Understanding Intracellular Signalling in Bacterial Chemotaxis

Marcus Tindall

RCUK Fellow in Emerging Technologies for Systems Biology

Institute for Cardiovascular and Metabolic Research,
School of Biological Sciences,
Department of Mathematics & Statistics,
University of Reading.
*E-mail: m.tindall@reading.ac.uk.*

Motivation

• Biological and biomedical sciences are today driven by discoveries at the subcellular level.

• Impact of changes at subcellular level is felt mostly on the multicellular or tissue level cell, e.g. drug discovery.

• Understanding many biological systems requires integrating information across various length scales and also timescales.

• Integrating such information is a major challenge for Systems Biology and related fields.
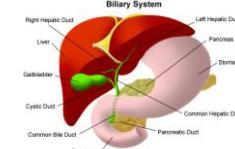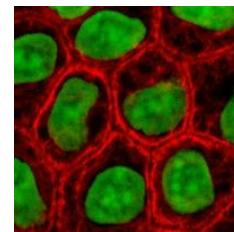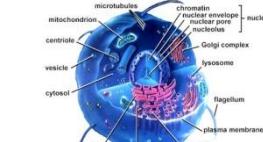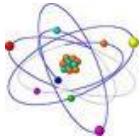

<nm   nm   µm   mm   cm   m

Motivation
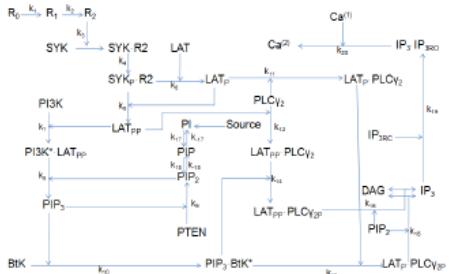
- Cellular biochemistry
- Cell mechanics
- Extracellular matrix
- Extracellular factors (e.g. Nutrients, growth factors)
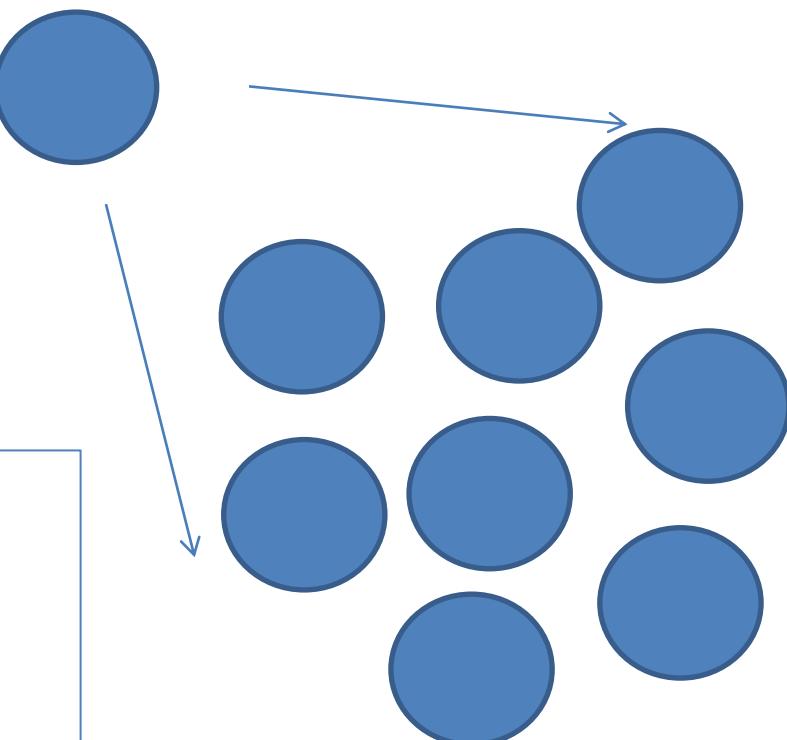
Mathematical Methods

• Deterministic ordinary and partial differential equations (ODEs and PDEs).

• Asymptotic methods.

• Numerical methods for solving ODEs and PDEs.

• Network theory.

• Inverse methods.

• Parameter estimation.

• Agent based modelling.

• Hybrid continuum/discrete methods.

• Multiscale modelling.

Life Science Application Areas

• Cardiovascular cell biology (platelets & cardiac myocytes).

• Lipoprotein metabolism (including models of endocytosis).

• Tumour growth.

• Bacterial chemotaxis.

