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The  
Synechocystis  
model

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Iceberg

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optimal  
solution space

# Exploring Synechocystis' Metabolic Potential in the Optimum

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Centrum Wiskunde en Informatica, Life Sciences

VU University Amsterdam, IBIVU Systems Bioinformatics

BioSolarCells Cluster Meeting, 18-06-2015

# What do we need?

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- 1 genome-scale stoichiometric metabolic model of Synechocystis'
- 2 method to explore the metabolic potential in the optimum

# Genome-scale stoichiometric models in a nutshell

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- 1 **metabolism:** the set of life-sustaining chemical transformations within the cells of living organisms

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- 1 **metabolism:** the set of life-sustaining chemical transformations within the cells of living organisms
- 2 **genome-scale:** cover the total metabolic potential that is encoded in the genome of an organism



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- 3 **stoichiometric:**
  - only relative quantities:  $A + B \rightarrow C$
  - no kinetics:  ~~$k_1 \times A[t] \times B[t]$~~
  - assume steady-state:  $\frac{dA}{dt} = \frac{dB}{dt} = \frac{dC}{dt} = 0$

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- 4 predict **steady-state flux distributions**

# The *Synechocystis* model

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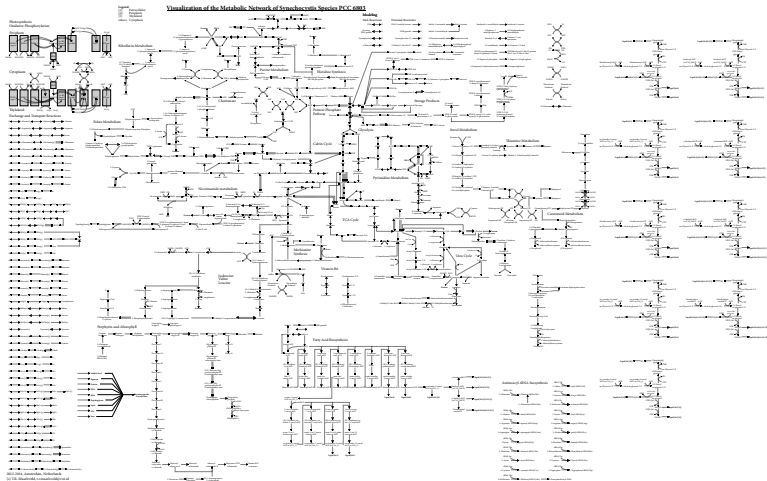
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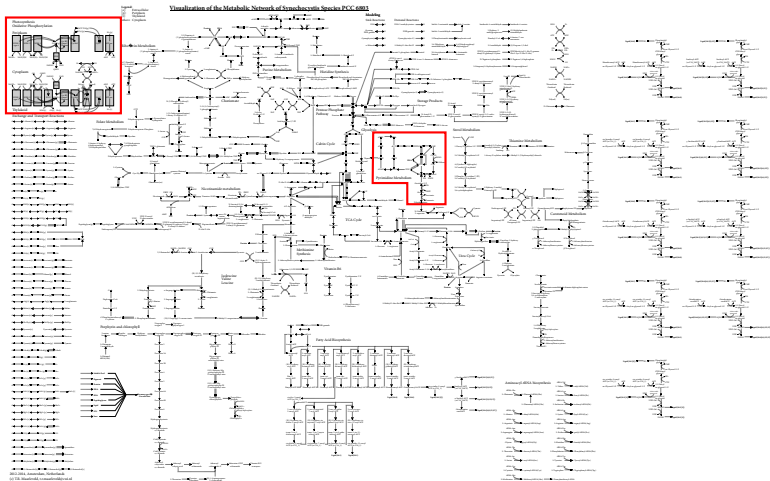
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# A real life example: cyanobacterium *Synechocystis*



