_D[e]
Keep:      GLNtm      gln_L[c]      ->      gln_L[m]
Duplicate:  HMR_5101      gln_L[c]      ->      gln_L[m]
Keep:      GLYC3Ptm      glyc3p[c]      ->      glyc3p[m]
Duplicate:  GLYC3Ptmc      glyc3p[m]      <=>      glyc3p[c]
Keep:      GLYt4      nal[e] + gly[e]      ->      nal[c] + gly[c]
Duplicate:  GLYSNAT5tc      nal[e] + gly[e]      <=>      nal[c] + gly[c]
Keep:      GSNT      gsn[e]      <=>      gsn[c]
Duplicate:  GSNT2r      gsn[e]      <=>      gsn[c]
Keep:      HIST4      nal[e] + his_L[e]      ->      nal[c] + his_L[c]
Duplicate:  HISSNAT5tc      nal[e] + his_L[e]      <=>      nal[c] + his_L[c]
Keep:      HISTiDF      his_L[e]      ->      his_L[c]
Duplicate:  HISCAT1      his_L[c]      <=>      his_L[e]
Keep:      HSD17B7r      nadph[r] + estrone[r]      ->      nadp[r] + estradiol[r]
Duplicate:  HMR_2041      nadph[r] + estrone[r]      ->      nadp[r] + estradiol[r]
Warning: Htg has more than one replicate
Keep:      Htg      <=>
Duplicate:  Htmi      ->
Keep:      INST      ins[e]      <=>      ins[c]
Duplicate:  INST2      ins[e]      <=>      ins[c]
Keep:      L_LACtcm      lac_L[c]      ->      lac_L[m]
Duplicate:  L_LACtm      lac_L[c]      ->      lac_L[m]
Keep:      LNLCCPT1      crn[c] + lnlccoa[c]      <=>      coa[c] + lnlccrn[c]
Duplicate:  LNLCCPT2rbc      coa[c] + lnlccrn[c]      <=>      crn[c] + lnlccoa[c]
Warning: NACUP has more than one replicate
Keep:      NACUP      nac[e]      ->      nac[c]
Duplicate:  NACHORCTL3le      nac[e]      ->      nac[c]
Keep:      NADHtpu      nadh[c]      ->      nadh[x]
Duplicate:  NADtpu      nadh[x]      ->      nadh[c]
Keep:      NAT      nal[e]      <=>      nal[c]
Duplicate:  NAT3_1      nal[c]      <=>      nal[e]
Keep:      NCAMUP      ncam[e]      ->      ncam[c]
Duplicate:  NCAMDe      ncam[c]      ->      ncam[e]
Keep:      NH4t3r      nh4[c]      <=>      nh4[e]
Duplicate:  NH4tb      nh4[e]      <=>      nh4[c]
Keep:      NOT      no[e]      <=>      no[c]

```



```

Duplicate:  NODe    no[c]    <=>    no[e]
Keep:      OCTAt   octa[e]    <=>    octa[c]
Duplicate:  OCTAt   octa[c]    <=>    octa[e]
Warning: ORNt4m has more than one replicate
Keep:      ORNt4m   orn[m] + citr_L[c]    <=>    orn[c] + citr_L[m]
Duplicate:  r2412   orn[c] + citr_L[m]    ->    orn[m] + citr_L[c]
Keep:      P5CRxm   nadh[m] + lpyr5c[m]    ->    nad[m] + pro_L[m]
Duplicate:  PRO1xm   nad[m] + pro_L[m]    ->    nadh[m] + lpyr5c[m]
Keep:      PItx     pi[c]    <=>    pi[x]
Duplicate:  HMR_5344 pi[c]    <=>    pi[x]
Keep:      PRODt2r   pro_D[e]    <=>    pro_D[c]
Duplicate:  PRO_Dtde pro_D[c]    <=>    pro_D[e]
Keep:      RIBt     rib_D[e]    <=>    rib_D[c]
Duplicate:  RIBt2    rib_D[e]    ->    rib_D[c]
Keep:      SRTNtu    srtn[e]    <=>    srtn[c]
Duplicate:  SRTNENT4tc srtn[e]    <=>    srtn[c]
Keep:      SUCCtp    succ[c]    <=>    succ[x]
Duplicate:  SUCCTD    succ[x]    <=>    succ[c]
Keep:      TAGt     tag_hs[e]    <=>    tag_hs[c]
Duplicate:  TAGHSTDe tag_hs[c]    ->    tag_hs[e]
Warning: THYMDt1 has more than one replicate
Keep:      THYMDt1   thymd[e]    ->    thymd[c]
Duplicate:  THMDt2r   thymd[e]    <=>    thymd[c]
Keep:      TRDRm     nadph[m] + trdox[m]    ->    nadp[m] + trdrd[m]
Duplicate:  r1433     nadp[m] + trdrd[m]    ->    nadph[m] + trdox[m]
Keep:      URIt     uri[e]    <=>    uri[c]
Duplicate:  URIt2r    uri[e]    <=>    uri[c]
Keep:      VITD3t    vitd3[c]    ->    vitd3[e]
Duplicate:  VITD3t2   vitd3[e]    ->    vitd3[c]
Warning: VITD3tm has more than one replicate
Keep:      VITD3tm    vitd3[m]    ->    vitd3[c]
Duplicate:  HMR_2116   vitd3[c]    <=>    vitd3[m]
Keep:      XOLEST2te  xolest2_hs[e]    <=>    xolest2_hs[c]
Duplicate:  XOLEST2HSTDle xolest2_hs[c]    ->    xolest2_hs[e]
Keep:      r0276     nh4[c] + nadp[c] + imp[c]    <=>    nadph[c] + gmp[c]
Duplicate:  GMPR     nadph[c] + gmp[c]    ->    nh4[c] + nadp[c] + imp[c]
Keep:      r0488     2 nadp[c] + coa[c] + mev_R[c]    <=>    2 nadph[c] + hmgcoa[c]
Duplicate:  HMGCOCARC 2 nadph[c] + hmgcoa[c]    ->    2 nadp[c] + coa[c] + mev_R[c]
Keep:      r0537     ethamp[c] + hxdcal[c]    ->    sphlp[c]
Duplicate:  SGPL11c   sphlp[c]    ->    ethamp[c] + hxdcal[c]
Keep:      r0561     coa[m] + 2mpdhl[m]    ->    ibcoa[m] + dhlam[m]
Duplicate:  RE3326M    ibcoa[m] + dhlam[m]    <=>    coa[m] + 2mpdhl[m]
Keep:      r0808     HC00004[c]    ->    HC00004[e]
Duplicate:  HC00004tle HC00004[e]    ->    HC00004[c]
Keep:      r0817     citr_L[c]    <=>    citr_L[e]
Duplicate:  CITRtr    citr_L[e]    <=>    citr_L[c]
Keep:      r0839     orot[e]    <=>    orot[c]
Duplicate:  OROte     orot[e]    ->    orot[c]
Keep:      r0899     ala_B[c]    <=>    ala_B[e]
Duplicate:  BALAPAT1tc ala_B[e]    ->    ala_B[c]
Keep:      r0913     icit[m] + mal_L[c]    <=>    mal_L[m] + icit[c]
Duplicate:  r2387     mal_L[m] + icit[c]    ->    icit[m] + mal_L[c]
Keep:      r0915     cit[c] + succ[m]    <=>    cit[m] + succ[c]
Duplicate:  r2382     cit[c] + succ[m]    ->    cit[m] + succ[c]
Keep:      r0944     spmd[c]    <=>    spmd[e]
Duplicate:  SPMTDe    spmd[e]    <=>    spmd[c]
Keep:      r1050     chsterol[e]    <=>    chsterol[c]
Duplicate:  CHOLESTDe chsterol[c]    ->    chsterol[e]
Keep:      r1067     his_L[l]    ->    his_L[c]
Duplicate:  HIShPTtc  his_L[l]    ->    his_L[c]
Keep:      r1078     tyr_L[c]    ->    tyr_L[m]
Duplicate:  HMR_5099   tyr_L[c]    <=>    tyr_L[m]
Keep:      r1127     HC00005[c]    ->    HC00005[r]
Duplicate:  HC00005t1r HC00005[r]    ->    HC00005[c]

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Keep:      r1128      HC00009[c]      ->      HC00009[r]
Duplicate:  HC00009t1r      HC00009[r]      ->      HC00009[c]
Keep:      r1129      HC00004[c]      ->      HC00004[r]
Duplicate:  HC00004t1r      HC00004[r]      ->      HC00004[c]
Keep:      r1131      HC00006[c]      ->      HC00006[r]
Duplicate:  HC00006t1r      HC00006[r]      ->      HC00006[c]
Keep:      r1132      HC00007[c]      ->      HC00007[r]
Duplicate:  HC00007t1r      HC00007[r]      ->      HC00007[c]
Keep:      r1133      HC00008[c]      ->      HC00008[r]
Duplicate:  HC00008t1r      HC00008[r]      ->      HC00008[c]
Keep:      r1147      akc[c] + icit[m]      <=>      akc[m] + icit[c]
Duplicate:  r2385      akc[m] + icit[c]      ->      akc[c] + icit[m]
Keep:      r1155      2obut[c]      ->      2obut[m]
Duplicate:  r1454      2obut[m]      ->      2obut[c]
Keep:      r1423      pi[c]      ->      pi[e]
Duplicate:  PIt6b      pi[e]      <=>      pi[c]
Keep:      r1427      his_L[c]      ->      his_L[m]
Duplicate:  r2416      his_L[m]      ->      his_L[c]
Keep:      r1429      glyc3p[c]      <=>      glyc3p[x]
Duplicate:  GLY3Pt      glyc3p[x]      ->      glyc3p[c]
Keep:      r1441      trdrd[c]      ->      trdrd[m]
Duplicate:  HMR_6618      trdrd[c]      <=>      trdrd[m]
Keep:      r1455      phe_L[c]      ->      phe_L[m]
Duplicate:  r1456      phe_L[m]      ->      phe_L[c]
Keep:      r1618      tyr_L[c] + phe_L[e]      <=>      phe_L[c] + tyr_L[e]
Duplicate:  TYRPHLAT2tc      phe_L[c] + tyr_L[e]      ->      tyr_L[c] + phe_L[e]
Keep:      r1619      cys_L[c] + phe_L[e]      <=>      cys_L[e] + phe_L[c]
Duplicate:  CYSPLAT2tc      cys_L[e] + phe_L[c]      ->      cys_L[c] + phe_L[e]
Keep:      r1620      leu_L[c] + phe_L[e]      <=>      leu_L[e] + phe_L[c]
Duplicate:  LEUPHAT2tc      leu_L[e] + phe_L[c]      <=>      leu_L[c] + phe_L[e]
Keep:      r1622      asn_L[c] + phe_L[e]      <=>      asn_L[e] + phe_L[c]
Duplicate:  ASNPPLAT2tc      asn_L[e] + phe_L[c]      ->      asn_L[c] + phe_L[e]
Keep:      r1623      phe_L[e] + val_L[c]      <=>      phe_L[c] + val_L[e]
Duplicate:  VALPHAT2tc      phe_L[c] + val_L[e]      ->      phe_L[e] + val_L[c]
Keep:      r1624      thr_L[c] + phe_L[e]      <=>      thr_L[e] + phe_L[c]
Duplicate:  THRPPLAT2tc      thr_L[e] + phe_L[c]      ->      thr_L[c] + phe_L[e]
Keep:      r1626      ile_L[c] + phe_L[e]      <=>      ile_L[e] + phe_L[c]
Duplicate:  ILEPLAT2tc      ile_L[e] + phe_L[c]      ->      ile_L[c] + phe_L[e]
Keep:      r1644      leu_L[e] + val_L[c]      <=>      leu_L[c] + val_L[e]
Duplicate:  VALLAT1tc      leu_L[c] + val_L[e]      ->      leu_L[e] + val_L[c]
Keep:      r1647      ile_L[c] + leu_L[e]      <=>      ile_L[e] + leu_L[c]
Duplicate:  ILELAT1tc      ile_L[e] + leu_L[c]      ->      ile_L[c] + leu_L[e]
Keep:      r1668      arg_L[e] + his_L[c]      <=>      arg_L[c] + his_L[e]
Duplicate:  HISyLATthc      arg_L[c] + his_L[e]      ->      arg_L[e] + his_L[c]
Keep:      r2009      ala_L[c] + arg_L[e]      ->      ala_L[e] + arg_L[c]
Duplicate:  ALAyLATthc      ala_L[e] + arg_L[c]      ->      ala_L[c] + arg_L[e]
Keep:      r2010      glN_L[c] + arg_L[e]      ->      glN_L[e] + arg_L[c]
Duplicate:  GLNyLATthc      glN_L[e] + arg_L[c]      ->      glN_L[c] + arg_L[e]
Keep:      r2012      arg_L[e] + met_L[c]      ->      arg_L[c] + met_L[e]
Duplicate:  METyLATthc      arg_L[c] + met_L[e]      ->      arg_L[e] + met_L[c]
Keep:      r2014      arg_L[e] + phe_L[c]      ->      arg_L[c] + phe_L[e]
Duplicate:  PHEyLATthc      arg_L[c] + phe_L[e]      ->      arg_L[e] + phe_L[c]
Keep:      r2017      arg_L[e] + leu_L[c]      ->      arg_L[c] + leu_L[e]
Duplicate:  LEUyLATthc      arg_L[c] + leu_L[e]      ->      arg_L[e] + leu_L[c]
Keep:      r2073      zn2[e]      ->      zn2[c]
Duplicate:  r2465      zn2[c]      ->      zn2[e]
Keep:      r2346      wharachd[e]      <=>      wharachd[c]
Duplicate:  WHARACHDtd      wharachd[c]      <=>      wharachd[e]
Keep:      r2355      HC02203[e]      <=>      HC02203[c]
Duplicate:  C05953td      HC02203[c]      <=>      HC02203[e]
Keep:      r2364      HC02213[e]      <=>      HC02213[c]
Duplicate:  C06439td      HC02213[c]      <=>      HC02213[e]
Keep:      r2373      akc[c] + cit[m]      <=>      akc[m] + cit[c]
Duplicate:  r2381      akc[m] + cit[c]      ->      akc[c] + cit[m]