Ligand

Receptor clusters

CheR

CheA → CheA$_P$

CheB
CheB$_P$

CheY

CheY$_P$

CheZ

E. coli

CheA$_2$ → CheA$_{2P}$

CheY$_3$ CheY$_4$ CheY$_6$

CheY$_{3P}$ CheY$_{4P}$ CheY$_{6P}$

CheA$_3$

CheA$_{3P}$

CheB$_1$

CheB$_2$

CheB$_{1P}$

CheB$_{2P}$

CheA$_2$ → CheA$_{2P}$
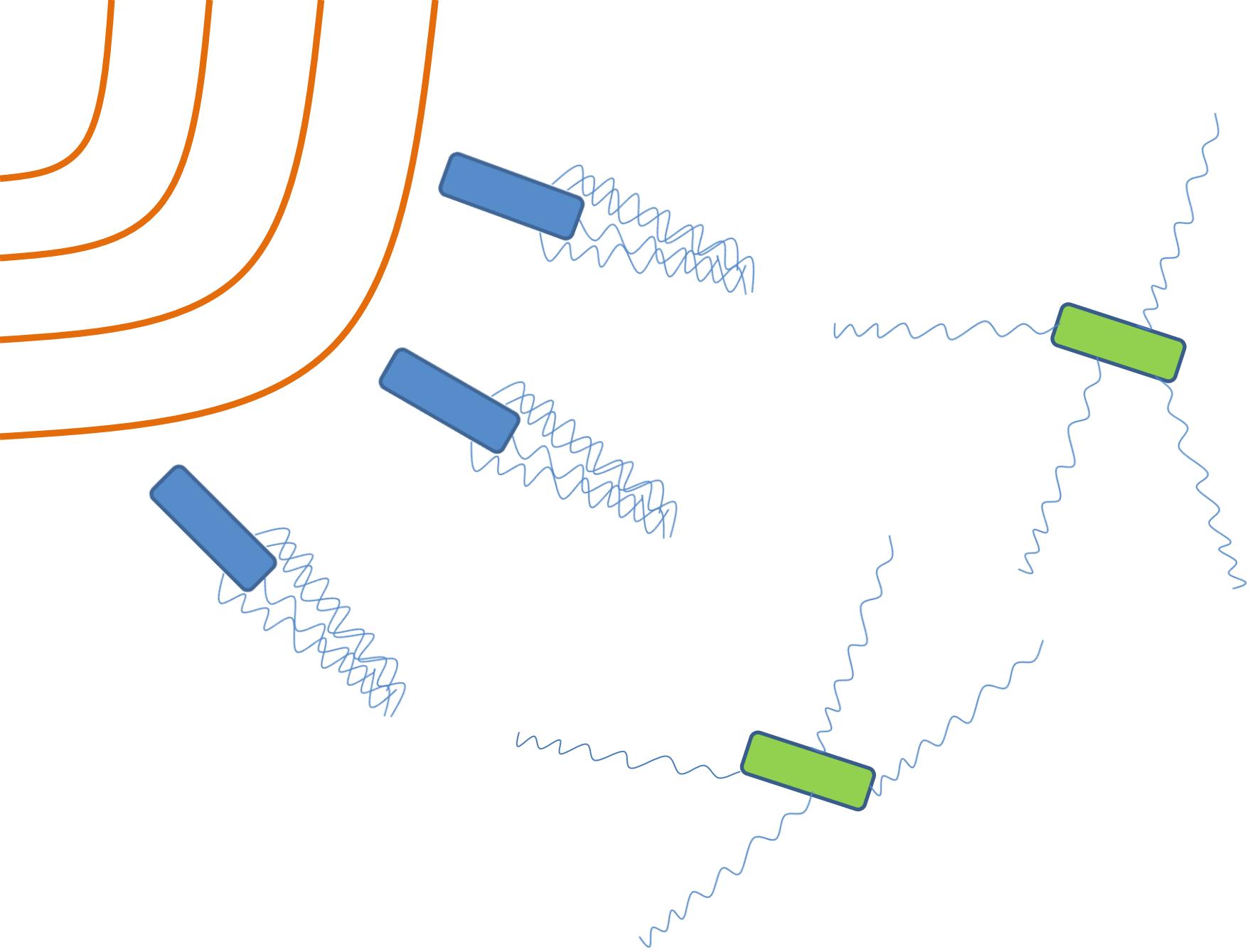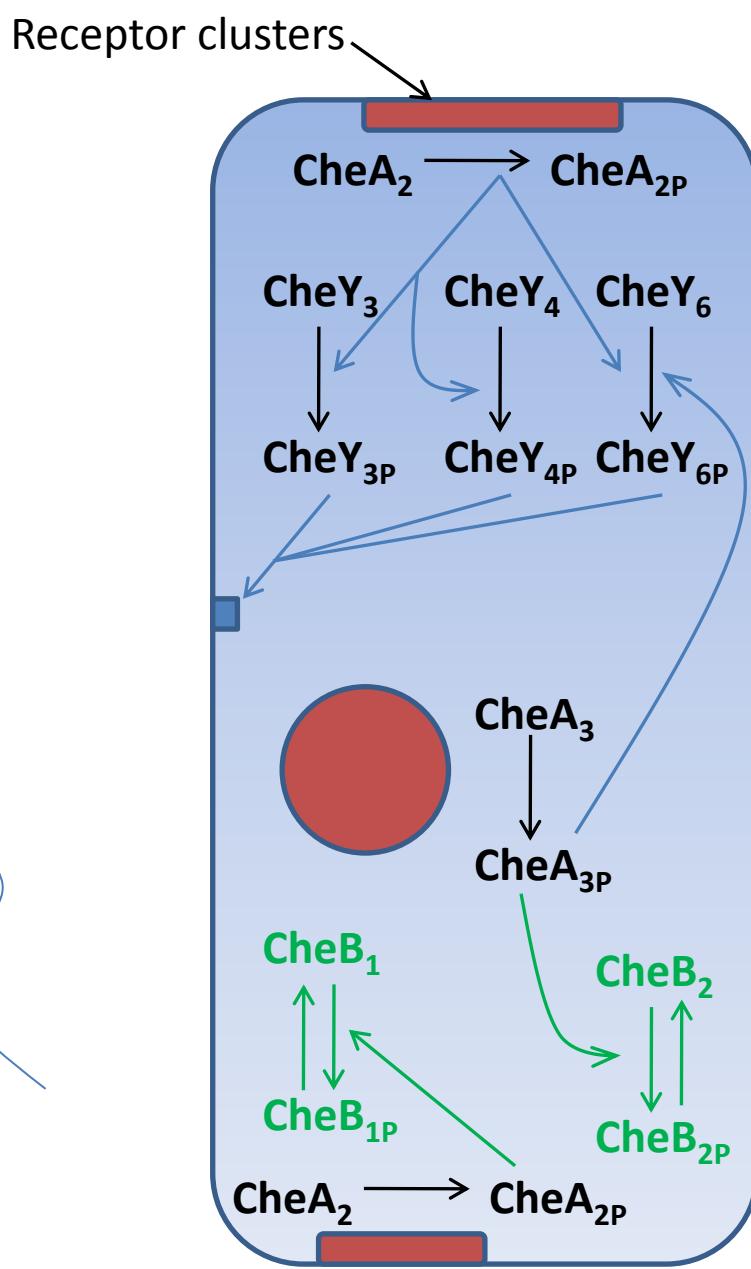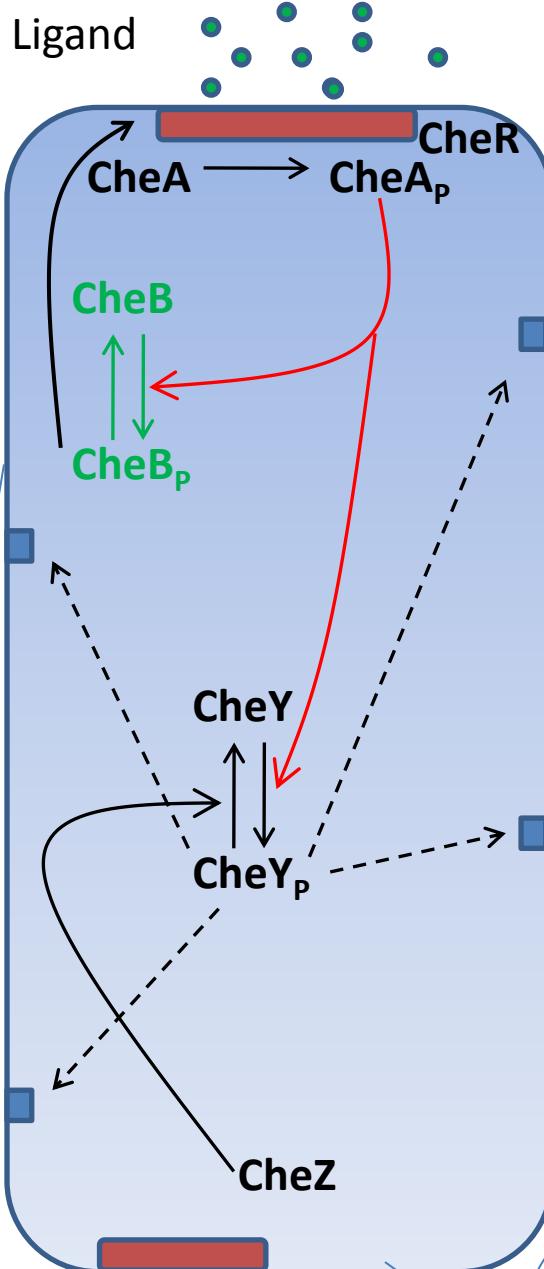
R. sphaeroides

