Modelling circadian rhythms using stochastic process algebra

Jane Hillston.
LFCS and CSBE, University of Edinburgh

22nd September 2010

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Outline

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

- Ostreococcus Tauri
- Stochastic modelling and model checking
- Investigating mutational effects

Outline

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

- Ostreococcus Tauri
- Stochastic modelling and model checking
- Investigating mutational effects

Current Hypothesis

Some of the techniques we have developed over the last thirty years for modelling complex software systems can be beneficially applied to the modelling aspects of systems biology.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Current Hypothesis

Some of the techniques we have developed over the last thirty years for modelling complex software systems can be beneficially applied to the modelling aspects of systems biology.

In particular formalisms which encompass support for

- Abstraction
- Modularity and
- Reasoning

have a key role to play.

Current Hypothesis

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Some of the techniques we have developed over the last thirty years for modelling complex software systems can be beneficially applied to the modelling aspects of systems biology.

In particular formalisms which encompass support for

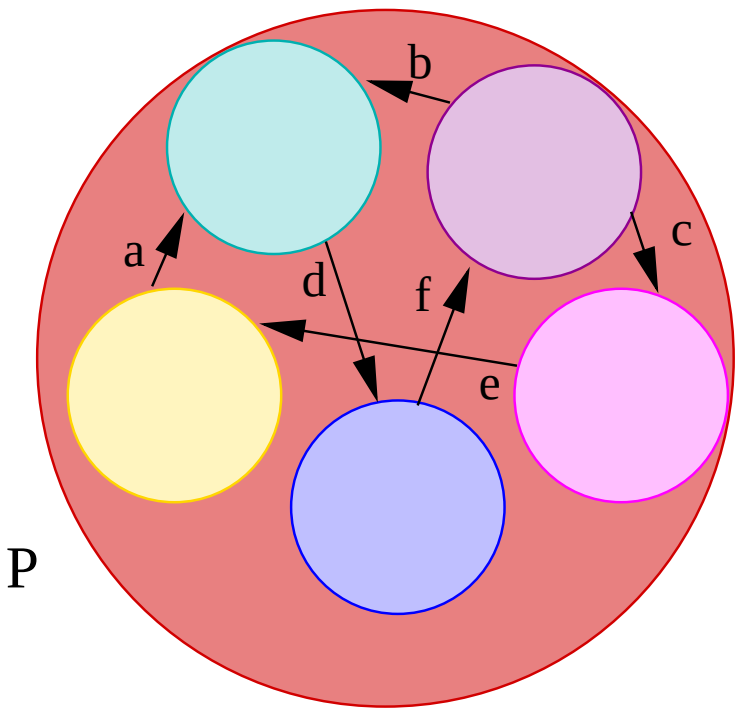
- Abstraction
- Modularity and
- Reasoning

have a key role to play.

Process algebras have mechanisms for each of these, and stochastic extensions which allow dynamic properties to be analysed.

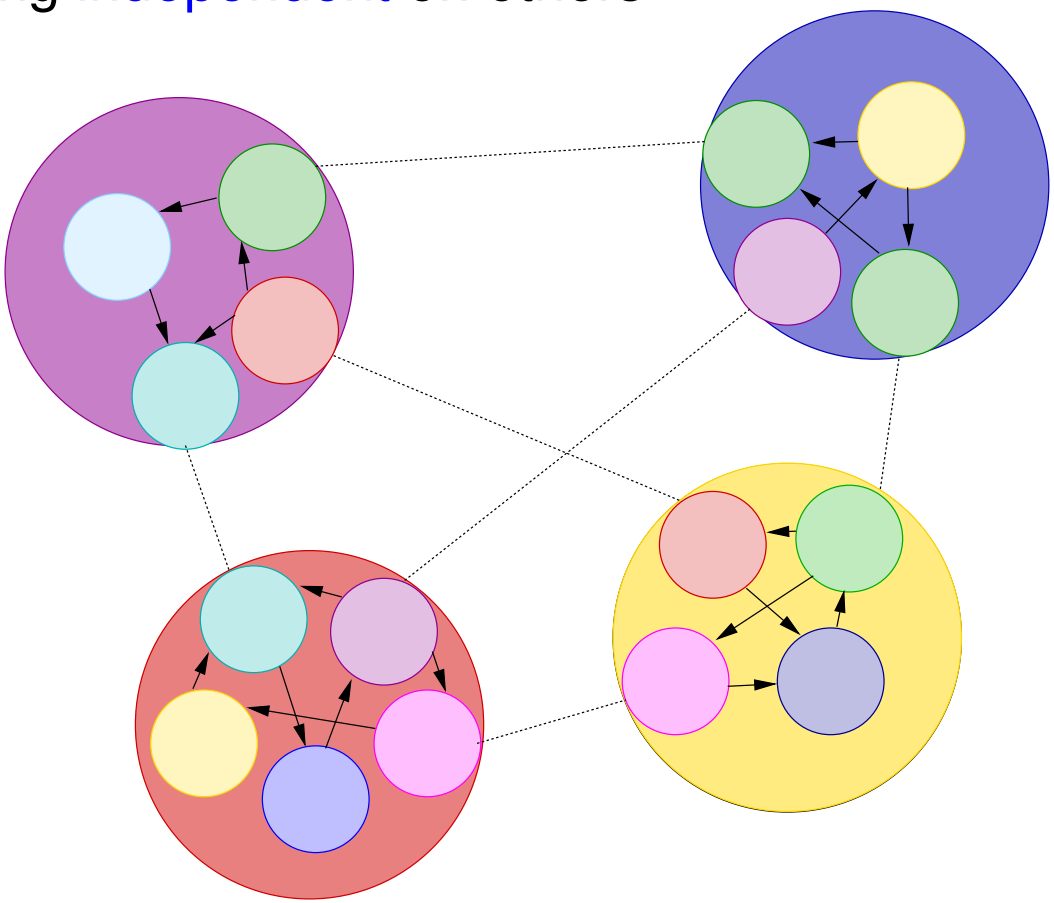
Process Algebra

- Models consist of agents which engage in actions and by doing so change state.



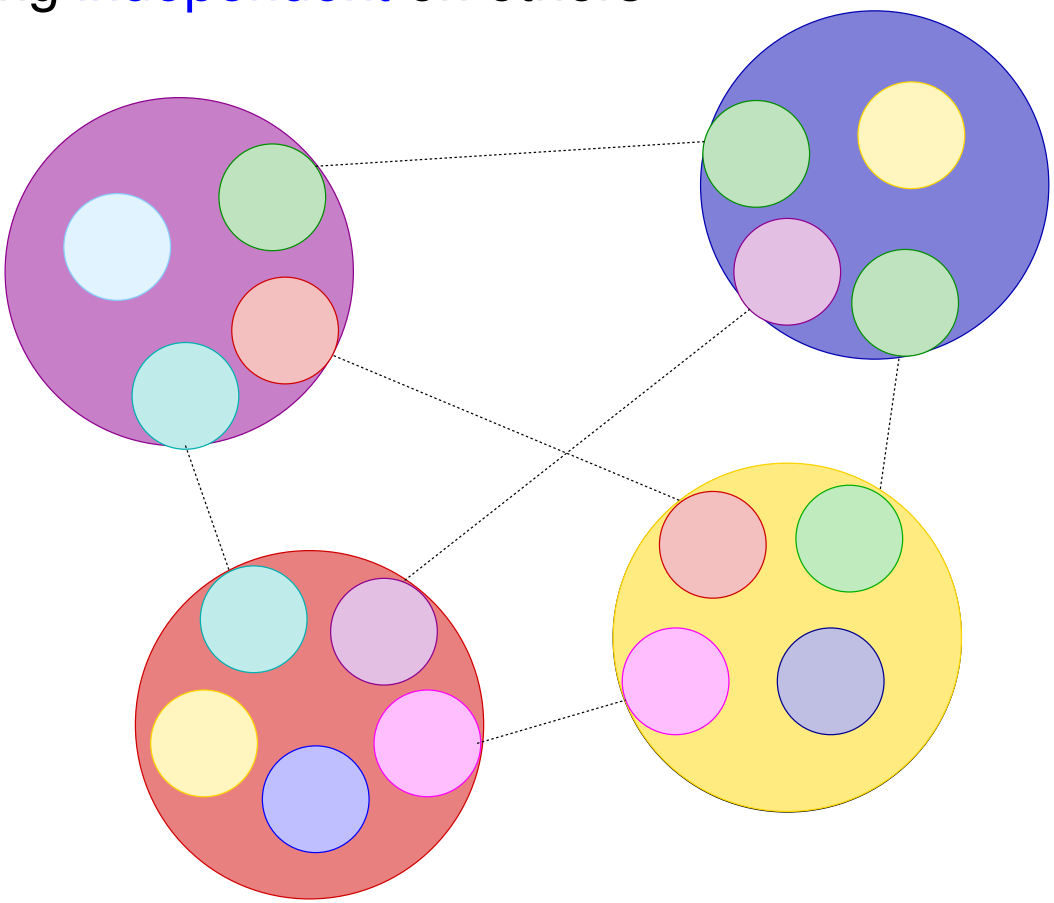
Process Algebra

- Models consist of agents which engage in actions and by doing so change state.
- Agents then interact, sharing some actions and being independent on others.