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Keep: r2374 cit[m] + oxa[c] <=> cit[c] + oxa[m]  
 Duplicate: r2384 cit[c] + oxa[m] -> cit[m] + oxa[c]  
 Keep: r2375 icit[m] + succ[c] <=> icit[c] + succ[m]  
 Duplicate: r2386 icit[c] + succ[m] -> icit[m] + succ[c]  
 Keep: r2376 icit[m] + oxa[c] <=> icit[c] + oxa[m]  
 Duplicate: r2388 icit[c] + oxa[m] -> icit[m] + oxa[c]  
 Keep: r2377 akc[c] + HC00342[m] <=> akc[m] + HC00342[c]  
 Duplicate: r2389 akc[m] + HC00342[c] -> akc[c] + HC00342[m]  
 Keep: r2378 succ[c] + HC00342[m] <=> succ[m] + HC00342[c]  
 Duplicate: r2390 succ[m] + HC00342[c] -> succ[c] + HC00342[m]  
 Keep: r2379 mal\_L[c] + HC00342[m] <=> mal\_L[m] + HC00342[c]  
 Duplicate: r2391 mal\_L[m] + HC00342[c] -> mal\_L[c] + HC00342[m]  
 Keep: r2380 oxa[c] + HC00342[m] <=> HC00342[c] + oxa[m]  
 Duplicate: r2392 HC00342[c] + oxa[m] -> oxa[c] + HC00342[m]  
 Keep: r2395 lys\_L[m] + arg\_L[c] -> lys\_L[c] + arg\_L[m]  
 Duplicate: r2399 lys\_L[c] + arg\_L[m] -> lys\_L[m] + arg\_L[c]  
 Keep: r2396 orn[c] + lys\_L[m] -> lys\_L[c] + orn[m]  
 Duplicate: r2403 lys\_L[c] + orn[m] -> orn[c] + lys\_L[m]  
 Keep: r2397 lys\_L[m] + his\_L[c] -> lys\_L[c] + his\_L[m]  
 Duplicate: r2406 lys\_L[c] + his\_L[m] -> lys\_L[m] + his\_L[c]  
 Keep: r2398 lys\_L[m] + citr\_L[c] -> lys\_L[c] + citr\_L[m]  
 Duplicate: r2410 lys\_L[c] + citr\_L[m] -> lys\_L[m] + citr\_L[c]  
 Keep: r2400 orn[c] + arg\_L[m] -> arg\_L[c] + orn[m]  
 Duplicate: r2404 arg\_L[c] + orn[m] -> orn[c] + arg\_L[m]  
 Keep: r2401 arg\_L[m] + his\_L[c] -> arg\_L[c] + his\_L[m]  
 Duplicate: r2407 arg\_L[c] + his\_L[m] -> arg\_L[m] + his\_L[c]  
 Keep: r2402 arg\_L[m] + citr\_L[c] -> arg\_L[c] + citr\_L[m]  
 Duplicate: r2411 arg\_L[c] + citr\_L[m] -> arg\_L[m] + citr\_L[c]  
 Keep: r2405 orn[m] + his\_L[c] -> orn[c] + his\_L[m]  
 Duplicate: r2408 orn[c] + his\_L[m] -> orn[m] + his\_L[c]  
 Keep: r2409 citr\_L[c] + his\_L[m] -> citr\_L[m] + his\_L[c]  
 Duplicate: r2413 citr\_L[m] + his\_L[c] -> citr\_L[c] + his\_L[m]  
 Keep: r2471 ser\_L[e] -> ser\_L[c]  
 Duplicate: r2526 ser\_L[e] <=> ser\_L[c]  
 Keep: r2516 lac\_L[x] <=> lac\_L[c]  
 Duplicate: LAClt lac\_L[x] -> lac\_L[c]  
 Keep: RE3628M dc2coa[m] <=> dece3coa[m]  
 Duplicate: FAOXC101m dece3coa[m] -> dc2coa[m]  
 Keep: BCRNe 3bcrn[c] -> 3bcrn[e]  
 Duplicate: 3BCRNtr 3bcrn[e] <=> 3bcrn[c]  
 Keep: C101CRNe c101crn[c] -> c101crn[e]  
 Duplicate: C101CRNtr c101crn[e] <=> c101crn[c]  
 Keep: C10CRNe c10crn[c] -> c10crn[e]  
 Duplicate: C10CRNtr c10crn[e] <=> c10crn[c]  
 Keep: C10DCe c10dc[c] -> c10dc[e]  
 Duplicate: C10Dctr c10dc[e] <=> c10dc[c]  
 Keep: C12DCe c12dc[c] -> c12dc[e]  
 Duplicate: C12Dctr c12dc[e] <=> c12dc[c]  
 Keep: C1410He 3tetd7ecoacrnc[c] -> 3tetd7ecoacrnc[e]  
 Duplicate: 3TETD7ECOACRNtr 3tetd7ecoacrnc[e] <=> 3tetd7ecoacrnc[c]  
 Keep: C1420He 3ttetddcoacrnc[c] -> 3ttetddcoacrnc[e]  
 Duplicate: 3TTETDDCOACRNtr 3ttetddcoacrnc[e] <=> 3ttetddcoacrnc[c]  
 Keep: C1620He 3thexddcoacrnc[c] -> 3thexddcoacrnc[e]  
 Duplicate: 3THEXDDCOACRNtr 3thexddcoacrnc[e] <=> 3thexddcoacrnc[c]  
 Keep: C16DCe c16dc[c] -> c16dc[e]  
 Duplicate: C16Dctr c16dc[e] <=> c16dc[c]  
 Keep: C3DCe c3dc[c] -> c3dc[e]  
 Duplicate: C3Dctr c3dc[e] <=> c3dc[c]  
 Keep: C4CRNe c4crn[c] -> c4crn[e]  
 Duplicate: C4CRNtr c4crn[e] <=> c4crn[c]  
 Keep: C4DCe c4dc[c] -> c4dc[e]  
 Duplicate: C4Dctr c4dc[e] <=> c4dc[c]  
 Keep: C5DCe c5dc[c] -> c5dc[e]  
 Duplicate: C5Dctr c5dc[e] <=> c5dc[c]