R. sphaeroides – Experimental Data
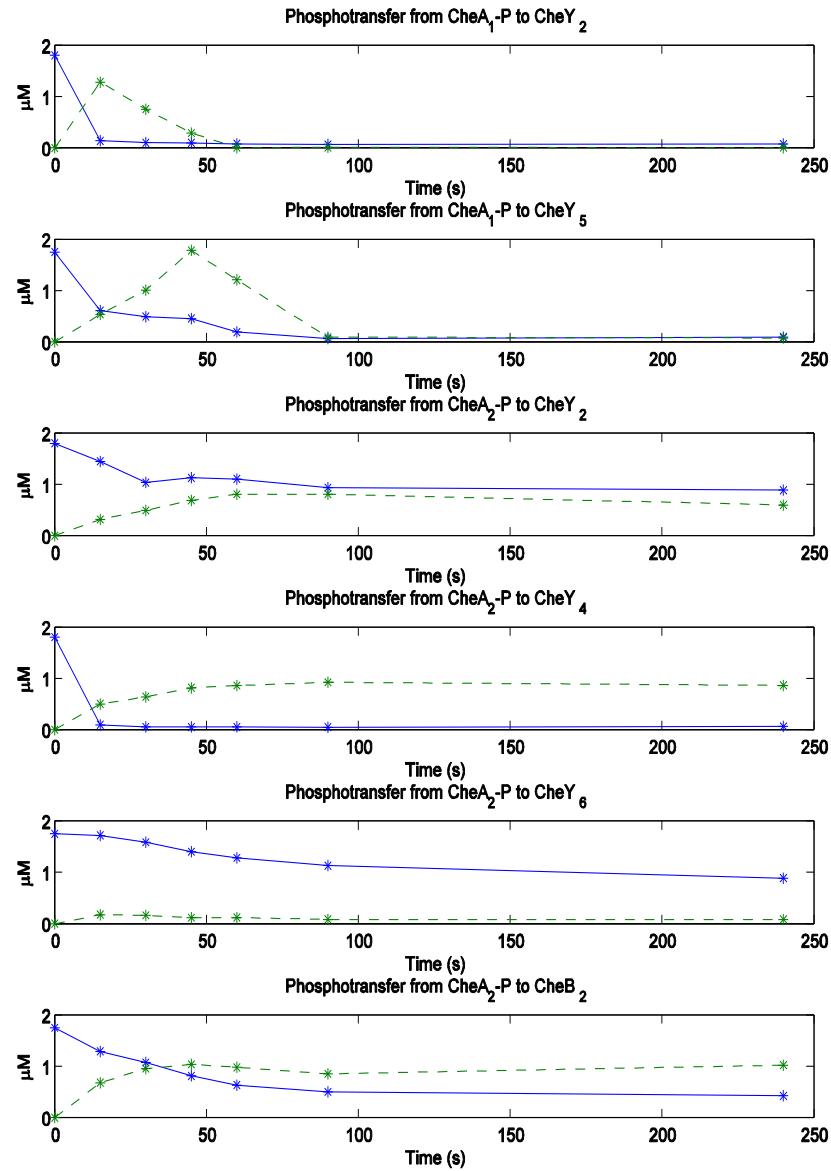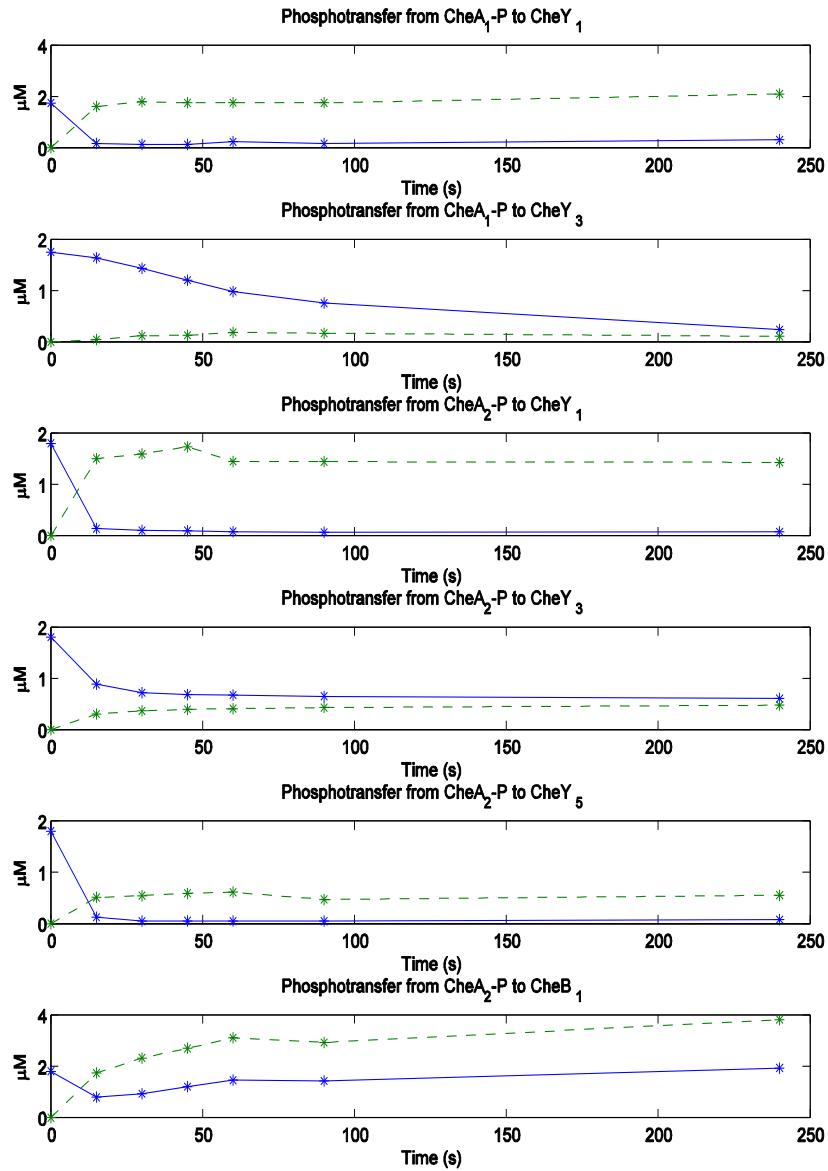
R. sphaeroides – Model parameterisation