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# After zooming in

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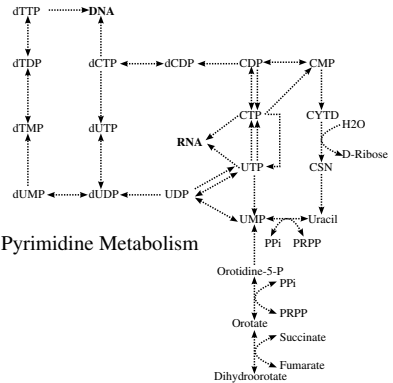
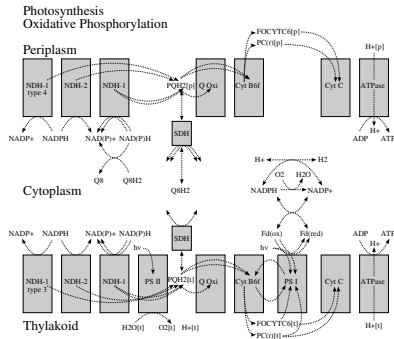
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Maarleveld et al, 2014. Plant Physiology

# How can we simulate this model?

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# Flux Balance Analysis

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**FBA**

# Flux Balance Analysis

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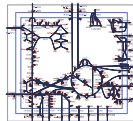
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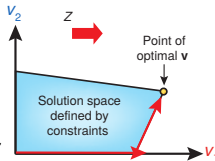
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Genome-scale metabolic model



**FBA**

Calculate fluxes that maximize  $Z$





# Exploring different scenario's

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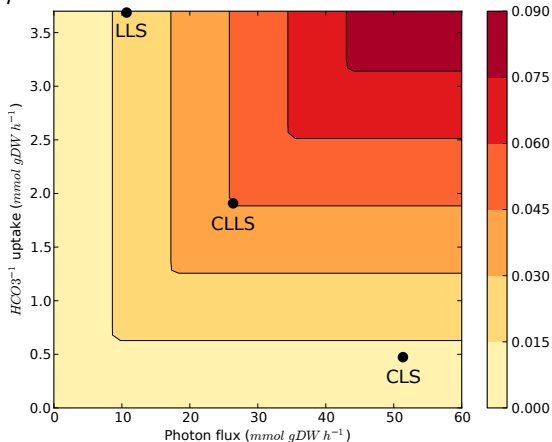
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$$Z = \max \mu$$



Visualization of flux data is now possible with

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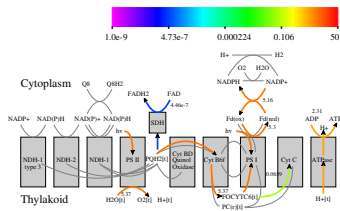
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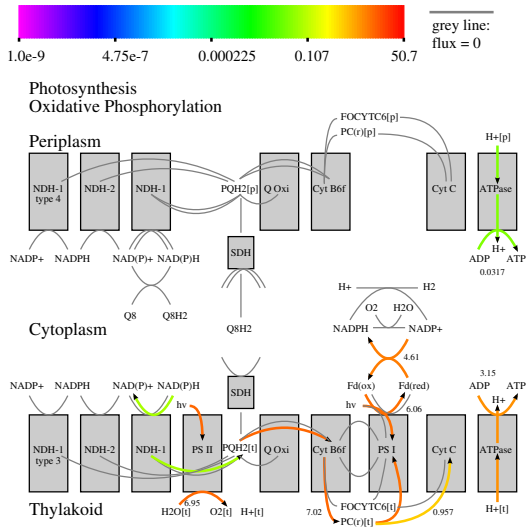
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# VoNDA

## *Visualization of Network Data*



# Carbon and Light Limiting State



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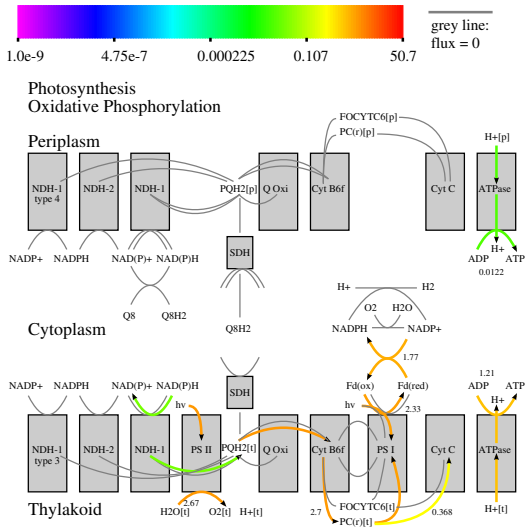
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# Light limiting state