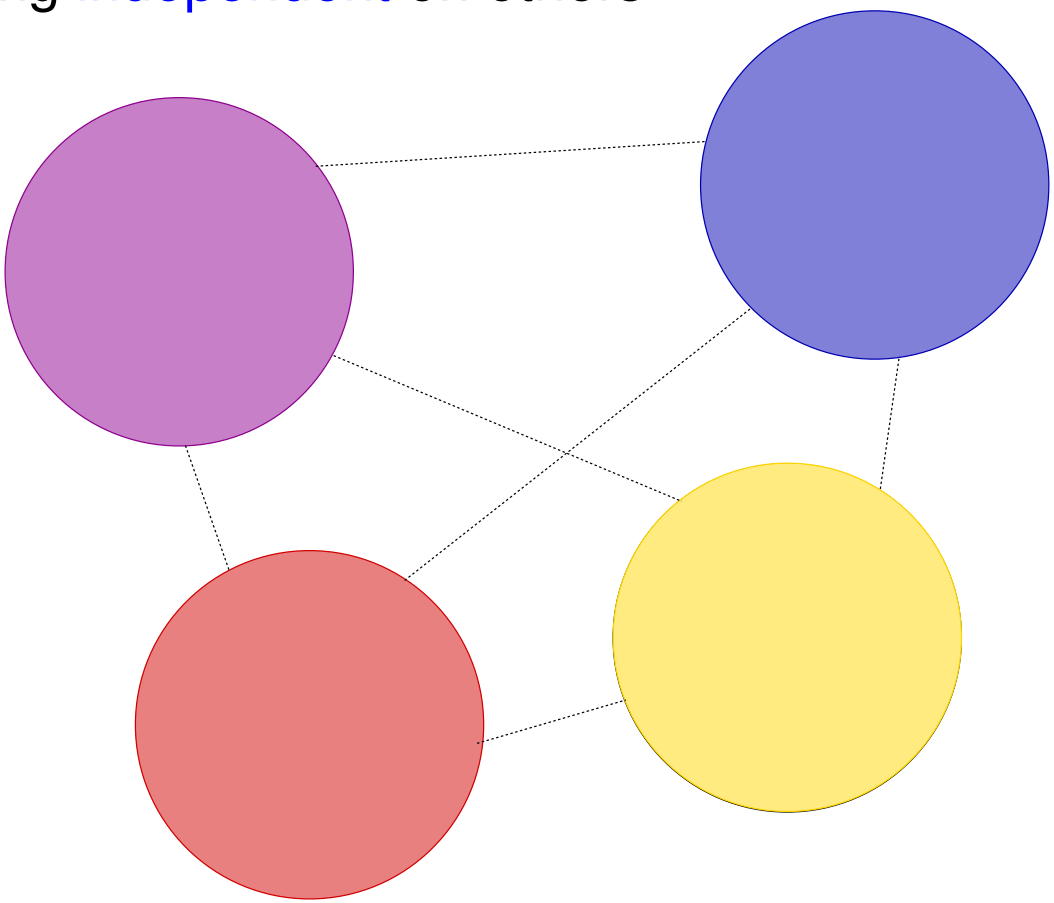
Process Algebra

- Models consist of agents which engage in actions and by doing so change state.
- Agents then interact, sharing some actions and being independent on others.



Process Algebra

- Models consist of agents which engage in actions and by doing so change state.
- Agents then interact, sharing some actions and being independent on others.



Process Algebra

- Models consist of agents which engage in actions and by doing so change state.
- Agents then interact, sharing some actions and being independent on others.

- Stochastic process algebra model in the same style but assume that each action has a delay governed by a random variable.

Deriving quantitative data

SPA models can be analysed for quantified dynamic behaviour in a number of different ways.

- The language may be used to generate a Markov Process (CTMC) to be solved numerically.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Deriving quantitative data

SPA models can be analysed for quantified dynamic behaviour in a number of different ways.

- The language may be used to generate a Markov Process (CTMC) to be solved numerically.
- The language also may be used to generate a stochastic simulation.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Deriving quantitative data

SPA models can be analysed for quantified dynamic behaviour in a number of different ways.

- The language may be used to generate a Markov Process (CTMC) to be solved numerically.
- The language also may be used to generate a stochastic simulation.
- The language may be used to generate a system of ordinary differential equations (ODEs).

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Deriving quantitative data

SPA models can be analysed for quantified dynamic behaviour in a number of different ways.

- The language may be used to generate a Markov Process (CTMC) to be solved numerically.
- The language also may be used to generate a stochastic simulation.
- The language may be used to generate a system of ordinary differential equations (ODEs).

Each of these has tool support so that the underlying model is derived automatically according to the predefined rules.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Molecular processes as concurrent computations

Concurrency	**Molecular Biology**
Concurrent computational processes	Molecules
Synchronous communication	Molecular interaction
Transition or mobility	Biochemical modification or relocation

[Regev *et al* 2000]

Molecular processes as concurrent computations

Concurrency	**Molecular Biology**
Concurrent computational processes	Molecules
Synchronous communication	Molecular interaction
Transition or mobility	Biochemical modification or relocation

[Regev *et al* 2000]

Molecular processes as concurrent computations

Concurrency	**Molecular Biology**
Concurrent computational processes	Molecules
Synchronous communication	Molecular interaction
Transition or mobility	Biochemical modification or relocation

[Regev *et al* 2000]

Molecular processes as concurrent computations

Concurrency	**Molecular Biology**
Concurrent computational processes	Molecules
Synchronous communication	Molecular interaction
Transition or mobility	Biochemical modification or relocation

[Regev *et al* 2000]

Formal Systems

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

There are two alternative approaches to constructing dynamic models of biochemical pathways commonly used by biologists:

Formal Systems

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

There are two alternative approaches to constructing dynamic models of biochemical pathways commonly used by biologists:

- Ordinary Differential Equations:
  - continuous time,
  - continuous behaviour (concentrations),
  - deterministic.

Formal Systems

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

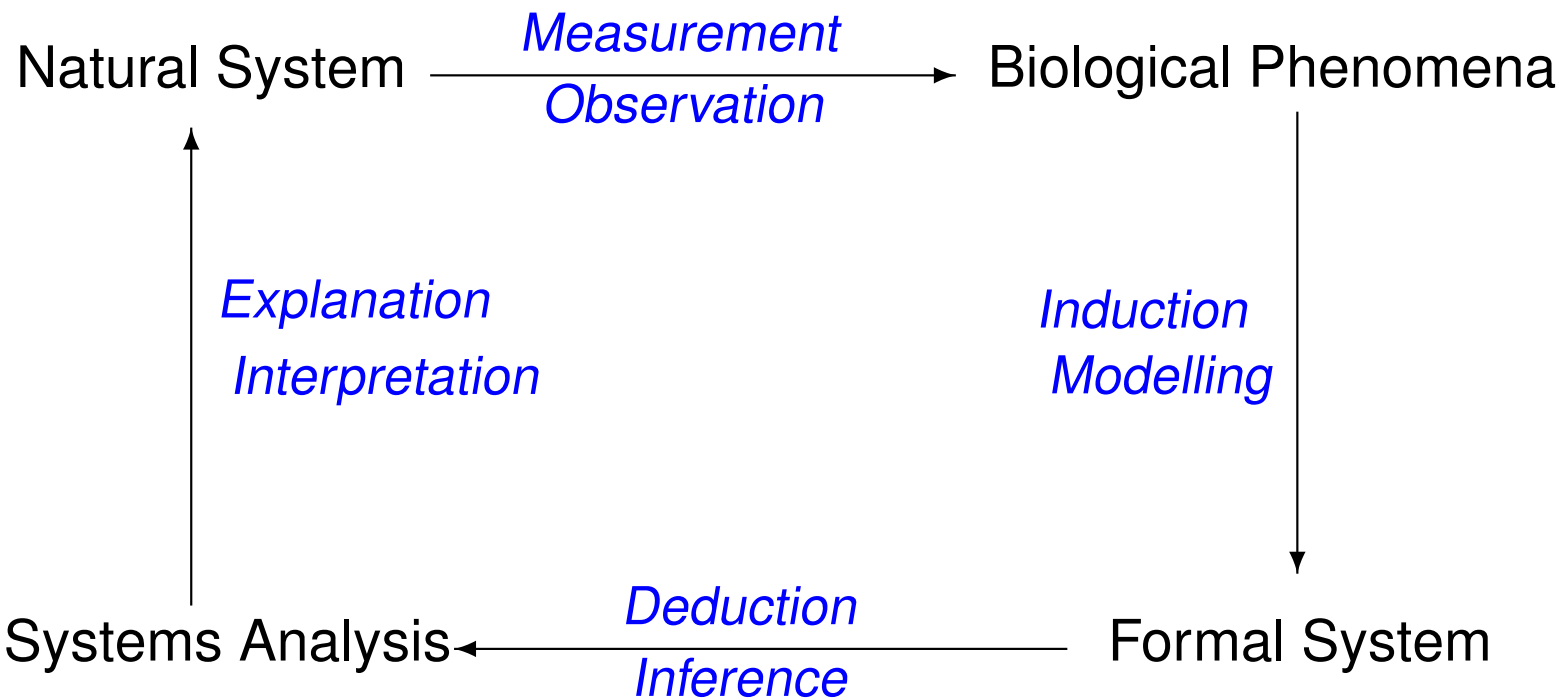
Stochastic modelling and model checking

Investigating mutational effects

There are two alternative approaches to constructing dynamic models of biochemical pathways commonly used by biologists:

- Ordinary Differential Equations:
  - continuous time,
  - continuous behaviour (concentrations),
  - deterministic.
- Stochastic Simulation:
  - continuous time,
  - discrete behaviour (no. of molecules),
  - stochastic.