Numerical optimisation (parameter fitting)

• Apply and compare a range of numerical optimisation methods for obtaining model parameters from experimental data.

• Allows us to compare various fits to the data to obtain the most robust set of parameters (as well as the method fits).
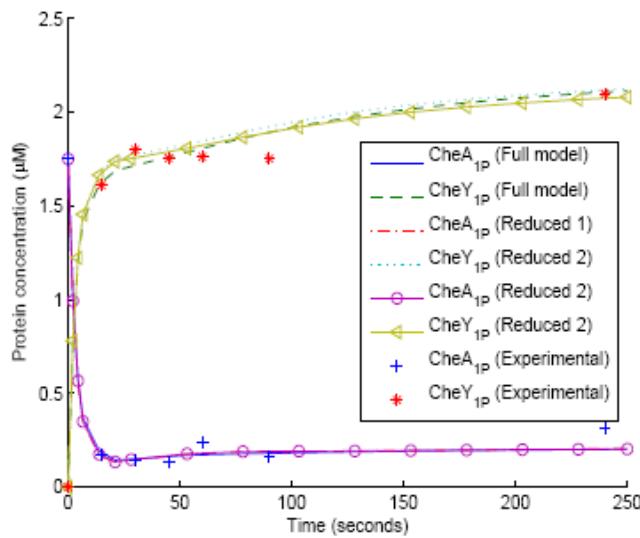
(a) $CheA_{1P}$ to $CheY_1$.
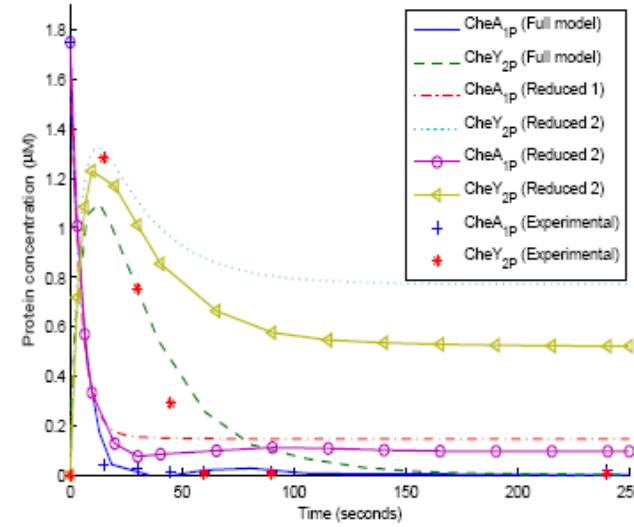
(b) $CheA_{1P}$ to $CheY_2$.