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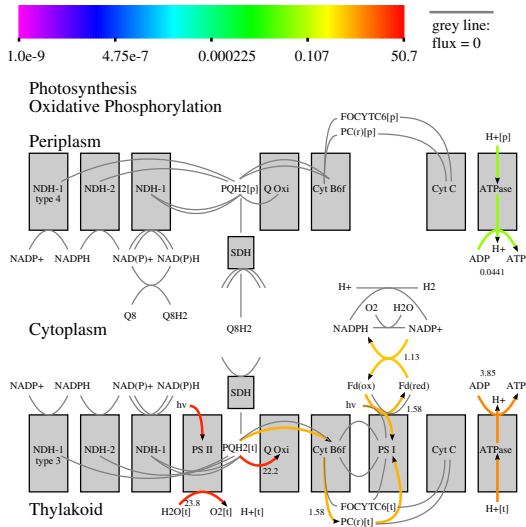
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# Carbon Limiting State



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# Net effect

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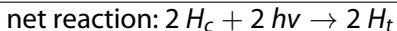
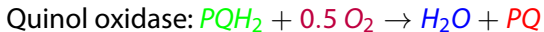
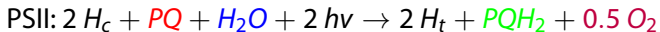
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- $J_{PSI}/J_{PSII} = 0.058$  (LEF gives 1)
- $J_{ATP}/J_{NADPH} = 7.2$  (LEF gives 1.28)



- net effect: ATP production!

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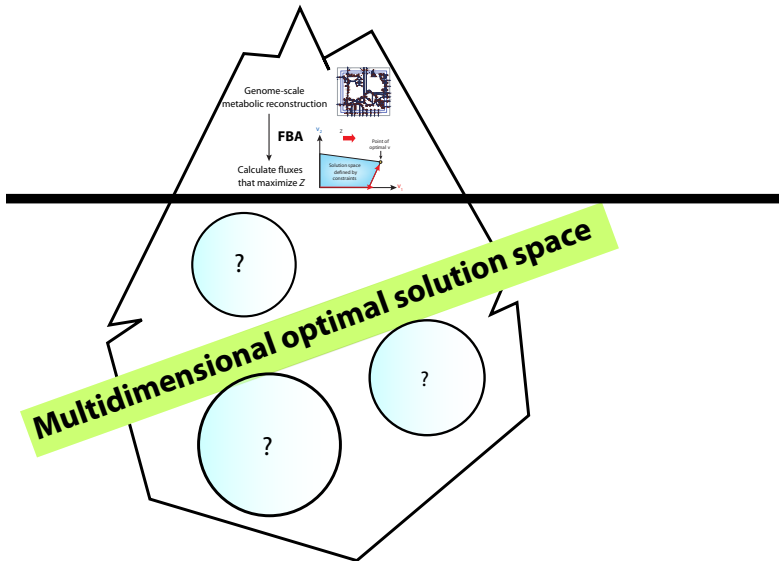
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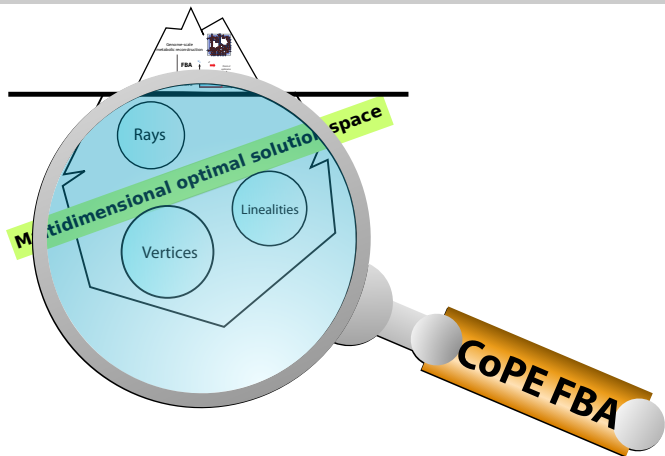
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Topological understanding of the metabolic capacity in terms of  
metabolic flux routes

Kelk et al, 2012. Scientific Reports.