Keep: C6CRNe c6crn[c] -> c6crn[e]  
 Duplicate: C6CRNtr c6crn[e] <=> c6crn[c]  
 Keep: C6DCe c6dc[c] -> c6dc[e]  
 Duplicate: C6DCTR c6dc[e] <=> c6dc[c]  
 Keep: C81CRNe c81crn[c] -> c81crn[e]  
 Duplicate: C81CRNtr c81crn[e] <=> c81crn[c]  
 Keep: C8CRNe c8crn[c] -> c8crn[e]  
 Duplicate: C8CRNtr c8crn[e] <=> c8crn[c]  
 Keep: C8DCe c8dc[c] -> c8dc[e]  
 Duplicate: C8DCTR c8dc[e] <=> c8dc[c]  
 Keep: DDCRNe 3ddcrn[c] -> 3ddcrn[e]  
 Duplicate: 3DDCRNtr 3ddcrn[e] <=> 3ddcrn[c]  
 Keep: DDECCRNe ddeccrn[c] -> ddeccrn[e]  
 Duplicate: DDECCRNtr ddeccrn[e] <=> ddeccrn[c]  
 Keep: DDECE1CRNe ddecelcrn[c] -> ddecelcrn[e]  
 Duplicate: DDECE1CRNtr ddecelcrn[e] <=> ddecelcrn[c]  
 Keep: DECCRNe 3deccrn[c] -> 3deccrn[e]  
 Duplicate: 3DECCRNtr 3deccrn[e] <=> 3deccrn[c]  
 Keep: DECDICRNe decdicrn[c] -> decdicrn[e]  
 Duplicate: DECDICRNtr decdicrn[e] <=> decdicrn[c]  
 Keep: HEDCECRNe 3hdececrn[c] -> 3hdececrn[e]  
 Duplicate: 3HDECECRNtr 3hdececrn[e] <=> 3hdececrn[c]  
 Keep: HEXDCRNe 3hexdcrn[c] -> 3hexdcrn[e]  
 Duplicate: 3HEXDCRNtr 3hexdcrn[e] <=> 3hexdcrn[c]  
 Keep: HIVCRNe 3ivcrn[c] -> 3ivcrn[e]  
 Duplicate: 3IVCRNtr 3ivcrn[e] <=> 3ivcrn[c]  
 Keep: HOCTDEC2CRNe 3octdec2crn[c] -> 3octdec2crn[e]  
 Duplicate: 3OCTDEC2CRNtr 3octdec2crn[e] <=> 3octdec2crn[c]  
 Keep: HOCTDECCRNe 3octdeccrn[c] -> 3octdeccrn[e]  
 Duplicate: 3OCTDECCRNtr 3octdeccrn[e] <=> 3octdeccrn[c]  
 Keep: HTDCRNe 3tdcrn[c] -> 3tdcrn[e]  
 Duplicate: 3TDCRNtr 3tdcrn[e] <=> 3tdcrn[c]  
 Keep: IVCrNe ivcrn[c] -> ivcrn[e]  
 Duplicate: IVCrNtr ivcrn[e] <=> ivcrn[c]  
 Keep: OCTDECE1CRNe 3octdecelcrn[c] -> 3octdecelcrn[e]  
 Duplicate: 3OCTDECE1CRNtr 3octdecelcrn[e] <=> 3octdecelcrn[c]  
 Keep: TDCRNe ttdcrn[c] -> ttdcrn[e]  
 Duplicate: TTDCRNtr ttdcrn[e] <=> ttdcrn[c]  
 Keep: TETDEC2CRNe tetdec2crn[c] -> tetdec2crn[e]  
 Duplicate: TETDEC2CRNtr tetdec2crn[e] <=> tetdec2crn[c]  
 Keep: TETDECE1CRNe tetdecelcrn[c] -> tetdecelcrn[e]  
 Duplicate: TETDECE1CRNtr tetdecelcrn[e] <=> tetdecelcrn[c]  
 Keep: TIGCRNe c51crn[c] -> c51crn[e]  
 Duplicate: C51CRNtr c51crn[e] <=> c51crn[c]  
 Keep: CARPEPT1tc carn[e] -> carn[c]  
 Duplicate: CARNtr carn[e] <=> carn[c]  
 Keep: CBLTDe adocbl[c] -> adocbl[e]  
 Duplicate: CBLtle adocbl[e] -> adocbl[c]  
 Keep: FOLTle fol[e] -> fol[c]  
 Duplicate: r0963 fol[e] -> fol[c]  
 Keep: GLYPROPEPT1tc glypro[e] -> glypro[c]  
 Duplicate: GLYPROt glypro[c] <=> glypro[e]  
 Keep: LEULEUPEPT1tc leuleu[e] -> leuleu[c]  
 Duplicate: LEULEUt leuleu[c] <=> leuleu[e]  
 Keep: PNTORDe pnto\_R[c] -> pnto\_R[e]  
 Duplicate: PNTote pnto\_R[e] <=> pnto\_R[c]  
 Keep: PROGLYPEPT1tc progly[e] -> progly[c]  
 Duplicate: PROGLyt progly[c] <=> progly[e]  
 Keep: SBTle sbt\_D[e] -> sbt\_D[c]  
 Duplicate: SBT\_Dtde sbt\_D[c] <=> sbt\_D[e]  
 Keep: TAUPAT1c taur[e] -> taur[c]  
 Duplicate: TAURCHAE taur[c] -> taur[e]  
 Keep: GLYCTDle glyc[e] <=> glyc[c]  
 Duplicate: GLYCT glyc[c] <=> glyc[e]

Keep: KHte k[e] <=> k[c]  
 Duplicate: r1492 k[c] -> k[e]  
 Keep: PHEMEe pheme[c] -> pheme[e]  
 Duplicate: PHEMEt pheme[e] -> pheme[c]  
 Keep: SPRMTDe sprm[e] <=> sprm[c]  
 Duplicate: SPRMt2r sprm[e] <=> sprm[c]  
 Keep: BALABETAtc2 cala[e] <=> cala[c]  
 Duplicate: CALAtr cala[e] <=> cala[c]  
 Keep: CRTNtr crtn[e] <=> crtn[c]  
 Duplicate: HMR\_9619 crtn[e] -> crtn[c]  
 Keep: ALAPAT4te ala\_L[e] <=> ala\_L[c]  
 Duplicate: ALAt2r ala\_L[e] <=> ala\_L[c]  
 Keep: PROPAT4te pro\_L[e] <=> pro\_L[c]  
 Duplicate: PROt2r pro\_L[e] <=> pro\_L[c]  
 Keep: 5AOpt 5aop[c] <=> 5aop[e]  
 Duplicate: 5AOpt2 5aop[e] -> 5aop[c]  
 Keep: ABT\_Dt abt\_D[e] <=> abt\_D[c]  
 Duplicate: ABT\_Dt2 abt\_D[e] <=> abt\_D[c]  
 Keep: ELAIDCRNtd elaidcrn[c] <=> elaidcrn[e]  
 Duplicate: ELAIDCRNtr elaidcrn[e] <=> elaidcrn[c]  
 Keep: HC02149td pcrn[c] <=> pcrn[e]  
 Duplicate: PCRNtr pcrn[e] <=> pcrn[c]  
 Keep: LNLCCRNdtd lnlccrn[c] <=> lnlccrn[e]  
 Duplicate: LNLCCRNtr lnlccrn[e] <=> lnlccrn[c]  
 Keep: PCSsec pcs[c] -> pcs[e]  
 Duplicate: PCSup pcs[e] -> pcs[c]  
 Keep: 3HCINNMap 3hcinnm[e] -> 3hcinnm[c]  
 Duplicate: 3HCINNMssec 3hcinnm[c] -> 3hcinnm[e]  
 Keep: 3HPPAup 3hppa[e] -> 3hppa[c]  
 Duplicate: 3HPPAsec 3hppa[c] -> 3hppa[e]  
 Keep: PACALDtm pacald[c] <=> pacald[m]  
 Duplicate: HMR\_4684 pacald[c] <=> pacald[m]  
 Keep: ACNAMt2 acnam[e] -> acnam[c]  
 Duplicate: ACNAMtr acnam[c] -> acnam[e]  
 Keep: ETHAt etha[e] <=> etha[c]  
 Duplicate: ETHAtr etha[c] -> etha[e]  
 Keep: THMtrbc thm[e] <=> thm[c]  
 Duplicate: THMt3 thm[e] <=> thm[c]  
 Keep: BUTt2r but[e] <=> but[c]  
 Duplicate: HMR\_0155 but[e] <=> but[c]  
 Keep: DIGALSGALSIDESEct digalsgalside\_hs[c] -> digalsgalside\_hs[e]  
 Duplicate: DIGALSGALSIDEt1e digalsgalside\_hs[e] -> digalsgalside\_hs[c]  
 Keep: PAIL\_hs\_SEct pail\_hs[c] -> pail\_hs[e]  
 Duplicate: PAIL\_hs\_t1e pail\_hs[e] -> pail\_hs[c]  
 Keep: PAILPALM\_HSSEct pailpalm\_hs[c] -> pailpalm\_hs[e]  
 Duplicate: PAILPALM\_HSt1e pailpalm\_hs[e] -> pailpalm\_hs[c]  
 Keep: PAILR\_HSSEct pailar\_hs[c] -> pailar\_hs[e]  
 Duplicate: PAILR\_HSt1e pailar\_hs[e] -> pailar\_hs[c]  
 Keep: PAILSTE\_HSSEct pailste\_hs[c] -> pailste\_hs[e]  
 Duplicate: PAILSTE\_HSt1e pailste\_hs[e] -> pailste\_hs[c]  
 Keep: SPHMYLN180241\_hs\_SEct sphmyln180241\_hs[c] -> sphmyln180241\_hs[e]  
 Duplicate: SPHMYLN180241\_hs\_t1 sphmyln180241\_hs[e] -> sphmyln180241\_hs[c]  
 Keep: SPHMYLN18114\_hs\_SEct sphmyln18114\_hs[c] -> sphmyln18114\_hs[e]  
 Duplicate: SPHMYLN18114\_hs\_t1 sphmyln18114\_hs[e] -> sphmyln18114\_hs[c]  
 Keep: SPHMYLN18115\_hs\_SEct sphmyln18115\_hs[c] -> sphmyln18115\_hs[e]  
 Duplicate: SPHMYLN18115\_hs\_t1 sphmyln18115\_hs[e] -> sphmyln18115\_hs[c]  
 Keep: SPHMYLN18116\_hs\_SEct sphmyln18116\_hs[c] -> sphmyln18116\_hs[e]  
 Duplicate: SPHMYLN18116\_hs\_t1 sphmyln18116\_hs[e] -> sphmyln18116\_hs[c]  
 Keep: SPHMYLN181161\_hs\_SEct sphmyln181161\_hs[c] -> sphmyln181161\_hs[e]  
 Duplicate: SPHMYLN181161\_hs\_t1 sphmyln181161\_hs[e] -> sphmyln181161\_hs[c]  
 Keep: SPHMYLN18117\_hs\_SEct sphmyln18117\_hs[c] -> sphmyln18117\_hs[e]  
 Duplicate: SPHMYLN18117\_hs\_t1 sphmyln18117\_hs[e] -> sphmyln18117\_hs[c]  
 Keep: SPHMYLN18118\_hs\_SEct sphmyln18118\_hs[c] -> sphmyln18118\_hs[e]  
 Duplicate: SPHMYLN18118\_hs\_t1 sphmyln18118\_hs[e] -> sphmyln18118\_hs[c]