Systems Biology Methodology



Systems Biology Modelling

- In most current work mathematics is being used directly as the formal system.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Systems Biology Modelling

- In most current work mathematics is being used directly as the formal system.
- Previous experience in computer performance modelling has shown us that there can be benefits to interposing a formal process algebra model between the system and the underlying mathematical model.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Systems Biology Modelling

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

- In most current work mathematics is being used directly as the formal system.
- Previous experience in computer performance modelling has shown us that there can be benefits to interposing a formal process algebra model between the system and the underlying mathematical model.
- Moreover taking this “high-level programming” style approach offers the possibility of different “compilations” to different mathematical models: integrated modelling and analysis.

Outline

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms
- Ostreococcus Tauri
- Stochastic modelling and model checking
- Investigating mutational effects

Bio-PEPA

In Bio-PEPA:

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Bio-PEPA

In Bio-PEPA:

- Unique rates are associated with each reaction (action) type, separately from the specification of the logical behaviour. These rates are typically specified by functions.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Bio-PEPA

In Bio-PEPA:

- Unique rates are associated with each reaction (action) type, separately from the specification of the logical behaviour. These rates are typically specified by functions.
- The representation of an action within a component (species) records the stoichiometry of that entity with respect to that reaction. The role of the entity is also distinguished.

Bio-PEPA

In Bio-PEPA:

- Unique rates are associated with each reaction (action) type, separately from the specification of the logical behaviour. These rates are typically specified by functions.
- The representation of an action within a component (species) records the stoichiometry of that entity with respect to that reaction. The role of the entity is also distinguished.
- The local states of components are quantitative rather than functional, i.e. distinct states of the species are represented as distinct components, not derivatives of a single component.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

The Bio-PEPA Language

Sequential component (species component)

$S ::= (\alpha, \kappa)\ \text{op}\ S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$

The Bio-PEPA Language

Sequential component (species component)

$S ::= (\alpha, \kappa)\ \text{op}\ S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$

The Bio-PEPA Language

Sequential component (species component)

$S ::= (\alpha, \kappa)\ \mathrm{op}\ S \mid S + S \mid C \qquad$ where $\mathrm{op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$

The Bio-PEPA Language

Sequential component (species component)

$S ::= (\alpha, \kappa)\ \text{op}\ S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$

The Bio-PEPA Language

Sequential component (species component)

$S ::= (\alpha, \kappa)\ \text{op}\ S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$

The Bio-PEPA Language

Sequential component (species component)

$$S ::= (\alpha, \kappa) \text{ op } S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$$

Model component

$$P ::= P \underset{\mathcal{L}}{\bowtie} P \mid S(l)$$

The Bio-PEPA Language

Sequential component (species component)

$$S ::= (\alpha, \kappa) \text{ op } S \mid S+S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$$

Model component

$$P ::= P \underset{\mathcal{L}}{\bowtie} P \mid S(l)$$

The Bio-PEPA Language

Sequential component (species component)

$$S ::= (\alpha, \kappa) \text{ op } S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$$

Model component

$$P ::= P \underset{\mathcal{L}}{\bowtie} P \mid S(l)$$

The Bio-PEPA Language

Sequential component (species component)

$$S ::= (\alpha, \kappa) \text{ op } S \mid S+S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$$

Model component

$$P ::= P \underset{\mathcal{L}}{\bowtie} P \mid S(l)$$

The parameter l is abstract, recording quantitative information about the species.

The Bio-PEPA Language

Sequential component (species component)

$$S ::= (\alpha, \kappa) \text{ op } S \mid S + S \mid C \qquad \text{where op} = \downarrow \mid \uparrow \mid \oplus \mid \ominus \mid \odot$$

Model component

$$P ::= P \underset{\mathcal{L}}{\bowtie} P \mid S(l)$$

The parameter l is abstract, recording quantitative information about the species.

The system description records the impact of action on this quantity which may be

- number of molecules (SSA),
- concentration (ODE) or
- a level within a semi-quantitative model (CTMC).

The Bio-PEPA system

A Bio-PEPA system $\mathcal{P}$ is a 6-tuple $\langle \mathcal{V}, \mathcal{N}, \mathcal{K}, \mathcal{F}_R, \mathit{Comp}, P \rangle$, where:

- $\mathcal{V}$ is the set of compartments;
- $\mathcal{N}$ is the set of quantities describing each species (step size, number of levels, location, ...);
- $\mathcal{K}$ is the set of parameter definitions;
- $\mathcal{F}_R$ is the set of functional rate definitions;
- *Comp* is the set of definitions of sequential components;
- P is the model component describing the system.

Outline

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms
- Ostreococcus Tauri
- Stochastic modelling and model checking
- Investigating mutational effects

Circadian Rhythms in *Ostreococcus Tauri*

Jane Hillston.
University of Edinburgh.

Process Algebras for Systems Biology

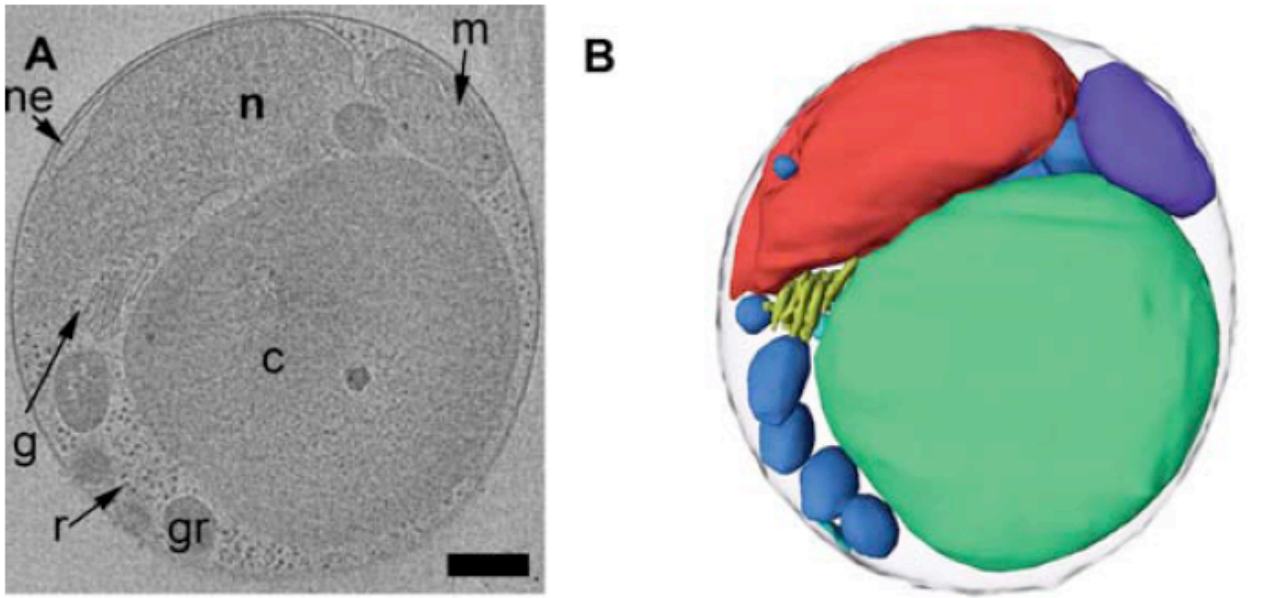
Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

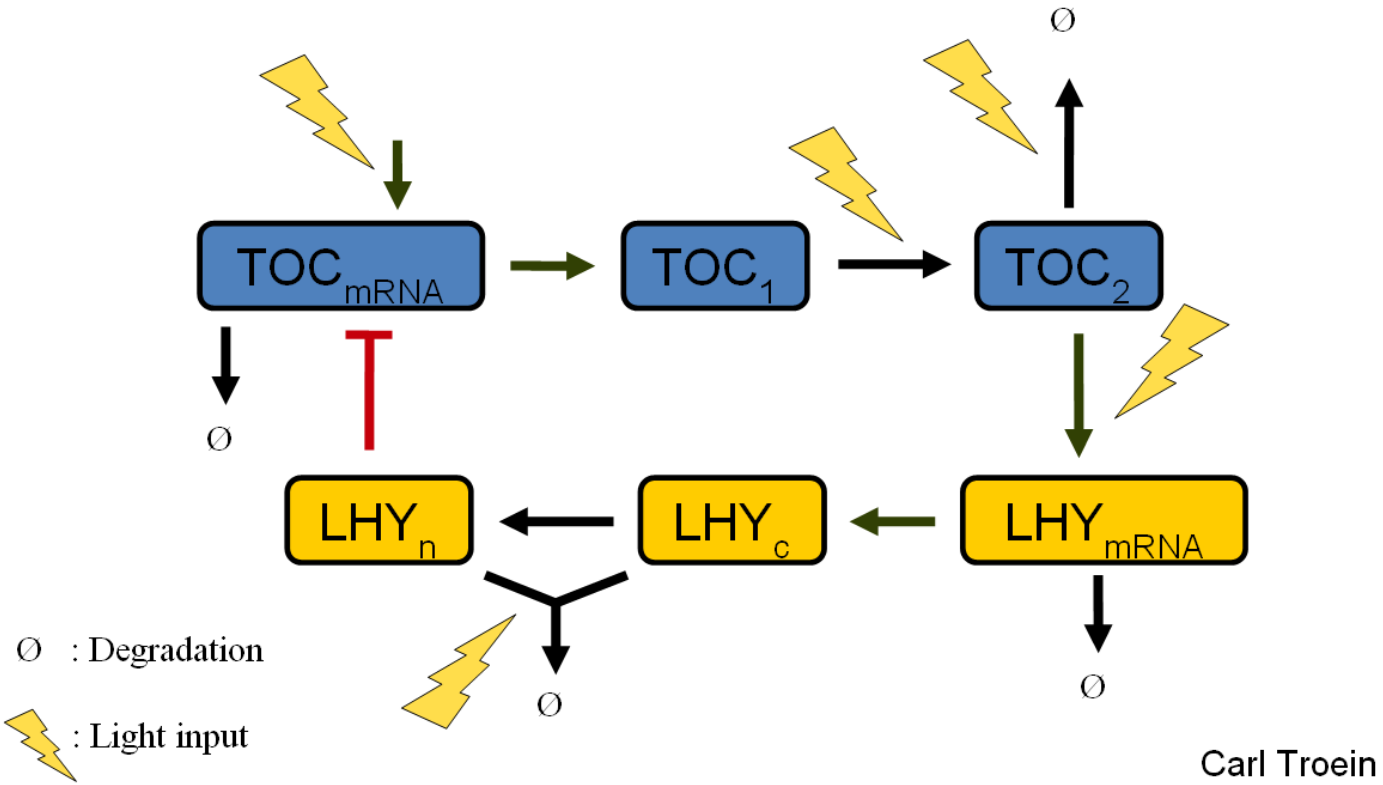
Investigating mutational effects



The green alga *Ostreococcus Tauri* is an ideal model system for understanding the function of plant clocks.