# CoPE-FBA is slow!

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# Can we speed-up CoPE-FBA?

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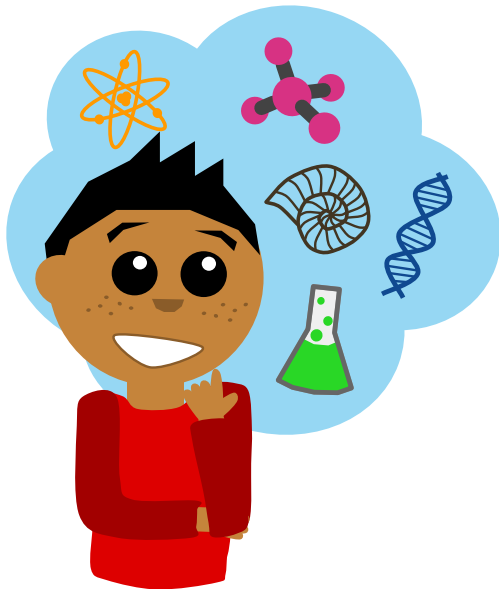
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# How fast is TimoTimo?

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Organism (model)	Source	Running time (s)	
		CoPE-FBA Kelk et al. 2012	TimoTimo
<i>S. PC6803</i> (iTm686)	glycogen	FAILED	117
<i>S. PC6803</i> (iTm686)	light	FAILED	26
<i>E. coli</i> (iAF1260,ox)	glucose	FAILED	36
<i>E. coli</i> (iAF1260,noox)	glucose	FAILED	27
<i>L. Lactis</i>	glucose	432495	16
<i>S. thermophilus</i>	lactose	345362	15
<i>M. tuberculosis</i> (iNJ661)	glycerol	FAILED	41
<i>M. barkeri</i> (iAF692)	methanol	265760	12
<i>E. coli</i> (iJR904)	malate	324793	10
<i>E. coli</i> (iJR904)	fumarate	584690	16

"Faster than lightning", but why?

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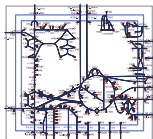
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# CoPE-FBA approach (in a nutshell)

**iAF1260 (glc, aerobic)**

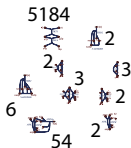


Vertices

**CoPE-FBA (weeks)**

120.932.352

Subnetworks



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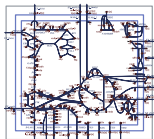
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# TimoTimo uses a divide-and-conquer approach

iAF1260 (glc, aerobic)



**CoPE-FBA (weeks)**

Vertices

120.932.352

Subnetworks

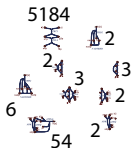


Subnetworks

**TimoTimo (minutes)**

Vertices

$6+3+5184+3+2+54+2+2 = 5256$



Enumerate 0.004% of the vertices to get all

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# Optimal solution space characterization after reversible-reaction splitting

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	<i>Synechocystis</i> iTM686	
	autotrophic (LLS)	heterotrophic (glycogen)
Simulation time (s)	135	228
Rays	242	242
Linealities	0	0
Vertices	<b>41.803.776</b>	<b>1.399.652.352</b>
Subnetworks	11	11

# Optimal solution space characterization after reversible-reaction splitting

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Why do we have so many ??

# Optimal solution space characterization after reversible-reaction splitting

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Why do we have so many ??