Keep: SPHMYLN181181\_hs\_SECT sphmyln181181\_hs[c] -> sphmyln181181\_hs[e]  
 Duplicate: SPHMYLN181181\_hs\_t1 sphmyln181181\_hs[e] -> sphmyln181181\_hs[c]  
 Keep: SPHMYLN18120\_hs\_SECT sphmyln18120\_hs[c] -> sphmyln18120\_hs[e]  
 Duplicate: SPHMYLN18120\_hs\_t1 sphmyln18120\_hs[e] -> sphmyln18120\_hs[c]  
 Keep: SPHMYLN181201\_hs\_SECT sphmyln181201\_hs[c] -> sphmyln181201\_hs[e]  
 Duplicate: SPHMYLN181201\_hs\_t1 sphmyln181201\_hs[e] -> sphmyln181201\_hs[c]  
 Keep: SPHMYLN18121\_hs\_SECT sphmyln18121\_hs[c] -> sphmyln18121\_hs[e]  
 Duplicate: SPHMYLN18121\_hs\_t1 sphmyln18121\_hs[e] -> sphmyln18121\_hs[c]  
 Keep: SPHMYLN18122\_hs\_SECT sphmyln18122\_hs[c] -> sphmyln18122\_hs[e]  
 Duplicate: SPHMYLN18122\_hs\_t1 sphmyln18122\_hs[e] -> sphmyln18122\_hs[c]  
 Keep: SPHMYLN181221\_hs\_SECT sphmyln181221\_hs[c] -> sphmyln181221\_hs[e]  
 Duplicate: SPHMYLN181221\_hs\_t1 sphmyln181221\_hs[e] -> sphmyln181221\_hs[c]  
 Keep: SPHMYLN18123\_hs\_SECT sphmyln18123\_hs[c] -> sphmyln18123\_hs[e]  
 Duplicate: SPHMYLN18123\_hs\_t1 sphmyln18123\_hs[e] -> sphmyln18123\_hs[c]  
 Keep: SPHMYLN1824\_hs\_SECT sphmyln1824\_hs[c] -> sphmyln1824\_hs[e]  
 Duplicate: SPHMYLN1824\_hs\_t1 sphmyln1824\_hs[e] -> sphmyln1824\_hs[c]  
 Keep: SPHMYLN1825\_hs\_SECT sphmyln1825\_hs[c] -> sphmyln1825\_hs[e]  
 Duplicate: SPHMYLN1825\_hs\_t1 sphmyln1825\_hs[e] -> sphmyln1825\_hs[c]  
 Keep: 3AIBt1 3aib[e] <=> 3aib[c]  
 Duplicate: HMR\_8090 3aib[c] -> 3aib[e]  
 Keep: 2HXIC\_Lt1e 2hxic\_L[e] -> 2hxic\_L[c]  
 Duplicate: 2HXIC\_Lt2e 2hxic\_L[c] -> 2hxic\_L[e]  
 Keep: MMat2e mma[c] <=> mma[e]  
 Duplicate: MMate mma[e] <=> mma[c]  
 Keep: CE4890te2 CE4890[c] <=> CE4890[e]  
 Duplicate: CE4890te CE4890[c] <=> CE4890[e]  
 Keep: MLTHFte mlthf[e] -> mlthf[c]  
 Duplicate: MLTHFte3 mlthf[e] -> mlthf[c]  
 Keep: TYMte2 tym[c] <=> tym[e]  
 Duplicate: TYMte tym[c] <=> tym[e]  
 Keep: 1A25DHVITD3te 1a25dhvitd3[e] -> 1a25dhvitd3[c]  
 Duplicate: 1A25DHVITD3t2e 1a25dhvitd3[c] -> 1a25dhvitd3[e]  
 Keep: ORN\_Dtx orn\_D[x] <=> orn\_D[c]  
 Duplicate: HMR\_9179 orn\_D[c] <=> orn\_D[x]  
 Keep: ORN\_Dte orn\_D[c] <=> orn\_D[e]  
 Duplicate: HMR\_9180 orn\_D[c] <=> orn\_D[e]  
 Keep: HC00005te HC00005[c] -> HC00005[e]  
 Duplicate: HC00005t1e HC00005[e] -> HC00005[c]  
 Keep: HC00006te HC00006[c] -> HC00006[e]  
 Duplicate: HC00006t1e HC00006[e] -> HC00006[c]  
 Keep: HC00007te HC00007[c] -> HC00007[e]  
 Duplicate: HC00007t1e HC00007[e] -> HC00007[c]  
 Keep: HC00008te HC00008[c] -> HC00008[e]  
 Duplicate: HC00008t1e HC00008[e] -> HC00008[c]  
 Keep: HC00009te HC00009[c] -> HC00009[e]  
 Duplicate: HC00009t1e HC00009[e] -> HC00009[c]  
 Keep: NO2te no2[e] <=> no2[c]  
 Duplicate: HMR\_6991 no2[c] <=> no2[e]  
 Keep: HMR\_0025 M01268[n] -> M01268[c]  
 Duplicate: HMR\_0030 M01268[c] -> M01268[n]  
 Keep: HMR\_9581 M02035[c] <=> M02035[e]  
 Duplicate: HMR\_9582 M02035[e] -> M02035[c]  
 Keep: HMR\_9583 M02467[c] <=> M02467[e]  
 Duplicate: HMR\_9584 M02467[e] -> M02467[c]  
 Keep: HMR\_0031 0.0024 ak2gchol\_hs[c] + 0.0008 dak2gpe\_hs[c] + 0.0016 pail\_hs[c] + 0.19 dag\_hs[c]  
 Duplicate: HMR\_0032 M02392[c] -> 0.0024 ak2gchol\_hs[c] + 0.0008 dak2gpe\_hs[c] + 0.0016 pail\_hs[c]  
 Keep: ALLOP2tu allop[e] -> allop[c]  
 Duplicate: ALLOPtepvb allop[e] <=> allop[c]  
 Keep: ATVACIDMCTtu atvacid[e] <=> atvacid[c]  
 Duplicate: ATVACIDtdu atvacid[e] <=> atvacid[c]  
 Keep: OXYPthc oxyp[e] <=> oxyp[c]  
 Duplicate: OXYPtepv oxyp[c] <=> oxyp[e]  
 Keep: PVSHtu pvs[e] <=> pvs[c]  
 Duplicate: PVStep pvs[c] <=> pvs[e]

Remove any duplicate reactions from the stoichiometric matrix, but do not remove the protons.

```
if length(removedRxnInd)>0
    irrevFlag=0;
    metFlag=0;%dont remove the protons
    model = removeRxns(model,model.rxns(removedRxnInd),irrevFlag,metFlag);
end
```

Display statistics of the removed reactions

```
if printLevel>0
    [nMet0,nRxn0]=size(modelOrig.S);
    [nMet,nRxn]=size(model.S);
    fprintf('%6s\t%6s\n','#mets','#rxns')
    fprintf('%6u\t%6u\t%s\n',nMet0,nRxn0,' totals.')
    fprintf('%6u\t%6u\t%s\n',nMet0-nMet,nRxn0-nRxn,' duplicate reactions
upto protons removed.')
    fprintf('%6u\t%6u\t%s\n',nMet,nRxn,' remaining.')
end
```

#mets	#rxns	
8399	13543	totals.
0	253	duplicate reactions upto protons removed.
8399	13290	remaining.

```
%model size
[nMet,nRxn]=size(model.S);
```

## Heuristically identify exchange reactions and metabolites exclusively involved in exchange reactions

An external reaction is one that is heuristically identified by a single stoichiometric coefficient in the corresponding column of *S*, or an (abbreviated) reaction name matching a pattern (e.g. prefix EX\_) or an external subsystem assignment. Any remaining reaction is assumed to be an internal reaction. If a reaction is not external then it is denoted an internal reaction. External reactants are exclusively involved in exchange reactions, and internal reactants otherwise. The findSExRxnInd function finds the external reactions in the model which export or import mass from or to the model, e.g. Exchange reactions, Demand reactions, Sink reactions.

```
if ~isfield(model,'SIntMetBool') || ~isfield(model,'SIntRxnBool')
    model = findSExRxnInd(model,[],printLevel-1);
end
```

Assuming biomass reaction is: biomass\_reaction

## EXPECTED RESULTS

In the returned model, model.SIntRxnBool, is a boolean of reactions heuristically though to be mass balanced, while model.SIntMetBool is a boolean of metabolites heuristically though to be involved in mass balanced reactions.

## CAUTION

The aforementioned assignments of external and internal reactions and reactants is the result of a heuristic and might result in one or more errors, either due to misspecification or because the names of external reactions and external subsystems often vary between laboratories.

## Find the reactions that are flux inconsistent

Ultimately we seek to identify the set of stoichiometrically consistent reactions that are also flux consistent, with no bounds on reaction rates. However, finding the stoichiometrically consistent subset can be demanding for large models so first we identify the subset of reactions that are flux consistent and focus on them.

```
modelOrig=model;
model.lb(~model.SIntRxnBool)=-1000;
model.ub(~model.SIntRxnBool)= 1000;
if 1
    if ~isfield(model,'fluxConsistentMetBool') ||
~isfield(model,'fluxConsistentRxnBool')
        param.modeFlag=0;
        param.method='null_fastcc';
        %param.method='fastcc';
        [fluxConsistentMetBool,fluxConsistentRxnBool,...
            fluxInConsistentMetBool,fluxInConsistentRxnBool,model]...
            = findFluxConsistentSubset(model,param,printLevel);
    end
    % Remove reactions that are flux inconsistent
    if any(fluxInConsistentRxnBool)
        irrevFlag=0;
        metFlag=1;
        model =
removeRxnS(model,model.rxnS(fluxInConsistentRxnBool),irrevFlag,metFlag);
        [nMet0,nRxn0]=size(modelOrig.S);
        [nMet,nRxn]=size(model.S);

        if printLevel>0
            fprintf('%s\n','-----')
            fprintf('%6s\t%6s\n','#mets','#rxns')
            fprintf('%6u\t%6u\t%6s\n',nMet0,nRxn0,' totals.')
            fprintf('%6u\t%6u\t%6s\n',nMet0-nMet,nRxn0-nRxn,' flux
inconsistent reactions removed.')
            fprintf('%6u\t%6u\t%6s\n',nMet,nRxn,' remaining.')
            fprintf('%s\n','-----')
            if printLevel>1
                for n=1:nRxn0
                    if fluxInConsistentRxnBool(n)

fprintf('%15s\t%-100s\n',modelOrig.rxnS{n},modelOrig.rxnNames{n})
                        end
                    end
                end
            end
        end
    end
end
```



```

        end
    end
    %revise model size
    [nMet,nRxn]=size(model.S);

    %Recompute
    %Heuristically identify exchange reactions and metabolites
    exclusively involved in exchange reactions
    %finds the reactions in the model which export/import from the model
    %boundary i.e. mass unbalanced reactions
    %e.g. Exchange reactions
    %     Demand reactions
    %     Sink reactions

    model = findSExRxnInd(model,[],0);
    if printLevel>0
        fprintf('%s\n','-----end-----')
    end
end
end
end

```

```

--- findFluxConsistentSubset START ---

```

```

12164    Total reactions
5974     Reversible reactions.
6190     Irreversible reactions.
6777 flux consistent metabolites
1622 flux inconsistent metabolites
11802 flux consistent reactions
1488 flux inconsistent reactions

```

```

--- findFluxConsistentSubset END ---

```

```

-----

```

#mets	#rxns	
8399	13290	totals.
1622	1488	flux inconsistent reactions removed.
6777	11802	remaining.

```

-----

```

3HPCOAHYD	3-Hydroxyisobutyryl-Coenzyme A Hydrolase
3HPPD	3-Hydroxypropionate Dehydrogenase
3NTD7l	3'-Nucleotidase (AMP), Lysosomal
4MPTNLtr	4-Methylpentanal Transport, Endoplasmatic Reticulum
5HOXINDACTOXm	5-Hydroxyindoleacetaldehyde:NAD+ Oxidoreductase, Mitochondrial
A_MANASE	Alpha-Mannosidase
ACSOMT	S-Adenosyl-L-Methionine:N-Acetylserotonin O-Methyltransferase
ADEt1	Adenine Faciliated Transport from Lysosome
ADPGLC	ADPGlucose Diphosphatase
ADPRDPm	ADPribose Diphosphatase, Mitochondrial
ADSELK	Adenylyl-Selenate Kinase
AGLPR	Alkyl Glycerol Phosphate Reductase
AGPex	Alkyl Glycerol Phosphate Transport
AGPRim	N-Acetyl-G-Glutamyl-Phosphate Reductase, Irreversible, Mitochondrial
AGPSx	Alkylglycerone Phosphate Synthase
ALKP	Alkaline Phosphatase
ALOX12R	Arachidonate 12-Lipoxygenase R
AMACR2r	Alpha-Methylacyl Coenzyme A Racemase (Reductase)
AMACRr	Alpha-Methylacyl Coenzyme A Racemase
AMPtr	AMP Transporter, Endoplasmic Reticulum
AP4AH1	Ap4A Hydrolase, Asymmetrically
BAMPPALDOXm	Beta-Aminopropion Aldehyde:NAD+ Oxidoreductase, Mitochondrial
BDG2HCGHD	Beta-D-Glucosyl-2-Coumarinate Glucohydrolase