We used a Bio-PEPA model to study the variability and robustness of the clock's behaviour with respect to internal stochastic noise, environmental changes and mutational changes.

The biological model



- Feedback loop between the two main genes (TOC1 and LHY).
- Effect of light on several parts of the network.

Modelling overview

- A previous deterministic (ODE) model of the system had been developed by hand.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Modelling overview

- A previous deterministic (ODE) model of the system had been developed by hand.
- We developed a Bio-PEPA model and validated that its continuous-deterministic interpretation coincided with the original model.

Modelling overview

- A previous deterministic (ODE) model of the system had been developed by hand.
- We developed a Bio-PEPA model and validated that its continuous-deterministic interpretation coincided with the original model.
- Stochastic simulation was used with the discrete-stochastic interpretation of the Bio-PEPA model to investigate stochastic fluctuations, focusing on clock phase and sensitivity analysis.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Modelling overview

- A previous deterministic (ODE) model of the system had been developed by hand.
- We developed a Bio-PEPA model and validated that its continuous-deterministic interpretation coincided with the original model.
- Stochastic simulation was used with the discrete-stochastic interpretation of the Bio-PEPA model to investigate stochastic fluctuations, focusing on clock phase and sensitivity analysis.
- Model-checking was also used to give more detailed analysis of variability within the system, via specific questions about model behaviour.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

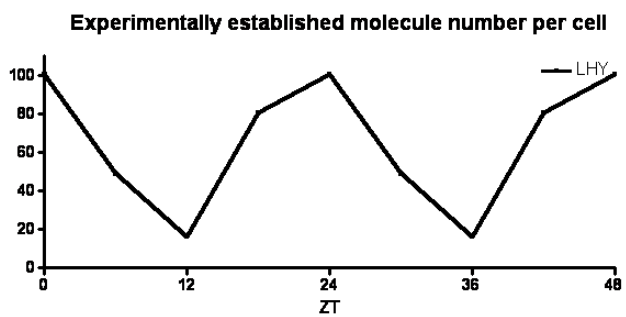
Modelling overview

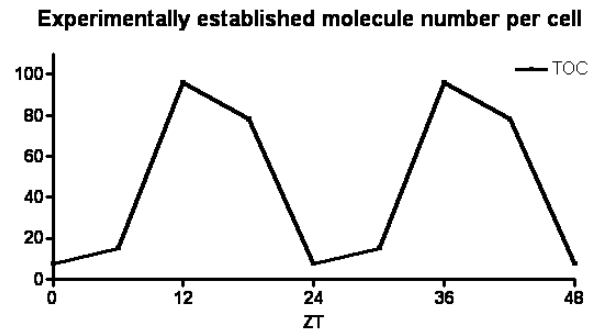
- A previous deterministic (ODE) model of the system had been developed by hand.
- We developed a Bio-PEPA model and validated that its continuous-deterministic interpretation coincided with the original model.
- Stochastic simulation was used with the discrete-stochastic interpretation of the Bio-PEPA model to investigate stochastic fluctuations, focusing on clock phase and sensitivity analysis.
- Model-checking was also used to give more detailed analysis of variability within the system, via specific questions about model behaviour.
- An evolutionary systems biology framework was used to investigate the potential affect low-level mutational effects might have on the phase of the clock.

Model and experimental settings

- Lab experiments in different light conditions and photoperiods
  - DD (24 hours dark)
  - LL (24 hours light)
  - LD 12:12 (12 hours light / 12 hours dark)
  - LD 6:18 (6 hours light / 18 hours dark)
  - LD 18:6 (18 hours light / 6 hours dark)

- Lab measurements on amounts of proteins

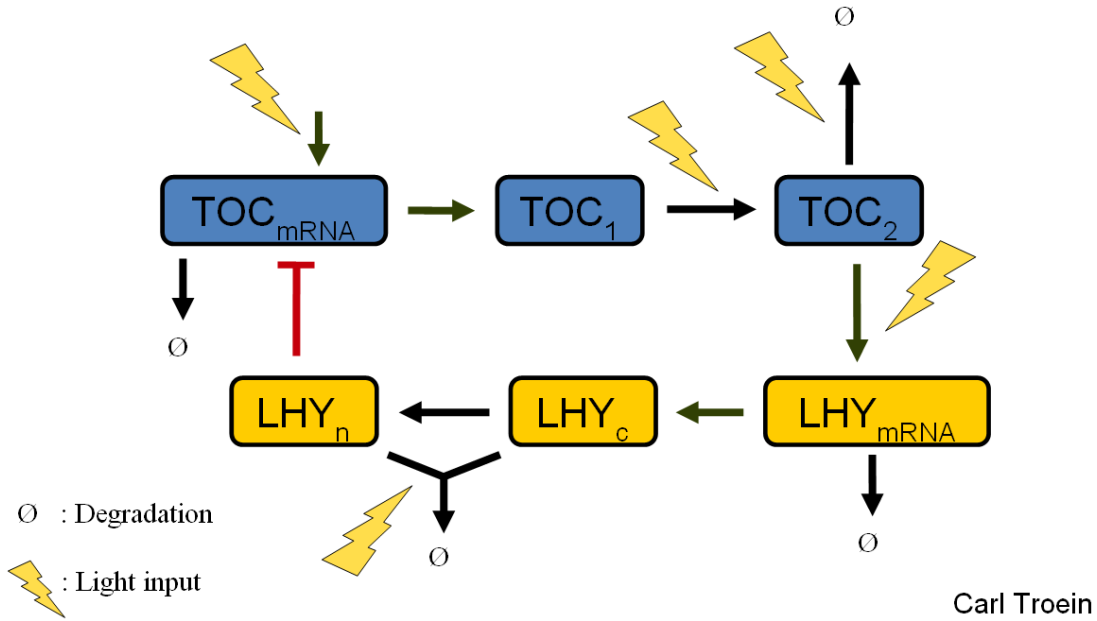




Modelling in Bio-PEPA

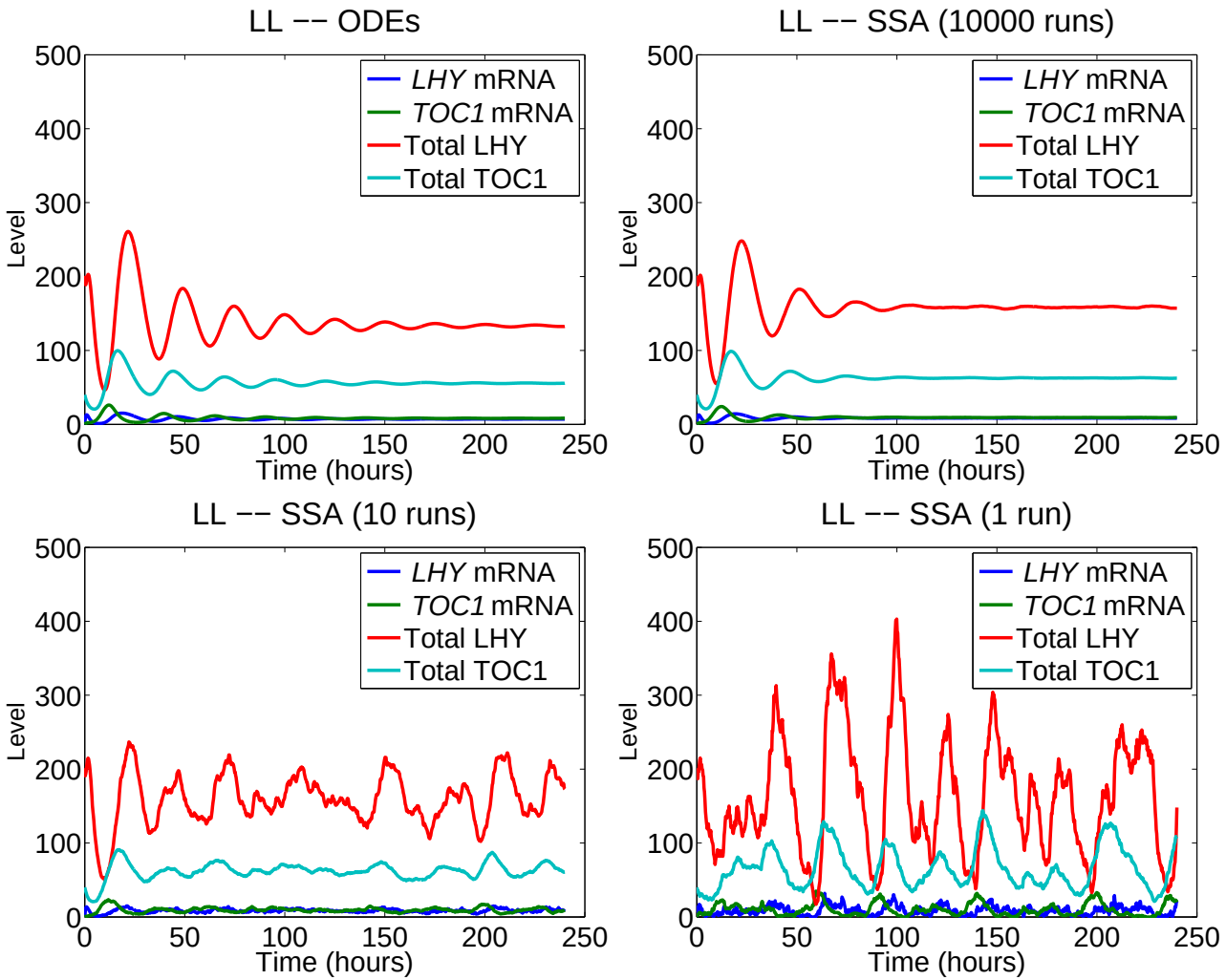
- **Biological species** $\Rightarrow$ interacting **species components**
- **Reactions** $\Rightarrow$ **actions**, with **functional rates** expressing the kinetic rate laws
- Light on/off mechanism for **entrainment to day/night cycle** $\Rightarrow$ **events** switching between day-time and night-time rates