R. sphaeroides - Stimulation of the cytoplasmic cluster
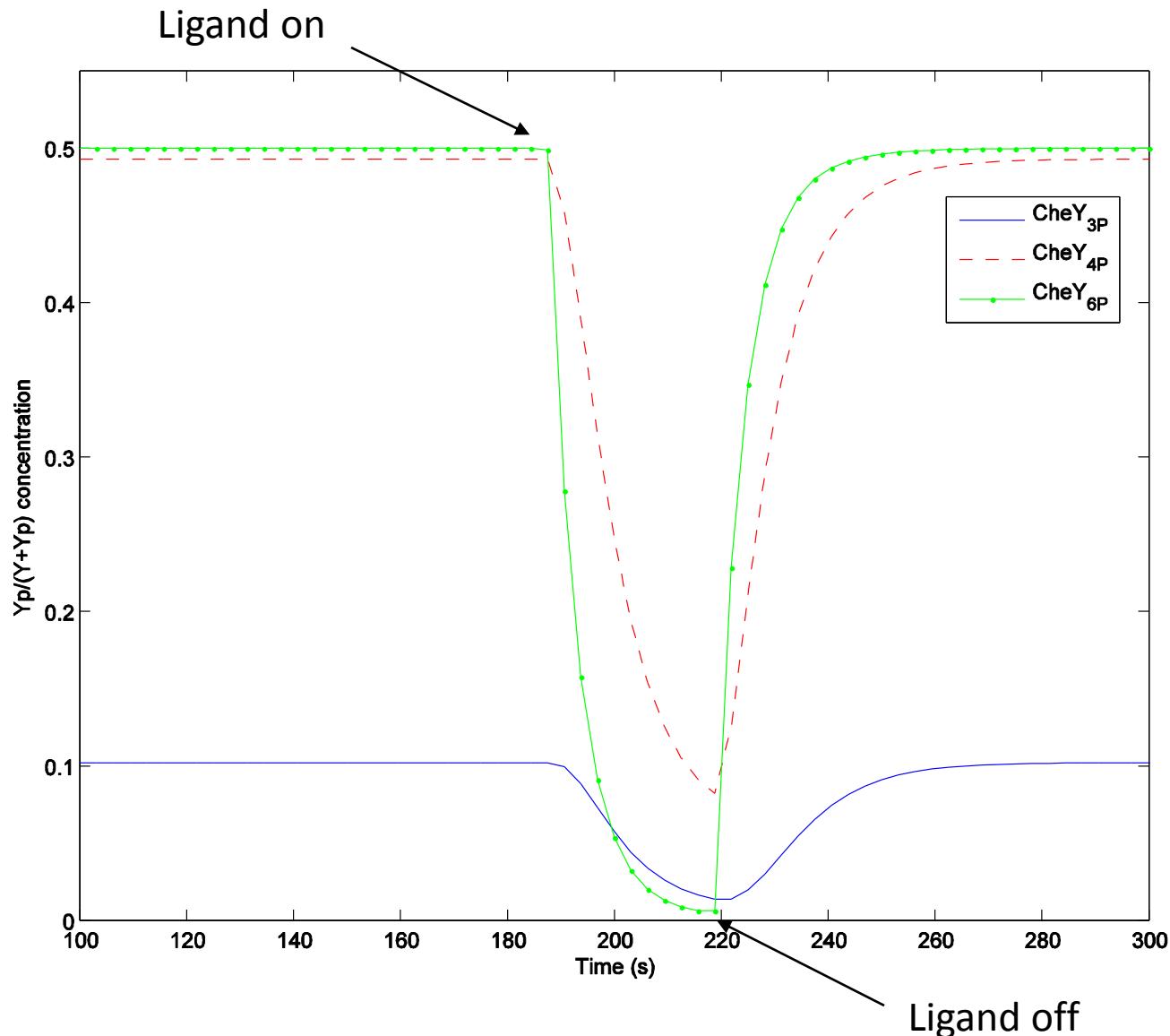
R. sphaeroides - Phosphorelay pathway

$$A_2 \xrightarrow{3.66 \times 10^{-2}} A_{2P}$$

$$A_3 \xrightarrow{4.02} A_{3P}$$

Gives back A_{2P}!