We do not have a enough (flux) constraints

# What's the effect of secondary objectives?

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$$P_L = |\{J_i : J_i \neq 0\}| \quad (1)$$

$$P_J = \sum_{i=1}^r |J_i| \quad (2)$$

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$\min(P_L)$	184.320	460.800
$\min(P_J)$	24	4

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**What kind of distributions do you expect?**

# Gaussian and bimodal distributions of secondary optimization of vertices

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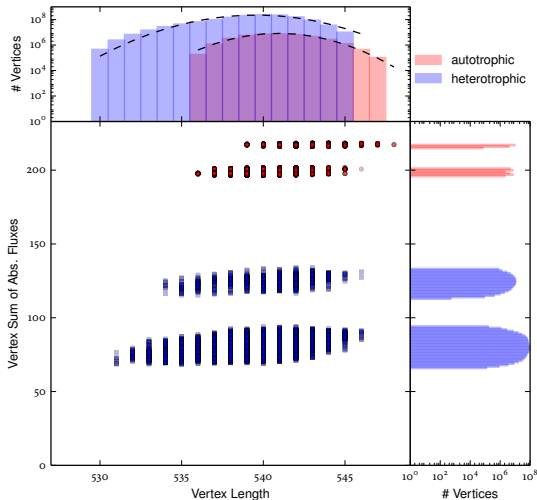
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# What explains the bimodal distributions?

## autotrophic growth

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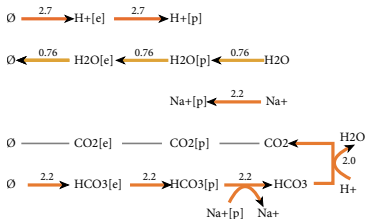
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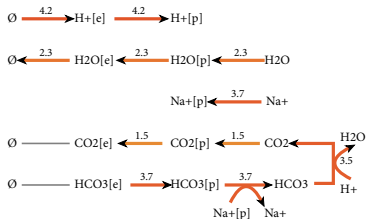
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**min  $P_j$**



**max  $P_j$**



- strategy: minimal  $\text{HCO}_3^-$  vs. maximal  $\text{HCO}_3^-$  import
- relative effect: 206 %
- total effect: 11 %



# What explains the bimodal distributions?

## autotrophic growth

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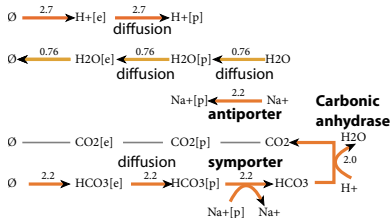
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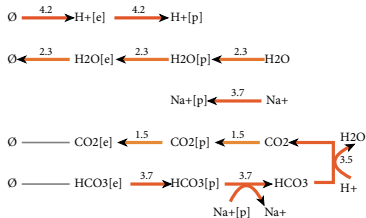
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**min  $P_j$**



**max  $P_j$**



- strategy: minimal  $\text{HCO}_3^-$  vs. maximal  $\text{HCO}_3^-$  import
- some reactions are related to diffusion
- requires additional expression of transporters and enzymes

# Summary

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we present

- a ready-to-use genome-scale stoichiometric model of the cyanobacterium *Synechocystis* with interactive map
- we present a **faster** and more **user-friendly** tool to analyze the metabolic potential in the optimum

we can use this to e.g.

- explore the metabolic potential for producing biofuels
- obtain a topological understanding of the metabolic capacity
- investigate flexibility in the optimum

# Acknowledgments



**BioSolar Cells**



- Meike Wortel , Brett Olivier, Bas Teusink, Frank Bruggeman (VU)
- Arne Reimers (CWI)
  
- Maarleveld et al. PLoS Comp. Biol. 2015 - Interplay between Constraints, Objectives, and Optimality for Genome-Scale Stoichiometric Models
- Maarleveld et al. Plant Physiology 2014 - A Data Integration and Visualization Resource for the Metabolic Network of *Synechocystis* sp. PCC 6803
  
- Contact information: [t.r.maarleveld\[at\]cwi.nl](mailto:t.r.maarleveld@cwi.nl)
- Website: [memesa-tools.sf.net](http://memesa-tools.sf.net)

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