C2M26DCOAHLM	Cis-2-Methyl-5-Isopropylhexa-2, 5-Dienoyl Coenzyme A Hydro-Lyase, Mitochondrial
C2M26DCOAHLX	Cis-2-Methyl-5-Isopropylhexa-2, 5-Dienoyl Coenzyme A Hydro-Lyase, Peroxisomal
CBR1	Carbonyl Reductase [NADPH] 1
CCA_D3t	Calcitroic Acid Transport from Cytosol
CCA_D3tm	Calcitroic Acid Transport from Mitochondria
CO2tn	CO2 Nuclear Transport via Diffusion
CPCTDTX	Choline-Phosphate Cytidylyltransferase
CRTSTRNtr	Corticosterone Intracellular Transport
CYSLYSL	L-Cystine Lysteine-Lyase (Deaminating)
CYSTAm	Cysteine Transaminase, Mitochondrial
DALAt2rL	D-Alanine Transport via Proton Symport, Lysosomal
DEDOLP1_U	Dehydrodolichol Diphosphate Phosphatase (Uterus)
DEDOLP2_U	Dehydrodolichol Phosphate Phosphatase (Uterus)
DEDOLR_U	Dehydrodolichol Reductase (Uterus)
DHAPAx	Dihydroxyacetone Phosphate Acyltransferase
DMHPTCRNCPT1	Carnitine Fatty-Acyl Transferase
DOGULND1	2, 3-Dioxo-L-Gulonate Decarboxylase (L-Lyxonate-Forming)
DOGULND2	2, 3-Dioxo-L-Gulonate Decarboxylase (L-Xylonate-Forming)
DOGULNO2	2, 3-Dioxo-L-Gulonate:Hydrogen Peroxide Oxireductase
DPROOp	D-Proline Oxidase, Perixosomal
ECGISOr	Ecgonine Isomerase, Endoplasmatic Reticulum
EGMESTr	Ecgonine Methyl Esterase, Endoplasmatic Reticulum
ENGASE	Endo-Beta-N-Acetylglucosaminidase
ENGASE2	Endo-Beta-N-Acetylglucosaminidase
ENMAN1g	Endomannosidase (Glc1Man-Producing), Golgi Apparatus
ENMAN2g	Endomannosidase (Glc2Man-Producing), Golgi Apparatus
ENMAN3g	Endomannosidase (Glc3Man-Producing), Golgi Apparatus
ENMAN4g	Endomannosidase (M6Masnc-Producing), Golgi Apparatus
ENMAN5g	Endomannosidase (M6Masnb2-Producing), Golgi Apparatus
ENMAN6g	Endomannosidase (M5Masnbl-Producing), Golgi Apparatus
EPCTX	Ethanolamine-Phosphate Cytidylyltransferase
EX_cca_d3[e]	Exchange of Calcitroic Acid (D3)
EX_pro_D[e]	Exchange of D-Proline
EX_sel[e]	Exchange of Selenate
EX_ser_D[e]	Exchange of D-Serine
EX_vitd2[e]	Exchange of Vitamin D2
FA120ACPH	Fatty-Acyl-Acp Hydrolase
FA140ACPH	Fatty-Acyl-Acp Hydrolase
FA141ACPH	Fatty-Acyl-Acp Hydrolase
FA161ACPH	Fatty-Acyl-Acp Hydrolase
FA180ACPH	Fatty-Acyl-Acp Hydrolase
FA181ACPH	Fatty-Acyl-Acp Hydrolase
FA1821ACPH	Fatty-Acyl-Acp Hydrolase
FA1822ACPH	Fatty-Acyl-Acp Hydrolase
FA182ACPH	Fatty-Acyl-Acp Hydrolase
G1M6MASNB1terg	Transport of Glucosyl-(Alpha-D-Mannosyl)6-Beta-D-Mannosyl-Diacetylchitobiosyl-L-Asparag
G1M7MASNBterg	Transport of Glucosyl-(Alpha-D-Mannosyl)7-Beta-D-Mannosyl-Diacetylchitobiosyl-L-Asparag
G1M7MASNCTerg	Transport of Glucosyl-(Alpha-D-Mannosyl)7-Beta-D-Mannosyl-Diacetylchitobiosyl-L-Asparag
G1M8MASNterg	Transport of (Alpha-D-Glucosyl)-(Alpha-D-Mannosyl)8-Beta-D-Mannosyl-Diacetylchitobiosyl
G2M8MASNterg	Transport of (Alpha-D-Glucosyl)2-(Alpha-D-Mannosyl)8-Beta-D-Mannosyl-Diacetylchitobiosy
G3M8MASNterg	Transport of (Alpha-D-Glucosyl)3-(Alpha-D-Mannosyl)8-Beta-D-Mannosyl-Diacetylchitobiosy
GGT_U	Geranylgeranyltransferase (Uterus)
GHMT3m	Glycine Hydroxymethyltransferase, Mitochondrial
GK1m	Guanylate Kinase (GMP:ATP), Mitochondrial
GLACOm	D-Glucuronolactone:NAD+ Oxidoreductase, Mitochondrial
GPAMm_hs	Glycerol-3-Phosphate Acyltransferase
GSNKm	Guanosine Kinase, Mitochondrial
GSNtm	Guanosine Faciliated Transport in Mitochondria
H8MTER_L	H8 Mannosyltransferase, Endoplasmic Reticulum
H8MTER_U	H8 Mannosyltransferase, Endoplasmic Reticulum
HEXCCPT2	Carnitine Transferase
HEXCCRnt	Transport into the Mitochondria (Carnitine)
HMGCOARr	Hydroxymethylglutaryl Coenzyme A Reductase (Ir)
HXANTl	Hypoxanthine Faciliated Transport from Lysosome

IMACTD_m	Imidazole Acetaldehyde Dehydrogenase, Mitochondrial
INSKm	Inosine Kinase, Mitochondrial
INSTl	Transport of Inosine, Facilitated, Lysosomal
INSTm	Transport of Inosine, Facilitated, Mitochondrial
IPDPtr	Isopentenyl Diphosphate Transport, Endoplasmic Reticulum
LACZly	B-Galactosidase, Lysosomal
LCADi_Dm	Lactaldehyde Dehydrogenase, Mitochondrial
LCTStl	Lactose Transport from Cytosol to Lysosome (Via Autophagocytosis)
LCYSTATm	L-Cysteate:2-Oxoglutarate Aminotransferase, Mitochondrial
LGNCCT2	Transport into the Mitochondria (Carnitine)
LGNCRRnt	Transport into the Mitochondria (Carnitine)
LS3	Lumisterol 3 Formation
LYSMTFln	Histone-Lysine N-Methyltransferase, Nuclear
LYSMTF2n	Histone-Lysine N-Methyltransferase, Nuclear
LYSMTF3n	Histone-Lysine N-Methyltransferase, Nuclear
M4ATAer	M4A Transamidase, Endoplasmic Reticulum
M4BET2er	M4B Phosphoethanolaminy Transferase, Endoplasmic Reticulum
MAN1_6Bler	Mannosidase I, Endoplasmic Reticulum (G1M6Masnb1-Producing)
MAN1_7Ber	Mannosidase I, Endoplasmic Reticulum (G1M7Masnb-Producing)
MAN2_6Bler	Mannosidase Ii, Endoplasmic Reticulum (G1M6Masnb1-Producing)
MAN2_7Cer	Mannosidase Ii, Endoplasmic Reticulum (G1M7Masnc-Producing)
MCOATAm	Malonyl Coenzyme A-Acp Transacylase, Mitochondrial
MEOHtr	Methanol Transporter, Endoplasmic Reticulum
MI1345PKn	Inositol-1, 3, 4, 5-Triphosphate 6-Kinase, Nucleus
MI1346PKn	Inositol-1, 3, 4, 6-Tetrakisphosphate 5-Kinase, Nucleus
MI1346Ptn	1D-Myo-Inositol 1, 3, 4, 6-Tetrakisphosphate Nuclear Transport (Diffusion)
MI134PK	Inositol-1, 3, 4-Trisphosphate 6-Kinase
MI1456PKn	Inositol-1, 4, 5, 6- Tetrakisphosphate 3-Kinase, Nucleus
MI145P6Kn	Inositol-1, 4, 5-Triphosphate 6-Kinase, Nucleus
MI145PKn	Inositol-1, 4, 5-Trisphosphate 3-Kinase, Nucleus
MI3456PK	Inositol-3, 4, 5, 6-Tetrakisphosphate 1-Kinase
MMCD	Methylmalonyl Coenzyme A Decarboxylase
MMCDp	Methylmalonyl Coenzyme A Decarboxylase, Peroxisomal
NABTN0m	N4-Acetylaminobutanal:NAD+ Oxidoreductase, Mitochondrial
NDPK10m	Nucleoside-Diphosphate Kinase (ATP:DIDP), Mitochondrial
NDPK2m	Nucleoside-Diphosphate Kinase (ATP:UDP), Mitochondrial
NDPK9m	Nucleoside-Diphosphate Kinase (ATP:IDP), Mitochondrial
NMPTRCOX	N-Methylputrescine:Oxygen Oxidoreductase (Deaminating)
NNATm	Nicotinate-Nucleotide Adenylyltransferase, Mitochondrial
NRVNCCPT2	Carnitine Transferase
NRVNCCRnt	Transport into the Mitochondria (Carnitine)
NTD2m	5'-Nucleotidase (UMP), Mitochondrial
NTD3l	5'-Nucleotidase (dCMP), Lysosomal
NTD6l	5'-Nucleotidase (dAMP), Lysosomal
NTD8l	5'-Nucleotidase (dGMP), Lysosomal
NTMELYstner	Protein Trimethyl Lysine Transport (Nucleus to Endoplasmic Reticulum)
NTPP1l	Nucleoside Triphosphate Pyrophosphorylase (XTP)
PE_HStg	Phosphatidylethanolamine Scramblase
PECGONCOATr	Pseudoecgonine Coenzyme A Transferase, Endoplasmic Reticulum
PEPLYStn	Peptide (Lysine) Nuclear Transport via Diffusion
PI45PLCn	Phosphatidylinositol 4, 5-Bisphosphate Phospholipase C, Nucleus
PI4P3Ker	Phosphatidylinositol 4-Phosphate 3-Kinase, Endoplasmic Reticulum
PI5P3Ker	Phosphatidylinositol-5-Phosphate 3-Kinase, Endoplasmic Reticulum
PIK3er	Phosphatidylinositol 3-Kinase, Endoplasmic Reticulum
PLYSPSer	Protein Lysine Peptidase (Endoplasmic Reticulum)
PNTKm	Pantothenate Kinase, Mitochondrial
PROAGOX1r	L-Proline, 2-Oxoglutarate:Oxygen Oxidoreductase (4-Hydroxylating), Endoplasmic Reticulum
PRODt2r	D-Proline Reversible Transport via Proton Symport
PRODt2rL	D-Proline Reversible Transport via Proton Symport, Lysosomal
PRPNCOAHYDx	Propenoyl Coenzyme A Hydrolase, Peroxisomal
PS_HStg	Phosphatidylserine Scramblase
PTE5x	Peroxisomal Acyl Coenzyme A Thioesterase
PYAM5Ptm	Pyridoxamine 5'-Phosphate Transport via Diffusion, Mitochondrial
PYDX5Ptm	Pyridoxal 5'-Phosphate Transport via Diffusion, Mitochondrial