Y_3

$12.73 \downarrow \uparrow 12.41$

Y_{3P}

$\downarrow 0.34$

X

Y_4

$1.71 \times 10^3 \downarrow \uparrow 246.11$

Y_{4P}

$\downarrow 1.26$

X

Y_6

$2.97 \downarrow \uparrow 3.00$

Y_{6P}

$\downarrow 1.00$

X

Y_6

$1.45 \times 10^3 \downarrow \uparrow 0.40$

Y_{6P}

$\downarrow 1.00$

X

Receptor cluster

Cytoplasmic cluster
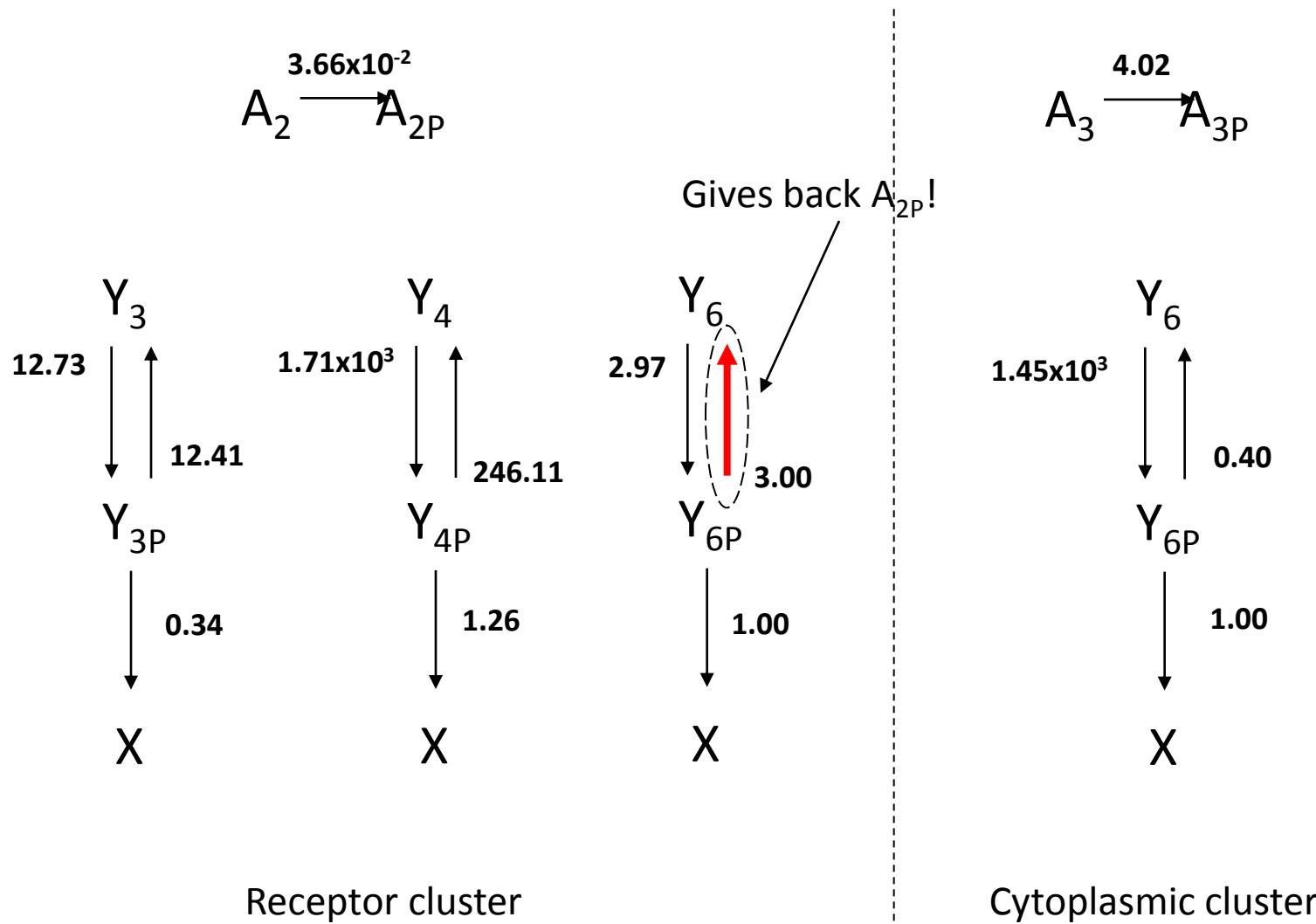
R. sphaeroides - Experimental Results

Time (s)	A3-P	B2-P	A2-P
0	21.21	0	0
15	20.58	0.97	0.03
30	21.55	0.92	0.03
60	20.55	0.92	0.05
120	19.70	0.94	0.07
240	19.29	1.00	0.11

E. coli – Phosphotransfer Reactions

Process	Reaction	Details
Autophosphorylation	$CheA \xrightarrow{k_1} CheA_P$	
Phosphotransfer	$CheA_P + CheY \xrightarrow{k_2} CheA + CheY_P$	$CheA_P$ to CheY
	$CheA_P + CheB \xrightarrow{k_3} CheA + CheB_P$	$CheA_P$ to CheB
Dephosphorylation	$CheY_P + CheZ \xrightarrow{k_4} CheY + CheZ$	Dephosphorylation by CheZ
	$CheY_P \xrightarrow{k_5} CheY$	Natural dephosphorylation
	$CheB_P \xrightarrow{k_5} CheB$	Natural dephosphorylation

E. coli - Complex formation

$$A \xrightarrow{k_1(L)} A_P$$

$$A_P + Y \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} \overset{(I_1)}{A_P \cdot Y} \underset{k_{-7}}{\overset{k_7}{\rightleftharpoons}} \overset{(I_2)}{A \cdot Y_P} \underset{k_{-8}}{\overset{k_8}{\rightleftharpoons}} A + Y_P$$

$$\Big\uparrow k_{10}(L)$$

$$A + Y \underset{k_{-9}}{\overset{k_9}{\rightleftharpoons}} \underset{(I_3)}{A \cdot Y} \xleftarrow{k_6}$$

$$Y_P + Z \underset{k_{-11}}{\overset{k_{11}}{\rightleftharpoons}} \overset{(I_4)}{Y_P \cdot Z} \xrightarrow{k_{12}} Y + Z \qquad Y_P \xrightarrow{k_6} Y$$

$$A_P + B \xrightarrow{k_3} A + B_P \qquad B_P \xrightarrow{k_5} B$$
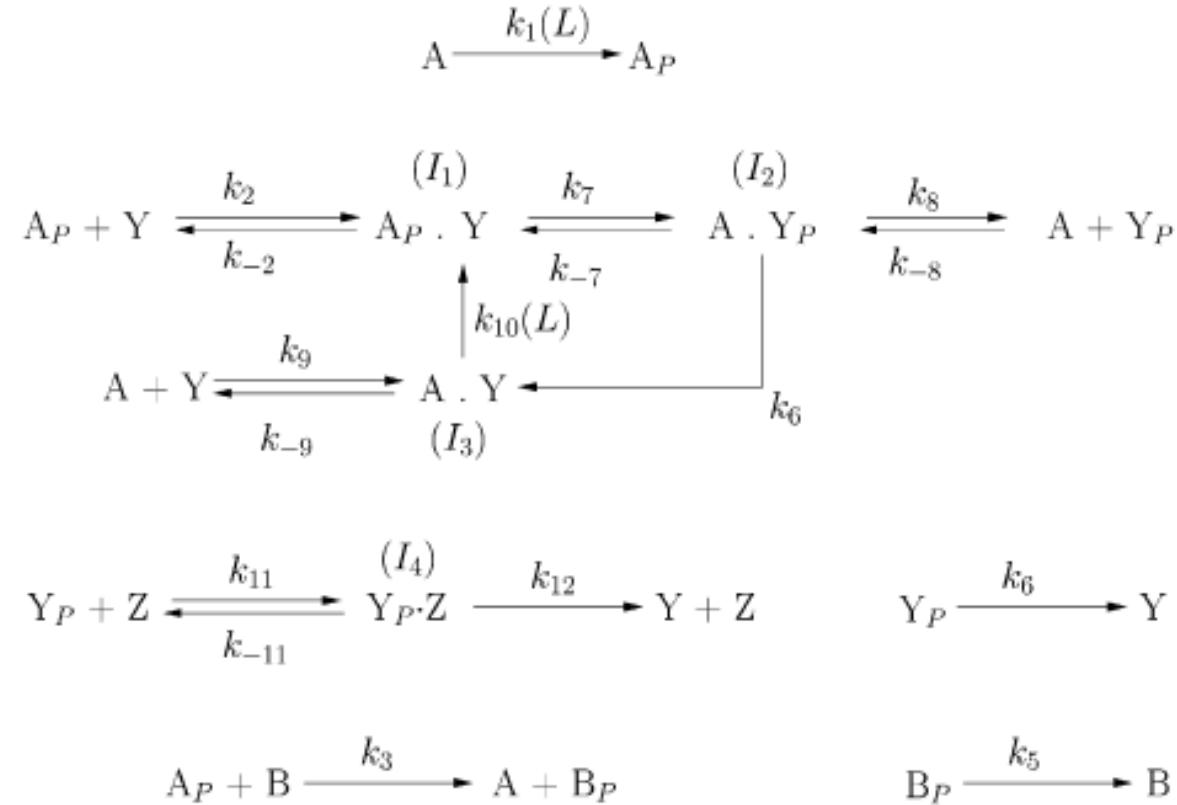
E. coli Complex formation - Parameterisation