Extracts from the Bio-PEPA model

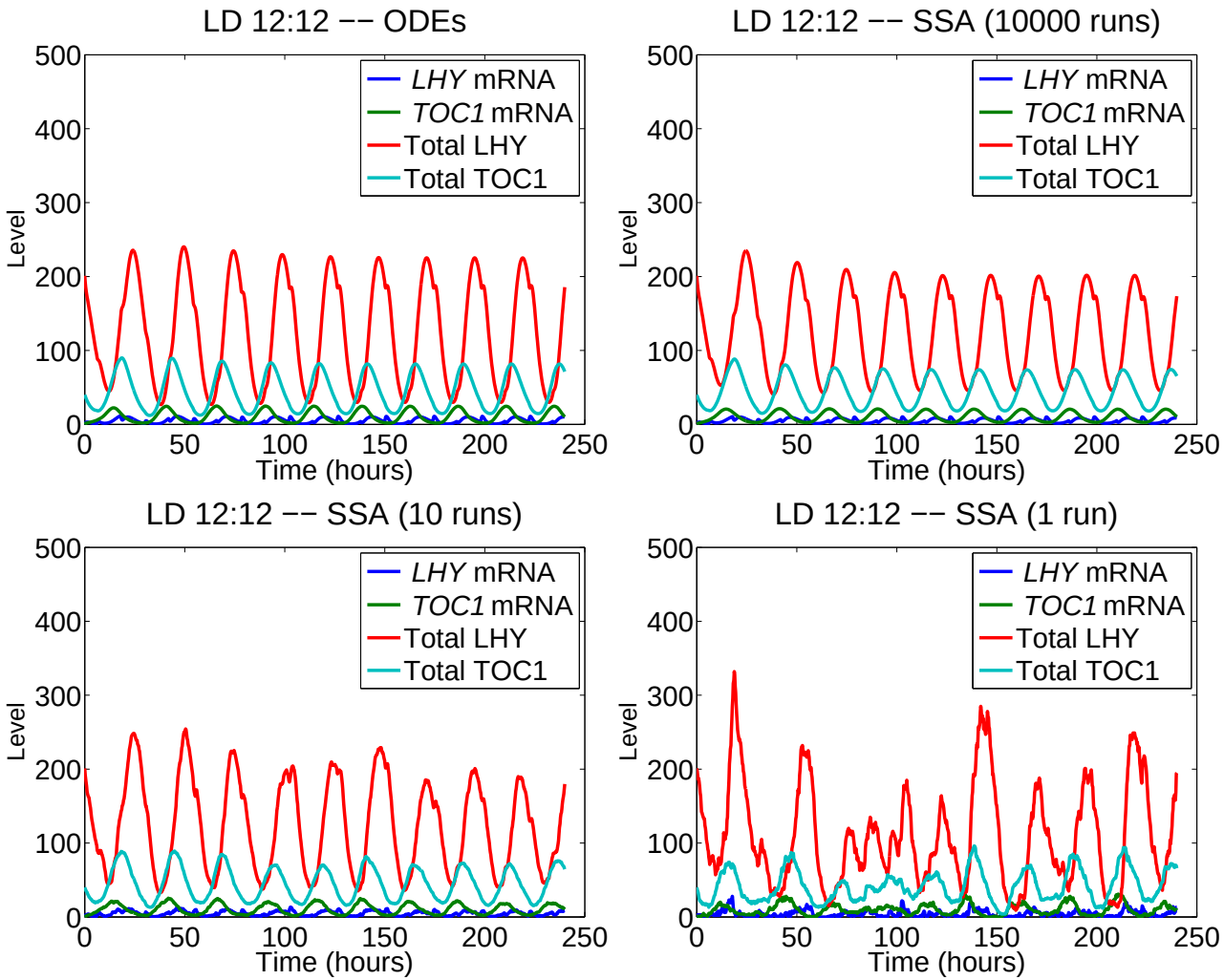


$$
\begin{aligned}
TOC1\_mRNA &= (transc_3, 1)\uparrow + (transl_5, 1) \oplus + (deg_7, 1)\downarrow \\
TOC1\_a &= (deg_4, 1)\downarrow + (conv_6, 1)\uparrow + (transc_8, 1)\oplus \\
TOC1\_i &= (transl_5, 1)\uparrow + (conv_6, 1)\downarrow \\
\\
LHY\_mRNA &= (transc_8, 1)\uparrow + (deg_9, 1)\downarrow + (transl_{10}, 1)\oplus \\
LHY\_c &= (transl_{10}, 1)\uparrow + (transp_{11}, 1)\downarrow + (deg_{12}, 1)\downarrow \\
LHY\_n &= (transc_3, 1)\ominus + (transp_{11}, 1)\uparrow + (deg_{13}, 1)\downarrow \\
\\
acc &= (prod_1, 1)\uparrow + (deg_2, 1)\downarrow + (transc_3, 1)\oplus
\end{aligned}
$$

Simulations – constant light (LL)



Simulations – light/dark (LD 12:12)



Statistical model-checking

- We use statistical model-checking (discrete event simulation and sampling over multiple runs) to make more elaborate queries of the model: approximate results.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Statistical model-checking

- We use statistical model-checking (discrete event simulation and sampling over multiple runs) to make more elaborate queries of the model: approximate results.
- The underlying simulation model is enhanced with a representation of time so that rates of reactions can change appropriately at dawn and dusk.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

Statistical model-checking

- We use statistical model-checking (discrete event simulation and sampling over multiple runs) to make more elaborate queries of the model: approximate results.
- The underlying simulation model is enhanced with a representation of time so that rates of reactions can change appropriately at dawn and dusk.
- The model checker can then be used to investigate the probability distribution of the number of molecules of a species over time.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