PYLALDOX	Perillyl Aldehyde:NAD+ Oxidoreductase
PYLALDOXm	Perillyl Aldehyde:NAD+ Oxidoreductase, Mitochondrial
RETNCOA	Retinoyl Coenzyme A Formation
Rtotaltp	Fatty Acid Intracellular Transport
SELADT	Selenate Adenylyltransferase
SELCYSLY	Selenocysteine Lyase
SELCYSLY2	Selenocysteine Lyase
SELNPS	Selenophosphate Synthase
SElt4_3	Selenate Transport via Sodium Symport
SGPL1lr	Sphingosine-1-Phosphate Lyase 1
SIAASE	Sialidase
SLDxm	L-Sulfolactate Dehydrogenase (NAD+), Mitochondrial
SOAT1lr	Sterol O-Acyltransferase (Acyl-Coenzyme A: Cholesterol Acyltransferase) 1
SOAT12r	Sterol O-Acyltransferase (Acyl-Coenzyme A: Cholesterol Acyltransferase) 1
SRTN23OX	5-Hydroxytryptamine:Oxygen 2, 3-Dioxygenase (Indole-Decyclizing)
SRTNMTX	S-Adenosyl-L-Methionine:Amine N-Methyltransferase (Srtn)
T2M26DCOAHLM	Trans-2-Methyl-5-Isopropylhexa-2, 5-Dienoyl Coenzyme A Hydro-Lyase, Mitochondrial
T2M26DCOHLx	Trans-2-Methyl-5-Isopropylhexa-2, 5-Dienoyl Coenzyme A Hydro-Lyase, Peroxisomal
T4HCINNOX	4-Coumarate:Oxygen Oxidoreductase
TDPDRR	DTDP-4-Dehydrorhamnose Reductase
TMLYster	Trimethyl-L-Lysine Transport (Er to Cytosol)
TRDRm	Thioredoxin Reductase (NADPH)
UDPGALt2g	UDPGalactose Transport, Golgi Apparatus
UDPGLCtg	UDP-Glc Golgi Transport via CMP Antiport
UGALNACter	UDP-Galnac Endoplasmic Reticulum Transport via CMP Antiport
UMPKm	UMP Kinase (Mitochondrial, ATP)
Uritm	Uridine Faciliated Transport in Mitochondria
VITD2Hm	Vitamin D-25-Hydroxylase (D2)
VITD2t	Vitamin D2 Release
VITD2tm	Vitamin D2 Transport from Mitochondria
XOL7AH2tr	Lipid, Flip-Flop Intracellular Transport
XOLDIOLONetm	Lipid, Flip-Flop Intracellular Transport
r0001	Virtual Reaction/Potential Definition
r0120	GTP 7, 8-8, 9-Dihydrolase
r0121	GTP 7, 8-8, 9-Dihydrolase
r0267	CMP-N-Acetylneuraminate, Ferrocytochrome-B5:Oxygen Oxidoreductase (N-Acetyl-Hydroxylating)
r0268	Cytidine Monophospho-N-Acetylneuraminic Acid Hydroxylase
r0400	N-Acetylneuraminate, Ferrocytochrome-B5:Oxygen Oxidoreductase (N-Acetyl-Hydroxylating)
r0598	L-Fucose Ketol-Isomerase
r0625	3Alpha, 7Alpha, 12Alpha-Trihydroxy-5Beta-Cholestan-26-Al:NAD+ 26-Oxidoreductase Bile Acid
r0626	5Beta-Cholestane-3Alpha, 7Alpha, 12Alpha, 26-Tetraol:NAD+ 26-Oxidoreductase Bile Acid
r0668	CTP:N-Acylneuraminate Cytidylyltransferase
r0678	Acyl-[Acyl-Carrier-Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating, C
r0681	(3R)-3-Hydroxybutanoyl-[Acyl-Carrier-Protein] Hydro-Lyase Fatty Acid Biosynthesis
r0682	Butyryl-[Acyl-Carrier Protein]:Malonyl Coenzyme A C-Acyltransferase (Decarboxylating, C
r0691	(3R)-3-Hydroxybutanoyl-[Acyl-Carrier Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis
r0692	(3R)-3-Hydroxydecanoyl-[Acyl-Carrier-Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis
r0693	(3R)-3-Hydroxybutanoyl-[Acyl-Carrier-Protein] Hydro-Lyase Fatty Acid Biosynthesis
r0694	(3R)-3-Hydroxyoctanoyl-[Acyl-Carrier-Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis
r0695	(3R)-3-Hydroxybutanoyl-[Acyl-Carrier-Protein] Hydro-Lyase
r0696	(3R)-3-Hydroxypalmitoyl-[Acyl-Carrier-Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis
r0697	(3R)-3-Hydroxypalmitoyl-[Acyl-Carrier-Protein] Hydro-Lyase Fatty Acid Biosynthesis
r0701	(3R)-3-Hydroxytetradecanoyl-[Acyl-Carrier-Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis
r0702	(3R)-3-Hydroxypalmitoyl-[Acyl-Carrier-Protein] Hydro-Lyase Fatty Acid Biosynthesis
r0708	2-Amino-4-Hydroxy-6- (Erythro-1, 2, 3-Trihydroxypropyl) Dihydropteridine Triphosphate 7
r0709	2-Amino-4-Hydroxy-6- (Erythro-1, 2, 3-Trihydroxypropyl) Dihydropteridine Triphosphate 7
r0712	Dodecanoyl-[Acyl-Carrier Protein]: Malonyl Coenzyme A C-Acyltransferase (Decarboxylating, C
r0713	Dodecanoyl-[Acyl-Carrier-Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating, C
r0760	Butyryl-[Acyl-Carrier Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating, C
r0761	(3R)-3-Hydroxyhexanoyl-[Acyl-Carrier-Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis
r0762	(3R)-3-Hydroxybutanoyl-[Acyl-Carrier-Protein] Hydro-Lyase Fatty Acid Biosynthesis
r0763	Hexanoyl-[Acyl-Carrier Protein]:Oxoacyl- And Enoyl-Reducing And Thioester-Hydrolysing)
r0764	Hexanoyl-[Acyl-Carrier Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating, C
r0765	Octanoyl-[Acyl-Carrier Protein]:Malonyl Coenzyme A C-Acyltransferase (Decarboxylating, C

r0766 Octanoyl-[Acyl-Carrier Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating,  
 r0767 Decanoyl-[Acyl-Carrier Protein]:Malonyl Coenzyme A C-Acyltransferase (Decarboxylating,  
 r0768 Decanoyl-[Acyl-Carrier Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating,  
 r0769 (3R)-3-Hydroxydodecanoyl-[Acyl-Carrier-Protein]:NADP+ Oxidoreductase Fatty Acid Biosynthesis  
 r0770 (3R)-3-Hydroxybutanoyl-[Acyl-Carrier-Protein] Hydro-Lyase Fatty Acid Biosynthesis  
 r0771 Tetradecanoyl-[Acyl-Carrier Protein]:Malonyl Coenzyme A C-Acyltransferase (Decarboxylating,  
 r0772 Tetradecanoyl-[Acyl-Carrier Protein]:Malonyl-[Acyl-Carrier-Protein] C-Acyltransferase (Decarboxylating,  
 r0773 Hexadecanoyl-[Acyl-Carrier Protein]:Malonyl Coenzyme A C-Acyltransferase (Decarboxylating,  
 r0775 Formamidopyrimidine Nucleoside Triphosphate 7, 8-8, 9-Dihydrolase  
 r0776 Formamidopyrimidine Nucleoside Triphosphate 7, 8-8, 9-Dihydrolase  
 r0777 GTP 7, 8-8, 9-Dihydrolase  
 r0778 GTP 7, 8-8, 9-Dihydrolase  
 r0800 Virtual ReactionPotential Definition  
 r0988 Postulated Transport Reaction  
 r0992 Na (+)Bile Acid Symporter Active Transport  
 r1021 Postulated Transport Reaction  
 r1027 Active Transport  
 r1131 Transport Reaction  
 r1132 Transport Reaction  
 r1133 Transport Reaction  
 r1319 Virtual ReactionPotential Definition  
 r1320 Virtual ReactionPotential Definition  
 r1321 Virtual ReactionPotential Definition  
 r1322 Virtual ReactionPotential Definition  
 r1323 Virtual ReactionPotential Definition  
 r1324 Virtual ReactionPotential Definition  
 r1325 Virtual ReactionPotential Definition  
 r1326 Virtual ReactionPotential Definition  
 r1327 Virtual ReactionPotential Definition  
 r1328 Virtual ReactionPotential Definition  
 r1329 Virtual ReactionPotential Definition  
 r1330 Virtual ReactionPotential Definition  
 r1331 Virtual ReactionPotential Definition  
 r1332 Virtual ReactionPotential Definition  
 r1430 [Acyl-Carrier-Protein] 4-Pantetheine-Phosphohydrolase  
 r1431 2-Deoxyuridine 5-Diphosphate:Oxidized-Thioredoxin 2-Oxidoreductase  
 r1432 2-Deoxyuridine 5-Diphosphate:Oxidized-Thioredoxin 2-Oxidoreductase  
 r1441 Active Transport  
 r1526 ATP-Binding Cassette (ABC) Tcdb:3.A.1.211.1  
 r2073 Zinc (Zn2+)-Iron (Fe2+) Permease (Zip), Tcdb:2.A.55.2.3  
 RE0066C Phosphatidylethanolamine N-Methyltransferase  
 RE0066M Phosphatidylethanolamine N-Methyltransferase  
 RE0066R Phosphatidylethanolamine N-Methyltransferase  
 RE0344M Palmitoyl Coenzyme A Hydrolase  
 RE0344X Palmitoyl Coenzyme A Hydrolase  
 RE0452N DTMP Kinase  
 RE0456M Ribonucleoside-Diphosphate Reductase  
 RE0512C 3-Hydroxyacyl Coenzyme A Dehydrogenase  
 RE0572N RE0572N  
 RE0573N RE0573N  
 RE0577M Palmitoyl Coenzyme A Hydrolase  
 RE0577X Palmitoyl Coenzyme A Hydrolase  
 RE0578M Palmitoyl Coenzyme A Hydrolase  
 RE0578X Palmitoyl Coenzyme A Hydrolase  
 RE0579M Palmitoyl Coenzyme A Hydrolase  
 RE0579X Palmitoyl Coenzyme A Hydrolase  
 RE0580L RE0580L  
 RE0580R RE0580R  
 RE0581R RE0581R  
 RE0582N RE0582N  
 RE0583N RE0583N  
 RE0688C RE0688C  
 RE0688X RE0688X  
 RE0689C RE0689C