Table 1

Parameter values used in our model as obtained from the indicated sources.

Parameter	Description	Value	Reference
k_1	Rate of CheA autophosphorylation	$3.75\ s^{-1}$	Assumed
k_2	Rate of phosphotransfer from $CheA_P$ + CheY to I_1	$2.50 \times 10^6\ (Ms)^{-1}$ [c]	Stewart (1997)
k_{-2}	Rate of I_1 to $CheA_P$ and CheY	$15\ s^{-1}$ [c]	Stewart (1997)
k_3	Rate of phosphotransfer from $CheA_P$ to CheB	$1.5 \times 10^7\ (Ms)^{-1}$	Bray website data[b]
k_5	Rate of $CheB_P$ natural dephosphorylation	$0.7\ s^{-1}$	Stewart (1993)
k_6	Rate of $CheY_P$ natural dephosphorylation and $A \cdot Y_p$ to $A \cdot Y$	$8.5 \times 10^{-2}\ s^{-1}$	Sourjik and Berg (2002a)
k_7	Rate of I_1 to I_2	$650\ s^{-1}$	Stewart (1997)
k_{-7}	Rate of I_2 to I_1	$50\ s^{-1}$	Stewart (1997)
k_8	Rate of I_2 to CheA + $CheY_P$	$250\ s^{-1}$ [d]	Li et al. (1995)
k_{-8}	Rate of CheA + $CheY_P$ to I_2	$2.08 \times 10^7\ (Ms)^{-1}$ [d]	Li et al. (1995)
k_9	Rate of CheA + CheY to I_3	$7.50 \times 10^6\ (Ms)^{-1}$	Stewart and van Bruggen (2004)
k_{-9}	Rate of I_3 to CheA + CheY	$15\ s^{-1}$	Stewart and van Bruggen (2004)
k_{10}	Rate of I_3 to I_1	$3.75\ s^{-1}$	Assumed equivalent to k_1
k_{11}	Rate of $CheY_P$ + CheZ to I_4	$5.60 \times 10^6\ (Ms)^{-1}$	Silversmith et al. (2008)
k_{-11}	Rate of I_4 to $CheY_P$ + CheZ	$0.04\ s^{-1}$	Silversmith et al. (2008)
k_{12}	Rate of I_4 to CheY + CheZ	$4.90\ s^{-1}$	Silversmith et al. (2008)
D_Y	CheY diffusion coefficient	$10\ \mu m^2\ s^{-1}$	Elowitz et al. (1999) Segall et al. (1985)
D_{Y_P}	$CheY_P$ diffusion coefficient	$10\ \mu m^2\ s^{-1}$	Segall et al. (1985) Elowitz et al. (1999)
D_B	CheB diffusion coefficient	$7\ \mu m^2\ s^{-1}$	Assumed[a]
D_{B_P}	$CheB_P$ diffusion coefficient	$7\ \mu m^2\ s^{-1}$	Assumed[a]
A_T	Total CheA concentration	$7.9\ \mu M$ [e]	Bray website data[b]
Y_T	Total CheY concentration	$9.70\ \mu M$	Bray website data[b]
B_T	Total CheB concentration	$0.28\ \mu M$	Bray website data[b]
Z_T	Total CheZ concentration	$3.8\ \mu M$ [e]	Bray website data[b]
L_x	Average length of a cell	$3\ \mu m$	Darnton et al. (2007)
L_y	Average width of a cell	$1\ \mu m$	Darnton et al. (2007)

[a] Calculated using the Einstein diffusion approximation.
[b] www.pdn.cam.ac.uk/groups/comp-cell/Rates.html.
[c] Approximated using the stated dissocation value of $K_D = 6\ \mu M$ as a guide.
[d] Calculated from a dissociation value of $K_D = 4.0\ \mu M$.
[e] In our model setup the total size of the polar region is approximately 1/10th the volume of the cell and thus these concentrations are increased tenfold.