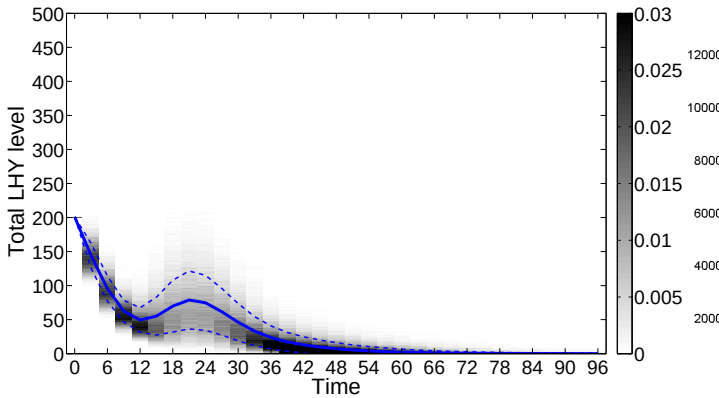
Ostreococcus Tauri

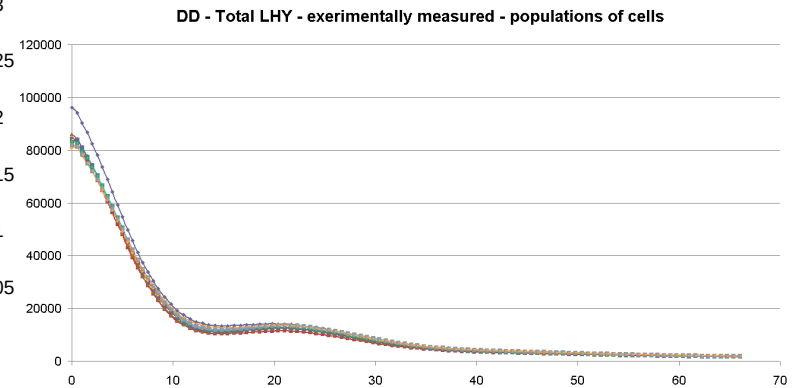
Stochastic modelling and model checking

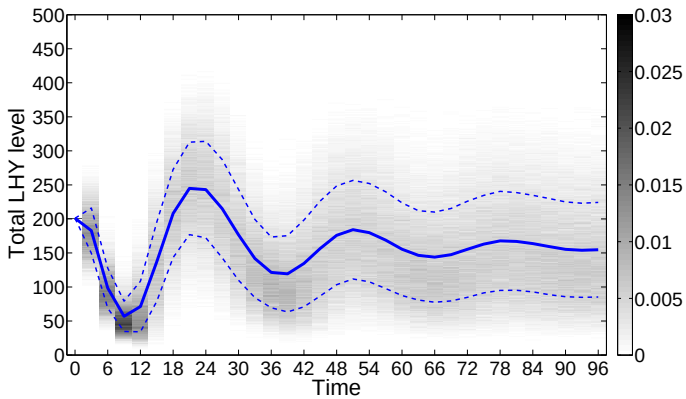
Investigating mutational effects

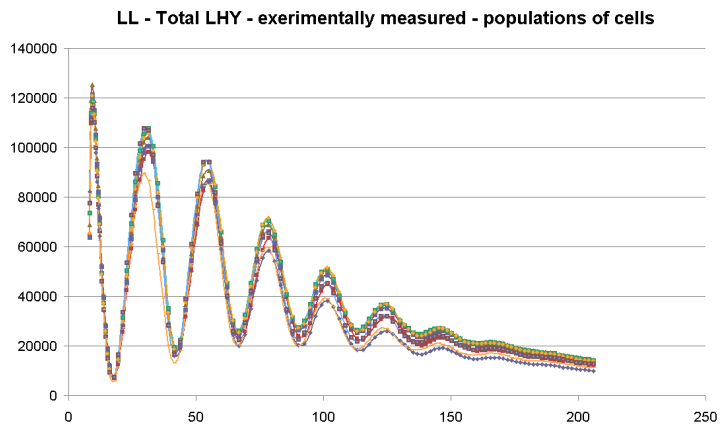
Model-checking – probability distribution of LHY value over 0-96 h

$$\mathcal{P}_{=?}[\mathbf{F}^{[T,T]}\ (\_LHY\_c + \_LHY\_n = level)], T = 0:3:96, level = 0:1:500$$









Probability that the total LHY stays below some threshold e in the long run

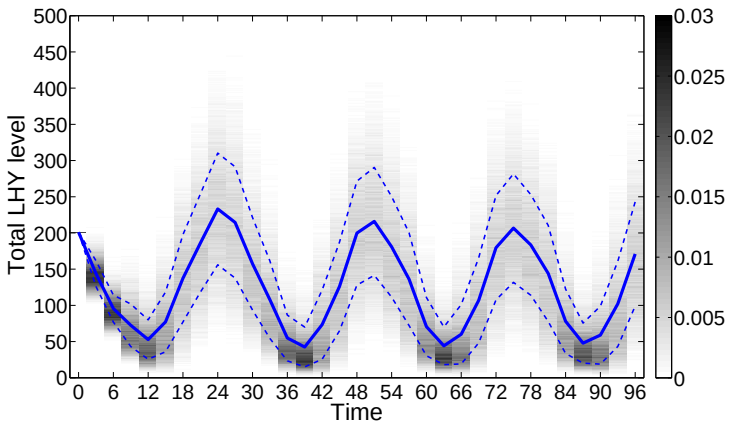
$$\mathcal{P}_{=?}[\mathbf{G}^{[96,500]}\ (LHY\_c + LHY\_n \leq 0 + e)]$$

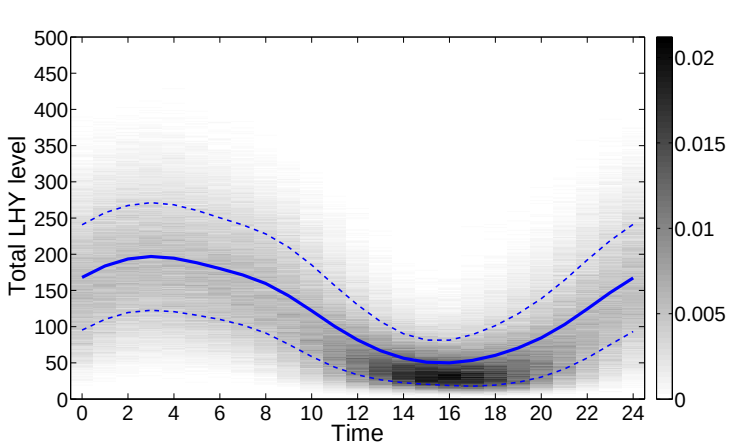
e	0	1	2	3	4
Prob	0.93	0.96	0.96	0.98	0.98

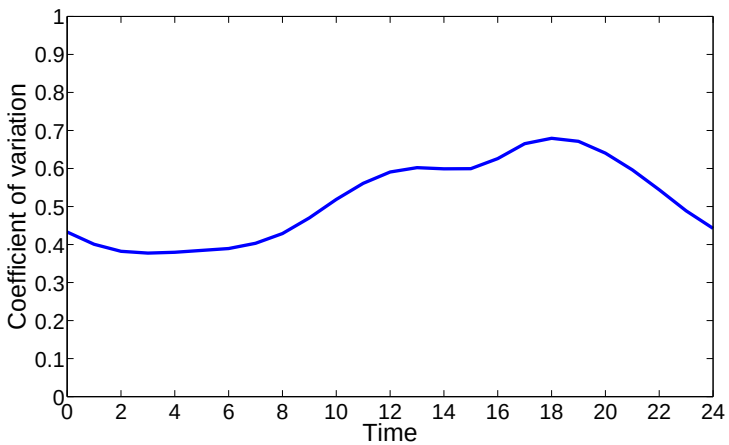
e	5	6	7	8	9	10
Prob	0.99	0.99	0.99	0.99	0.99	1.0

Probability distribution/coefficient of variation of LHY value – LD 12:12

$\mathcal{P}_{=?}[\mathbf{F}^{[T,T]}\ (\_LHY\_c + \_LHY\_n = level)], T = 120 : 1 : 144, level = 0 : 1 : 500$