RE0689X	RE0689X
RE0690C	RE0690C
RE0690X	RE0690X
RE0702C	Dihydrolipoyl Dehydrogenase
RE0702L	Dihydrolipoyl Dehydrogenase
RE0702M	Dihydrolipoyl Dehydrogenase
RE0702N	Dihydrolipoyl Dehydrogenase
RE0827C	RE0827C
RE0827X	RE0827X
RE0828C	RE0828C
RE0828X	RE0828X
RE0864C	RE0864C
RE0875C	RE0875C
RE0908G	Steryl-Sulfatase
RE0908R	Steryl-Sulfatase
RE0916G	Steryl-Sulfatase
RE0916R	Steryl-Sulfatase
RE0918G	Steryl-Sulfatase
RE0918R	Steryl-Sulfatase
RE0919C	Glucuronosyltransferase
RE0919R	Glucuronosyltransferase
RE0920C	Glucuronosyltransferase
RE0920R	Glucuronosyltransferase
RE0921C	Glucuronosyltransferase
RE0921R	Glucuronosyltransferase
RE0922C	Glucuronosyltransferase
RE0922R	Glucuronosyltransferase
RE0923C	Glucuronosyltransferase
RE0923R	Glucuronosyltransferase
RE0924C	Glucuronosyltransferase
RE0924R	Glucuronosyltransferase
RE0925C	Glucuronosyltransferase
RE0925R	Glucuronosyltransferase
RE0926C	Alpha-Amylase
RE0927C	Glucuronosyltransferase
RE0927R	Glucuronosyltransferase
RE0928C	Glucuronosyltransferase
RE0928R	Glucuronosyltransferase
RE0935C	Alpha-Amylase
RE0936C	RE0936C
RE0937C	RE0937C
RE0938C	RE0938C
RE0944C	Alpha-Amylase
RE1050C	Peroxidase
RE1050L	Peroxidase
RE1050N	Peroxidase
RE1062C	Neurolysin
RE1062M	Neurolysin
RE1063C	Thimet Oligopeptidase
RE1064C	Thimet Oligopeptidase
RE1096M	RE1096M
RE1096R	RE1096R
RE1099G	Steryl-Sulfatase
RE1099L	Steryl-Sulfatase
RE1099R	Steryl-Sulfatase
RE1100G	Steryl-Sulfatase
RE1100L	Steryl-Sulfatase
RE1134M	RE1134M
RE1134R	RE1134R
RE1135G	Steryl-Sulfatase
RE1135L	Steryl-Sulfatase
RE1233M	Kynurenine-Oxoglutarate Transaminase
RE1236C	RE1236C
RE1238X	Diamine N-Acetyltransferase

RE1240C	RE1240C
RE1317C	L-Iditol 2-Dehydrogenase
RE1441G	1-Phosphatidylinositol-4-Phosphate 5-Kinase
RE1473C	Gamma-Glutamyltransferase
RE1508C	RE1508C
RE1514M	Long-Chain-Fatty-Acid- Coenzyme A Ligase
RE1514X	Long-Chain-Fatty-Acid- Coenzyme A Ligase
RE1525C	3-Hydroxyacyl Coenzyme A Dehydrogenase
RE1526C	3-Hydroxyacyl Coenzyme A Dehydrogenase
RE1527C	3-Hydroxyacyl Coenzyme A Dehydrogenase
RE1537C	RE1537C
RE1537X	RE1537X
RE1538C	RE1538C
RE1538X	RE1538X
RE1539C	RE1539C
RE1539X	RE1539X
RE1582L	Quinine 3-Monooxygenase
RE1587L	Quinine 3-Monooxygenase
RE1651C	NADPH:Quinone Reductase
RE1653C	NADPH:Quinone Reductase
RE1711M	Alcohol Dehydrogenase
RE1796C	Steroid Delta-Isomerase
RE1806C	Quinine 3-Monooxygenase
RE1809C	Quinine 3-Monooxygenase
RE1809R	Quinine 3-Monooxygenase
RE1811C	Quinine 3-Monooxygenase
RE1811R	Quinine 3-Monooxygenase
RE1812C	Quinine 3-Monooxygenase
RE1812R	Quinine 3-Monooxygenase
RE1818C	Glutathione Transferase
RE1818M	Glutathione Transferase
RE1818R	Glutathione Transferase
RE1818X	Glutathione Transferase
RE1819C	Carbonyl Reductase (NADPH)
RE1819M	Carbonyl Reductase (NADPH)
RE1819X	Carbonyl Reductase (NADPH)
RE1826M	RE1826M
RE1827M	RE1827M
RE1828C	RE1828C
RE1828M	RE1828M
RE1829C	RE1829C
RE1829M	RE1829M
RE1830C	RE1830C
RE1830M	RE1830M
RE1835M	Palmitoyl Coenzyme A Hydrolase
RE1835X	Palmitoyl Coenzyme A Hydrolase
RE1836M	Propionyl Coenzyme A C2-Trimethyltridecanoyltransferase
RE1846X	Bile Acid-CoA:Amino Acid N-Acyltransferase
RE1860C	2',3'-Cyclic-Nucleotide 3'-Phosphodiesterase
RE1899C	Deoxyhypusine Synthase
RE1907C	RE1907C
RE1916X	Glutathione Transferase
RE1917C	RE1917C
RE1920C	Catechol O-Methyltransferase
RE1922C	Catechol O-Methyltransferase
RE1927C	RE1927C
RE1942C	RE1942C
RE1942R	RE1942R
RE1952C	Microsomal Epoxide Hydrolase
RE1952R	Microsomal Epoxide Hydrolase
RE1952X	Microsomal Epoxide Hydrolase
RE1954C	RE1954C
RE1956C	Microsomal Epoxide Hydrolase
RE1956R	Microsomal Epoxide Hydrolase

RE1956X	Microsomal Epoxide Hydrolase
RE1957G	1-Phosphatidylinositol-4-Phosphate 5-Kinase
RE2026C	RE2026C
RE2027C	RE2027C
RE2028C	RE2028C
RE2029C	RE2029C
RE2031M	Amino-Acid N-Acetyltransferase
RE2032M	Amino-Acid N-Acetyltransferase
RE2040C	Gamma-Glutamylcyclotransferase
RE2041C	Gamma-Glutamylcyclotransferase
RE2048N	Arachidonate 5-Lipoxygenase
RE2048R	Arachidonate 5-Lipoxygenase
RE2049C	RE2049C
RE2050C	Prostaglandin-Endoperoxide Synthase
RE2050R	Prostaglandin-Endoperoxide Synthase
RE2051C	Phosphatidate Phosphatase
RE2051G	Phosphatidate Phosphatase
RE2051R	Phosphatidate Phosphatase
RE2067C	RE2067C
RE2068C	RE2068C
RE2070C	RE2070C
RE2081C	Peptide Alpha-N-Acetyltransferase
RE2117M	Glycine N-Acyltransferase
RE2124C	Catechol O-Methyltransferase
RE2128C	RE2128C
RE2129C	RE2129C
RE2131C	RE2131C
RE2133C	Catechol O-Methyltransferase
RE2138C	RE2138C
RE2139C	RE2139C
RE2140C	RE2140C
RE2141C	RE2141C
RE2146C	Glucuronosyltransferase
RE2146R	Glucuronosyltransferase
RE2149C	Glucuronosyltransferase
RE2149R	Glucuronosyltransferase
RE2150C	Glucuronosyltransferase
RE2150R	Glucuronosyltransferase
RE2152C	RE2152C
RE2155R	Steroid 21-Monooxygenase
RE2156M	Amino-Acid N-Acetyltransferase
RE2202C	RE2202C
RE2203C	RE2203C
RE2221C	RE2221C
RE2221M	RE2221M
RE2248C	RE2248C
RE2250C	RE2250C
RE2251C	RE2251C
RE2252C	RE2252C
RE2265C	Tissue Kallikrein
RE2269C	Chymase
RE2270C	Carboxypeptidase A
RE2272C	Tripeptidyl-Peptidase I
RE2272L	Tripeptidyl-Peptidase I
RE2273C	Carboxypeptidase A
RE2292C	RE2292C
RE2296X	Glutathione Transferase
RE2306C	Pyroglutamyl-Peptidase Ii
RE2333C	RE2333C
RE2334C	RE2334C
RE2335C	RE2335C
RE2349M	Kynurenine-Oxoglutarate Transaminase
RE2360C	RE2360C
RE2360N	RE2360N



RE2373C	RE2373C
RE2375C	RE2375C
RE2377C	RE2377C
RE2384C	RE2384C
RE2404R	Glucuronosyltransferase
RE2405R	Glucuronosyltransferase
RE2410N	7-Dehydrocholesterol Reductase
RE2440C	RE2440C
RE2443C	Glutathione Transferase
RE2443M	Glutathione Transferase
RE2444C	RE2444C
RE2445C	Peptidyl-Dipeptidase A
RE2452C	RE2452C
RE2453M	Methylcrotonoyl Coenzyme A Carboxylase
RE2454M	Methylcrotonoyl Coenzyme A Carboxylase
RE2459C	Sterol Esterase
RE2474C	Quinine 3-Monooxygenase
RE2474R	Quinine 3-Monooxygenase
RE2476C	RE2476C
RE2477C	RE2477C
RE2493C	Methionine Synthase
RE2513C	Peroxidase
RE2513L	Peroxidase
RE2513N	Peroxidase
RE2514C	Peroxidase
RE2514L	Peroxidase
RE2514N	Peroxidase
RE2520C	NADPH:Quinone Reductase
RE2521C	NADPH:Quinone Reductase
RE2522C	Glutathione Transferase
RE2522X	Glutathione Transferase
RE2523C	Glutathione Transferase
RE2523X	Glutathione Transferase
RE2524C	Glutathione Transferase
RE2524X	Glutathione...

Found biomass reaction: biomass\_reaction  
 -----end-----

## Find mass leaks or siphons within the heuristically internal part, without using the bounds given by the model

```

if 1
  modelBoundsFlag=0;
  leakParams.method='dc';
  leakParams.theta=0.5;

  [leakMetBool,leakRxnBool,siphonMetBool,siphonRxnBool,leakY,siphonY,statp,stat
n] =...
    findMassLeaksAndSiphons(model,model.SIntMetBool,model.SIntRxnBool,...
    modelBoundsFlag,leakParams,printLevel);
end

```

6776	9958	subset tested for leakage (dc method, with infinite flux bounds)...
5028	6528	semipositive leaking metabolites (and exclusive reactions).
983.289	10fthf5glu[c]	
983.289	10fthf5glu[l]	
983.289	10fthf5glu[m]	
1111.32	10fthf6glu[c]	
1111.32	10fthf6glu[l]	

```

1111.32    10fthf6glu[m]
1239.36    10fthf7glu[c]
1239.36    10fthf7glu[l]
1239.36    10fthf7glu[m]
471.15     10fthf[c]
...
11DOCRSLtm
11DOCRSLtr
11DOCRSTRNtm
11DOCRSTRNtr
13DAMPPOX
1MNCAMti
1PPDCRp
24_25DHVITD3t
24_25DHVITD3tm
25HVITD3t
    5028      6528      seminegative siphon metabolites (and exclusive reactions).
983.289    10fthf5glu[c]
983.289    10fthf5glu[l]
983.289    10fthf5glu[m]
1111.32    10fthf6glu[c]
1111.32    10fthf6glu[l]
1111.32    10fthf6glu[m]
1239.36    10fthf7glu[c]
1239.36    10fthf7glu[l]
1239.36    10fthf7glu[m]
471.15     10fthf[c]
...
11DOCRSLtm
11DOCRSLtr
11DOCRSTRNtm
11DOCRSTRNtr
13DAMPPOX
1MNCAMti
1PPDCRp
24_25DHVITD3t
24_25DHVITD3tm
25HVITD3t