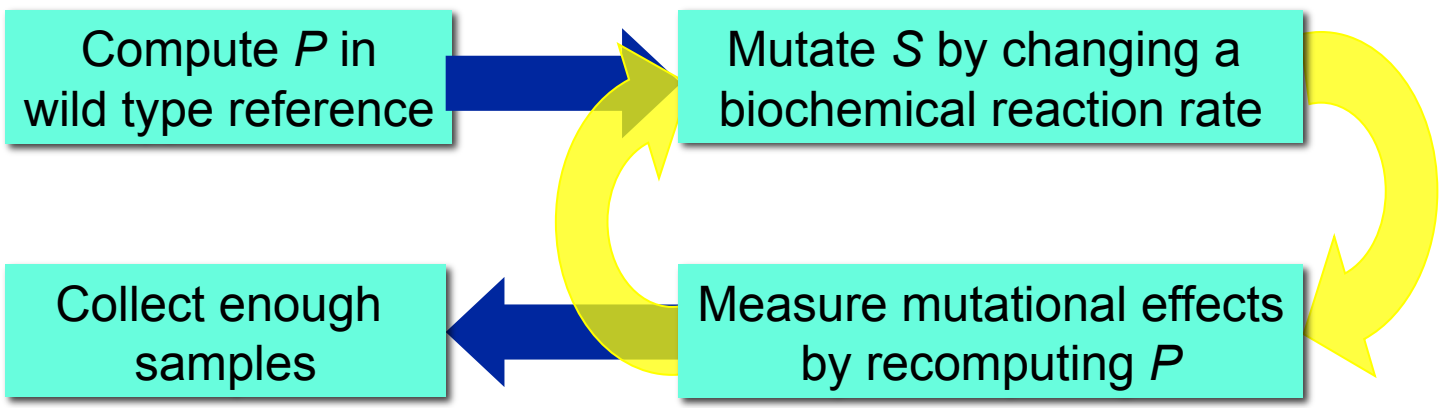


Evolutionary Systems Biology

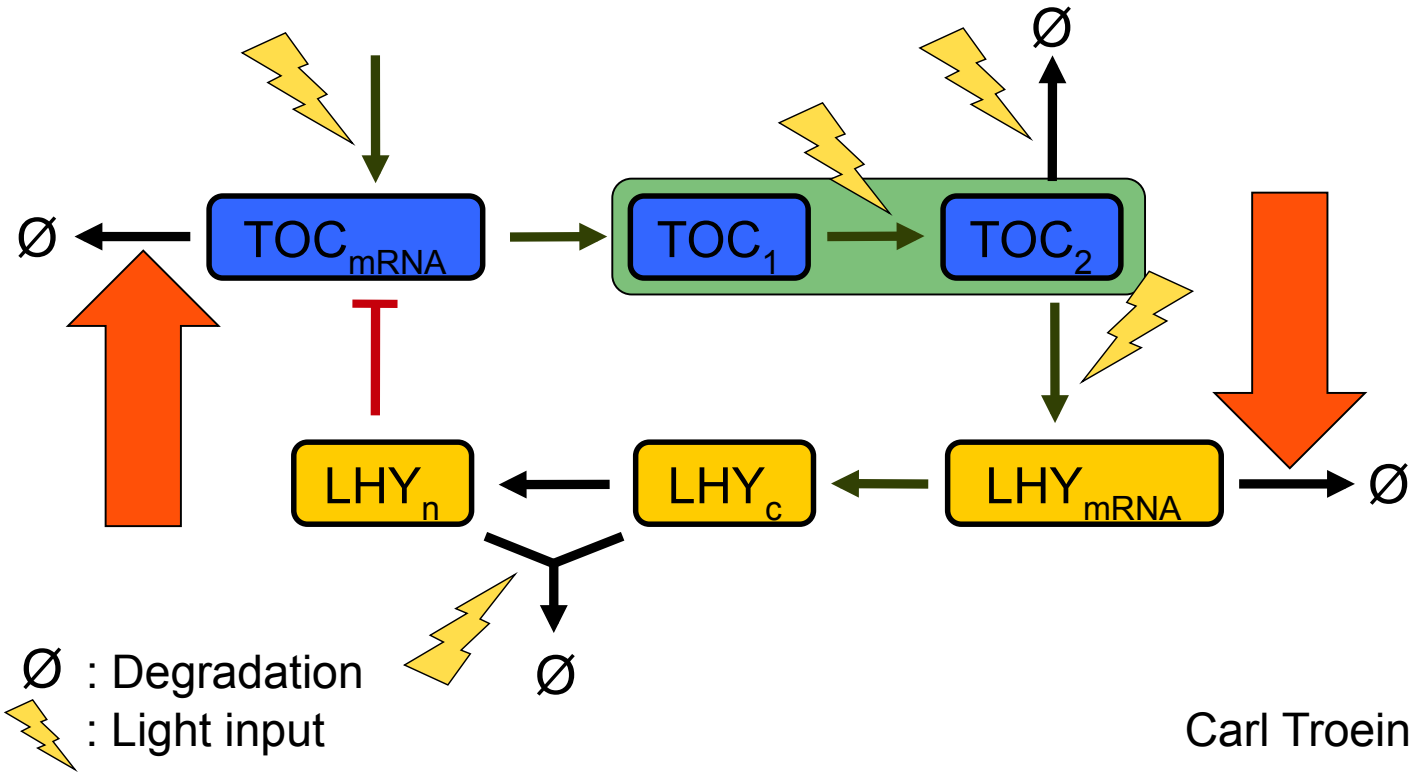
Define system property *P* for systems biology model *S*.

P is a computable property with a link to fitness (e.g. energy costs, accuracy of clock, survival rate).

Assume that non-lethal mutations manifest as variations in the rates of reactions (e.g. due to conformational changes of proteins or mRNAs).

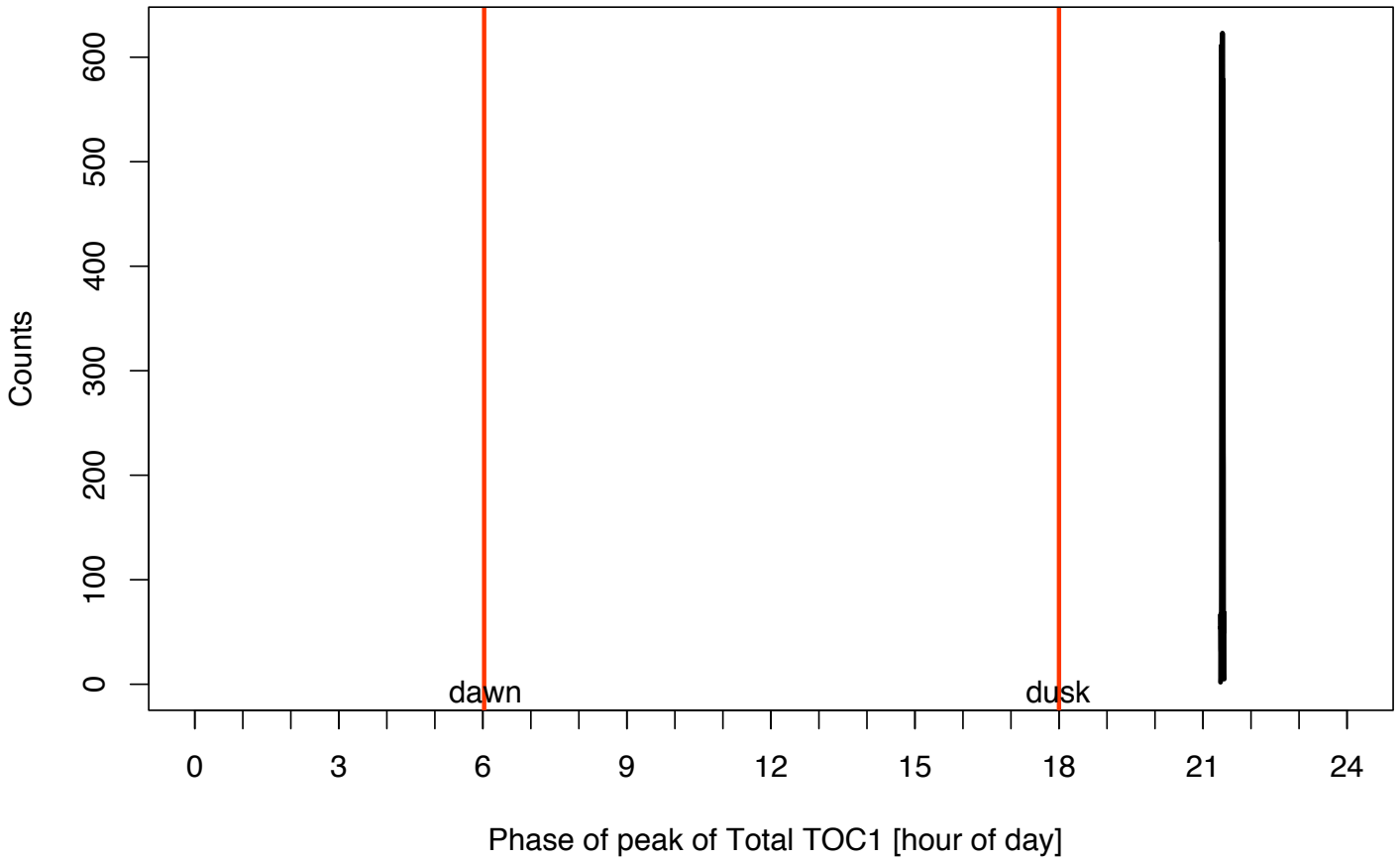


Sensitivity of clock phase to mRNA degradation

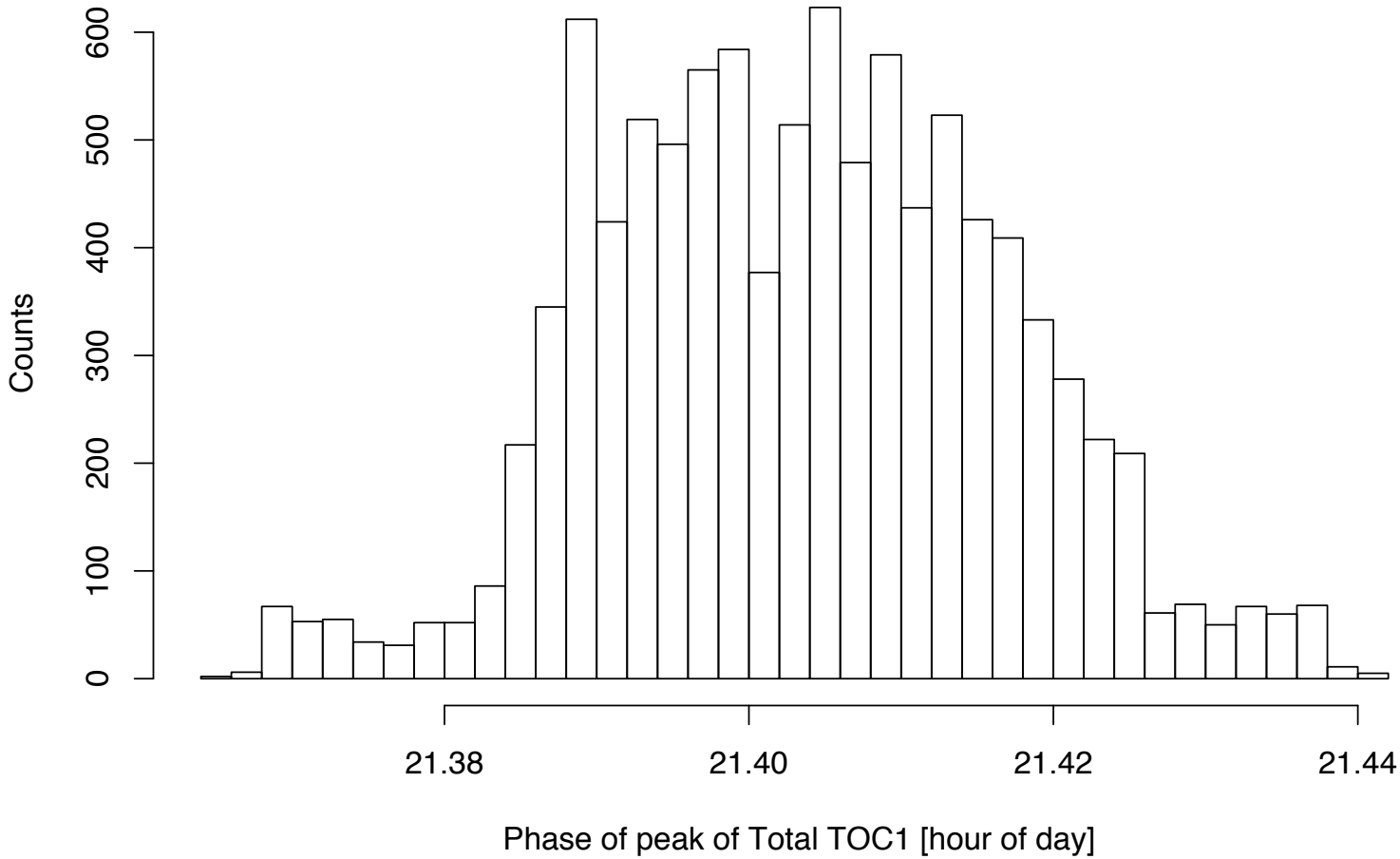


Previous work suggested that the peak of the total amount of TOC is a good indicator of the clock phase.

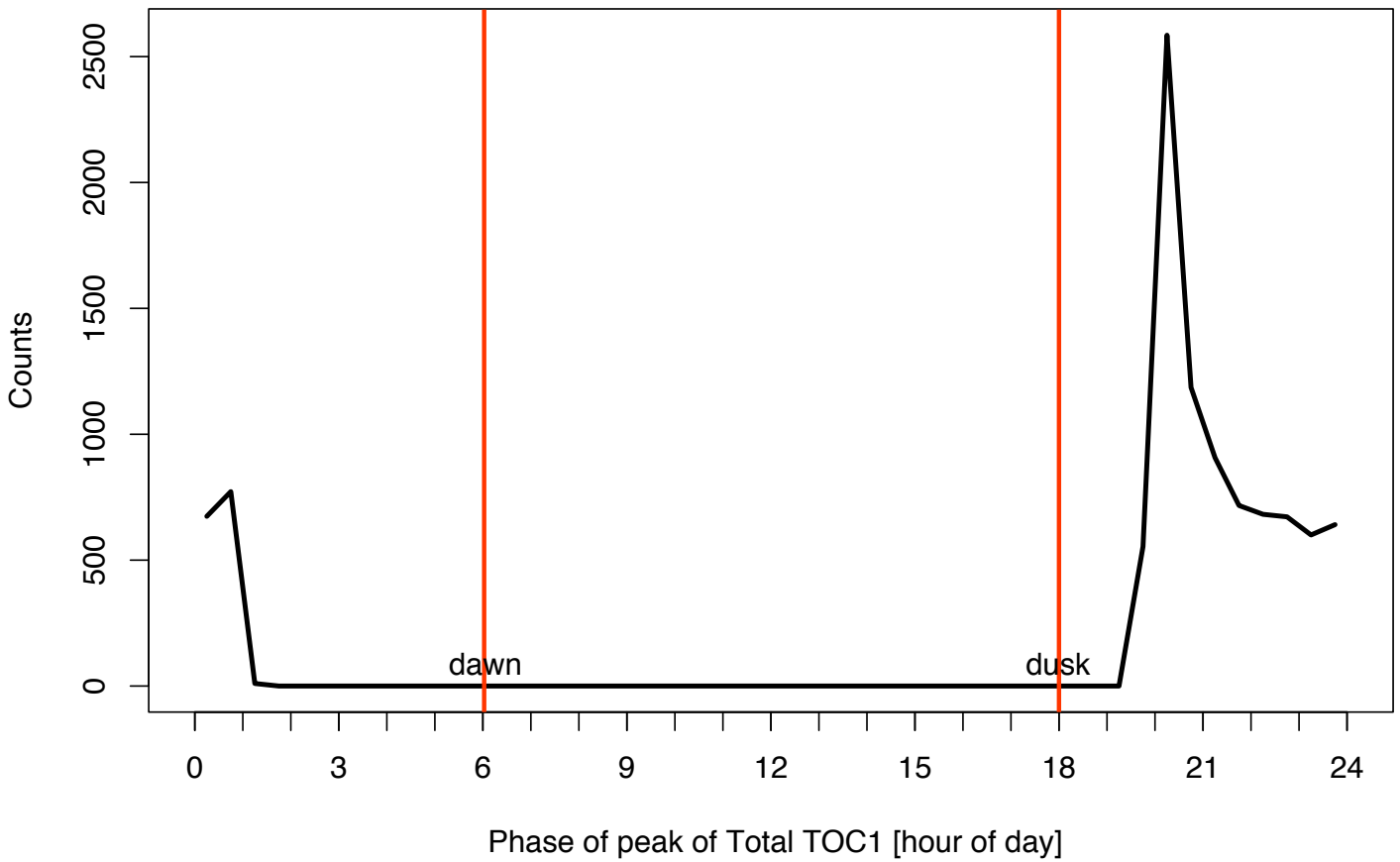
Distribution of phase in wildtype (10,000 runs), large Ω



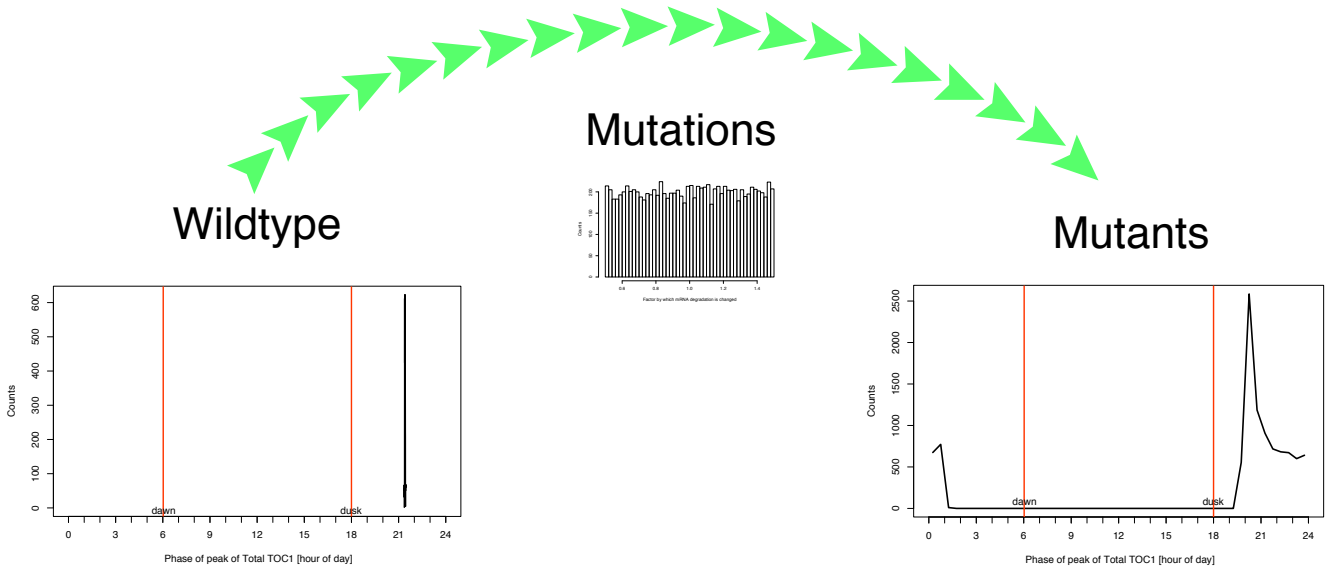
Distribution of phase in wildtype (10,000 runs), large Ω



Distribution of phase in mutant (10,000 runs), large Ω



Evolutionary Systems Biology Framework



Loewe, L. 2009 A framework for evolutionary systems biology. *BMC Systems Biology* 3:27.

- Using this evolutionary systems biology framework we can investigate how low-level mutational effects affect fitness.
- Such small effects are very hard to measure in the lab but become accessible through simulations.
- Bio-PEPA provides a sound modelling basis on which the framework can be built.

On-going work

- Work is continuing with Ostreococcus for both the stochastic simulations and the analysis of mutational effects.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

On-going work

- Work is continuing with Ostreococcus for both the stochastic simulations and the analysis of mutational effects.
- We have considered variations in all rates within the pathway, and found very different impacts on fitness, where fitness is interpreted in terms of how well the biochemical clock corresponds to actual variations in environmental light.

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

On-going work

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

- Work is continuing with Ostreococcus for both the stochastic simulations and the analysis of mutational effects.
- We have considered variations in all rates within the pathway, and found very different impacts on fitness, where fitness is interpreted in terms of how well the biochemical clock corresponds to actual variations in environmental light.
- We have also started working with a more complex organism, the plant Aribadopsis thaliana.

On-going work

Process Algebras for Systems Biology

Bio-PEPA

Modelling Circadian Rhythms

Ostreococcus Tauri

Stochastic modelling and model checking

Investigating mutational effects

- Work is continuing with Ostreococcus for both the stochastic simulations and the analysis of mutational effects.
- We have considered variations in all rates within the pathway, and found very different impacts on fitness, where fitness is interpreted in terms of how well the biochemical clock corresponds to actual variations in environmental light.
- We have also started working with a more complex organism, the plant Aribadopsis thaliana.
- WIthin this context we are investigating means of comparison between stochastic oscillations.

Thank you!

People involved in the work:

- Federica Ciocchetta
- Adam Duguid
- Stephen Gilmore
- Maria Luisa Guerriero
- Laurence Loewe
- Ozgur Akman
- Laura Dixon
- Andrew Millar
- Carl Troein
- Gerben van Ooijen

Thank you!

People involved in the work:

- Federica Ciocchetta
- Adam Duguid
- Stephen Gilmore
- Maria Luisa Guerriero
- Laurence Loewe
- Ozgur Akman
- Laura Dixon
- Andrew Millar
- Carl Troein
- Gerben van Ooijen

Acknowledgements: Engineering and Physical Sciences Research Council (EPSRC) and Biotechnology and Biological Sciences Research Council (BBSRC).