```

## Find the maximal set of reactions that are stoichiometrically consistent

```

if ~isfield(model,'SConsistentMetBool') ||
~isfield(model,'SConsistentRxnBool')
    if strcmp(model.modelID,'HMRdatabase2_00')
        massBalanceCheck=0;
    else
        massBalanceCheck=1;
    end
    if 1

[SConsistentMetBool,SConsistentRxnBool,SInConsistentMetBool,SInConsistentRxnB
ool,unknownSConsistencyMetBool,unknownSConsistencyRxnBool,model]...
        =findStoichConsistentSubset(model,massBalanceCheck,printLevel);
    else
        %print out problematic reactions to file
        resultsFileName=[resultsPath filesep model.modelID];
    end

```

```
[SConsistentMetBool,SConsistentRxnBool,SInConsistentMetBool,SInConsistentRxnB
ool,unknownSConsistencyMetBool,unknownSConsistencyRxnBool,model]...
```

```
=findStoichConsistentSubset(model,massBalanceCheck,printLevel,resultsFileName
);
    end
end
```

```
--- findStoichConsistentSubset START ---
```

```
-----
#mets      #rxns
```

```
6777      11802
-----
1          1844
6776      9958
3676      9238
3676      9238
3101      2564
```

```
-----
3676      9238
3676      9238
3100      720
-----
```

```
Iteration #1 minimum cardinality of conservation relaxation vector.
6776      9958      unknown consistency.
6776      9958      being tested.
6776      9475      ... of which are stoichiometrically consistent by min cardinality of stoich consistent
Infeasibility while detecting semipositive leaking metabolites.
6776      9475      Confirmed stoichiometrically consistent by leak/siphon testing.
0          483      ... of which are of unknown consistency.
0          101      removed.
```

```
-----
Iteration #2 minimum cardinality of conservation relaxation vector.
0          382      unknown consistency.
6776      9857      being tested.
6776      9601      ... of which are stoichiometrically consistent by min cardinality of stoich consistent
6776      9601      Confirmed stoichiometrically consistent by leak/siphon testing.
0          256      ... of which are of unknown consistency.
0          22      removed.
```

```
-----
Iteration #3 minimum cardinality of conservation relaxation vector.
0          234      unknown consistency.
6776      9835      being tested.
6776      9605      ... of which are stoichiometrically consistent by min cardinality of stoich consistent
Infeasibility while detecting seminegative leaking metabolites.
6776      9605      Confirmed stoichiometrically consistent by leak/siphon testing.
0          230      ... of which are of unknown consistency.
0          8        removed.
```

```
-----
Iteration #4 minimum cardinality of conservation relaxation vector.
0          222      unknown consistency.
6776      9827      being tested.
6776      9610      ... of which are stoichiometrically consistent by min cardinality of stoich consistent
Infeasibility while detecting semipositive leaking metabolites.
6776      9610      Confirmed stoichiometrically consistent by leak/siphon testing.
0          217      ... of which are of unknown consistency.
0          3        removed.
```

```
-----
Iteration #5 minimum cardinality of conservation relaxation vector.
0          214      unknown consistency.
6776      9824      being tested.
```

```

6776      9609      ... of which are stoichiometrically consistent by min cardinality of stoich consistent
Infeasibility while detecting seminegative leaking metabolites.
6776      9609      Confirmed stoichiometrically consistent by leak/siphon testing.
0         215       ... of which are of unknown consistency.
-----
Iteration #6 minimum cardinality of conservation relaxation vector.
0         215       unknown consistency.
6776      9824       being tested.
6776      9609      ... of which are stoichiometrically consistent by min cardinality of stoich consistent
Infeasibility while detecting seminegative leaking metabolites.
6776      9609      Confirmed stoichiometrically consistent by leak/siphon testing.
0         215       ... of which are of unknown consistency.
Infeasibility while detecting seminegative leaking metabolites.
--- Summary of stoichiometric consistency ---
6777      11802      totals.
1         1844      heuristically external.
6776      9958      heuristically internal:
6776      9609      ... of which are stoichiometrically consistent.
0         134       ... of which are stoichiometrically inconsistent.
0         215       ... of which are of unknown consistency.
---
0         349       heuristically internal and stoichiometrically inconsistent or unknown consistency.
644       134       ... of which are elementally imbalanced (inclusively involved metabolite).
0         134       ... of which are elementally imbalanced (exclusively involved metabolite).
6776      9609      Confirmed stoichiometrically consistent by leak/siphon testing.
--- findStoichConsistentSubset END ---

```

```

rxnBool=model.SInConsistentRxnBool & model.SIntRxnBool;
if any(rxnBool)
    if printLevel>0
        fprintf('%s\n','Stoichiometrically inconsistent heuristically non-
exchange reactions:')
    end
    for n=1:nRxn
        if rxnBool(n)
            fprintf('%20s\t%50s\t%s\n',model.rxns{n},model.rxnNames{n})
        end
    end
    if printLevel>0
        fprintf('%s\n','-----')
    end
end
end

```

```

Stoichiometrically inconsistent heuristically non-exchange reactions:
      AB07g      Abo Blood Group (Transferase A, Alpha 1-3-N-Acetylgalactosaminytransferase
-----

```

```

rxnBool=model.unknownSConsistencyRxnBool & model.SIntRxnBool;
if any(rxnBool)
    if printLevel>0
        fprintf('%s\n','Unknown consistency heuristically non-exchange
reactions:')
    end
end

```

```

for n=1:nRxn
    if rxnBool(n)
        fprintf('%20s\t%50s\t%s\n',model.rxns{n},model.rxnNames{n})
    end
end
if printLevel>0
    fprintf('%s\n','-----')
end
end

```

Unknown consistency heuristically non-exchange reactions:

3MOBt2im      3-Methyl-2-Oxobutanoate Mitochondrial Transport via Proton Symport

-----

## Sanity check of stoichiometric and flux consistency of model with open external reactions

```

if all(model.SIntMetBool & model.SConsistentMetBool)...
    && nnz(model.SIntRxnBool &
model.SConsistentRxnBool)==nnz(model.SIntRxnBool)...
    && all(model.fluxConsistentMetBool)...
    && all(model.fluxConsistentRxnBool)

[nMet,nRxn]=size(model.S);
if printLevel>1
    fprintf('%6s\t%6s\n','#mets','#rxns')
    fprintf('%6u\t%6u\t%s\n',nMet,nRxn,' totals.')

fprintf('%6u\t%6u\t%s\n',nnz(~model.SIntMetBool),nnz(~model.SIntRxnBool),'
heuristically exchange.')
end

checksPassed=0;
%Check that all heuristically non-exchange reactions are also
stoichiometrically consistent

%exchange reactions
model.EXRxnBool=strncmp('EX_', model.rxns, 3)==1;
%demand reactions going out of model
model.DMRxnBool=strncmp('DM_', model.rxns, 3)==1;
%sink reactions going into or out of model
model.SinkRxnBool=strncmp('sink_', model.rxns, 5)==1;
%all heuristic non-exchanges, i.e., supposedly all external reactions
bool=~(model.EXRxnBool | model.DMRxnBool | model.SinkRxnBool);
if nnz(bool & model.SIntRxnBool &
model.SConsistentRxnBool)==nnz(model.SConsistentRxnBool)
    checksPassed=checksPassed+1;
    if printLevel>1

fprintf('%6u\t%6u\t%s\n',nnz(model.SIntMetBool),nnz(model.SIntRxnBool),' All

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internally stoichiometrically consistent. (Check 1: minimum cardinality of
conservation relaxation vector.));
    end
end

    %Check for mass leaks or siphons in the stoichiometrically
consistent part
    %There should be no leaks or siphons in the stiochiometrically
consistent part
    modelBoundsFlag=0;
    leakParams.epsilon=1e-4;
    leakParams.eta = getCobraSolverParams('LP', 'feasTol')*100;
    leakParams.method='dc';

[leakMetBool,leakRxnBool,siphonMetBool,siphonRxnBool,leakY,siphonY,statp,stat
n]...

=findMassLeaksAndSiphons(model,model.SConsistentMetBool,model.SConsistentRxnB
ool,modelBoundsFlag,leakParams,printLevel);

    if nnz(leakMetBool)==0 && nnz(leakRxnBool)==0 &&
nnz(siphonMetBool)==0 && nnz(siphonRxnBool)==0
        checksPassed=checksPassed+1;
        if printLevel>1
            fprintf('%6u\t%6u\t%s\n',nnz(leakMetBool |
siphonMetBool),nnz(leakRxnBool | siphonRxnBool),' No internal leaks or
siphons. (Check 2: leak/siphon tests.));
        end
    end

    %Check that the maximal conservation vector is nonzero for each the
%internal stoichiometric matrix
    maxCardinalityConsParams.epsilon=1e-4;%1/epsilon is the largest mass
considered, needed for numerical stability
    maxCardinalityConsParams.method = 'quasiConcave';%seems to work the
best, but sometimes infeasible
    maxCardinalityConsParams.theta = 0.5;
    maxCardinalityConsParams.eta=getCobraSolverParams('LP',
'feasTol')*100;

[maxConservationMetBool,maxConservationRxnBool,solution]=maxCardinalityConser
vationVector(model.S(model.SConsistentMetBool,model.SConsistentRxnBool),
maxCardinalityConsParams);

    if nnz(maxConservationMetBool)==size(model.S,1) &&
nnz(maxConservationRxnBool)==nnz(model.SIntRxnBool)
        checksPassed=checksPassed+1;
        if printLevel>1

fprintf('%6u\t%6u\t%s\n',nnz(maxConservationMetBool),nnz(maxConservationRxnBo

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ol),' All internally stoichiometrically consistent. (Check 3: maximim
cardinality conservation vector.)');
    end
end

    %Check that each of the reactions in the model (with open external
reactions) is flux consistent
    modelOpen=model;
    modelOpen.lb(~model.SIntRxnBool)=-1000;
    modelOpen.ub(~model.SIntRxnBool)= 1000;
    param.epsilon=1e-4;
    param.modeFlag=0;
    param.method='null_fastcc';

[fluxConsistentMetBool,fluxConsistentRxnBool,fluxInConsistentMetBool,fluxInCo
nsistentRxnBool,modelOpen] =
findFluxConsistentSubset(modelOpen,param,printLevel-2);

    if nnz(fluxConsistentMetBool)==size(model.S,1) &&
nnz(fluxConsistentRxnBool)==size(model.S,2)
        checksPassed=checksPassed+1;
        if printLevel>1

fprintf('%6u\t%6u\t%s\n',nnz(fluxConsistentMetBool),nnz(fluxConsistentRxnBool
),' All flux consistent. (Check 4: maximim cardinality constrained right
nullspace.)');
            end
        end

        if checksPassed==4
            %save the model with open exchanges as the default generic
            %model
            model=modelOpen;
            if printLevel>0
                fprintf('%s\n','Open external reactions is
stoichiometrically and flux consistent. A flux balance model generated from
a reconstruction. GREAT!!!!');
            end
        end
        save([resultsFileName '_consistent.mat'],'model')
    end
end

```

## REFERENCES

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