E. coli Complex formation – Wild-type cell
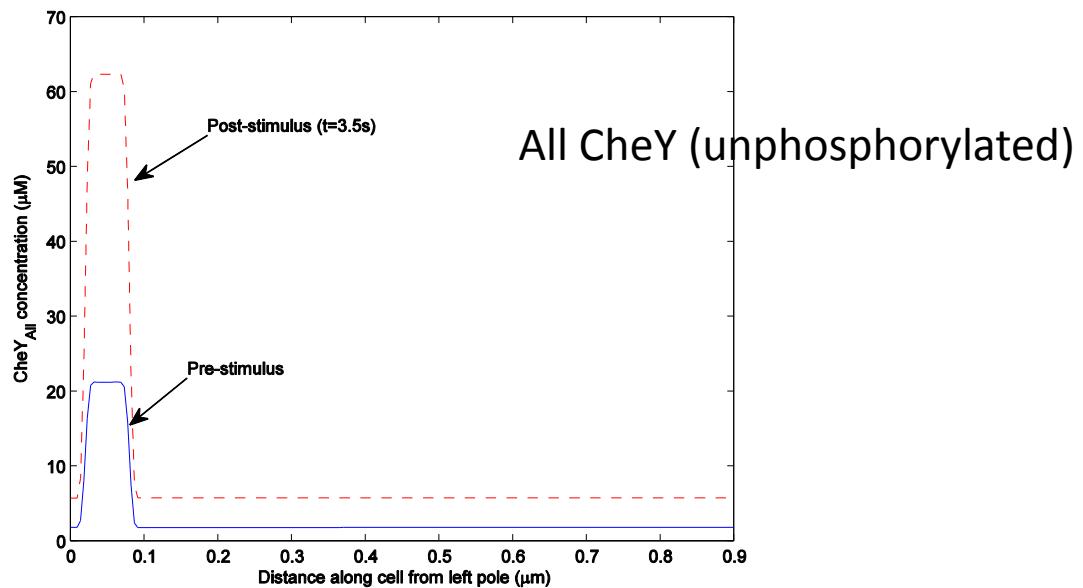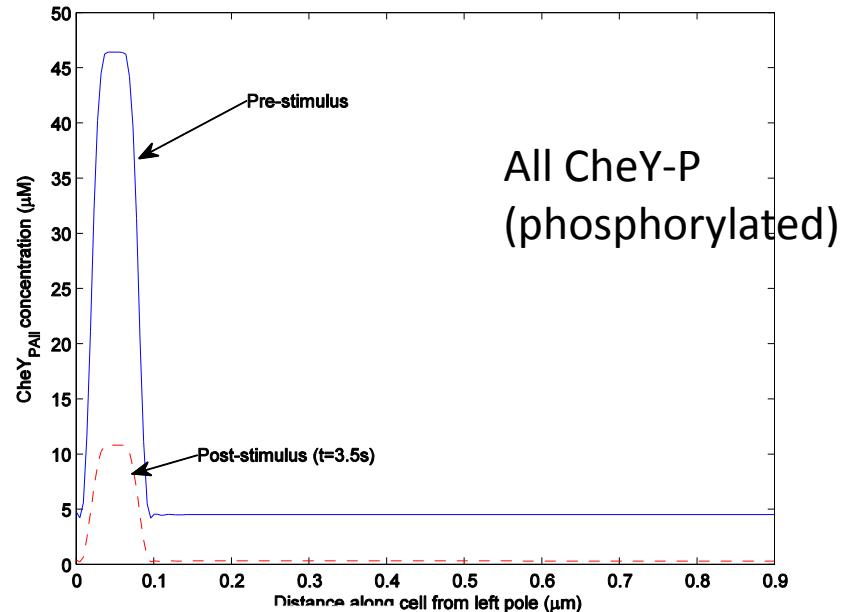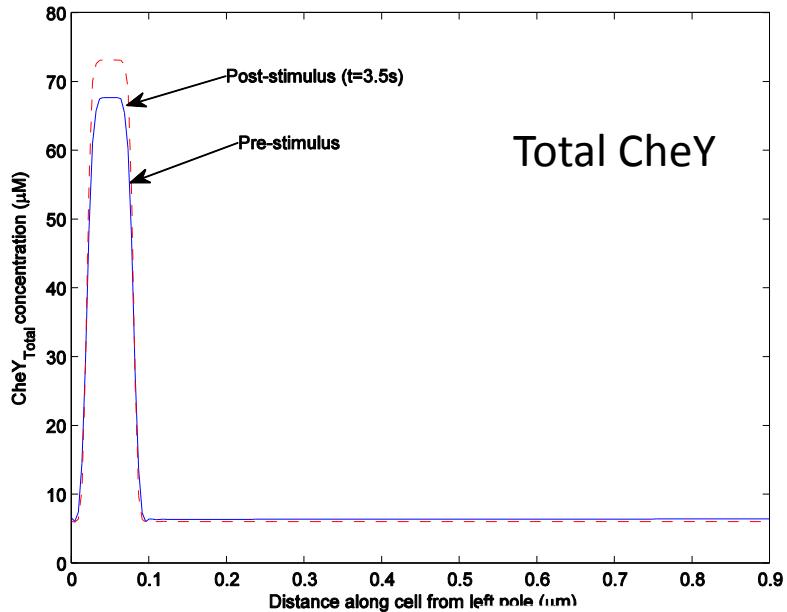
E. coli Complex formation

	CheY free	CheY$_P$ free	CheA·CheY	CheA·CheY$_P$	CheA$_P$·CheY	CheY$_P$·CheZ
Wild-type ($Z_T = 3.8$ μM)						
Pre-stimulus	18.56	46.97	10.20	5.83	0.59	17.91
Post-stimulus	58.62	3.15	32.22	0.41	0.07	5.51
Low CheZ ($Z_T = 0.1$ μM)						
Pre-stimulus	1.74	95.23	0.16	2.17	0.18	0.51
Post-stimulus	4.7	81.98	2.85	9.22	0.73	0.51
High CheZ ($Z_T = 12$ μM)						
Pre-stimulus	40.64	6.55	22.77	1.33	0.32	28.39
Post-stimulus	60.81	0.81	32.56	0.18	0.06	5.57

R. sphaeroides - Adaptation
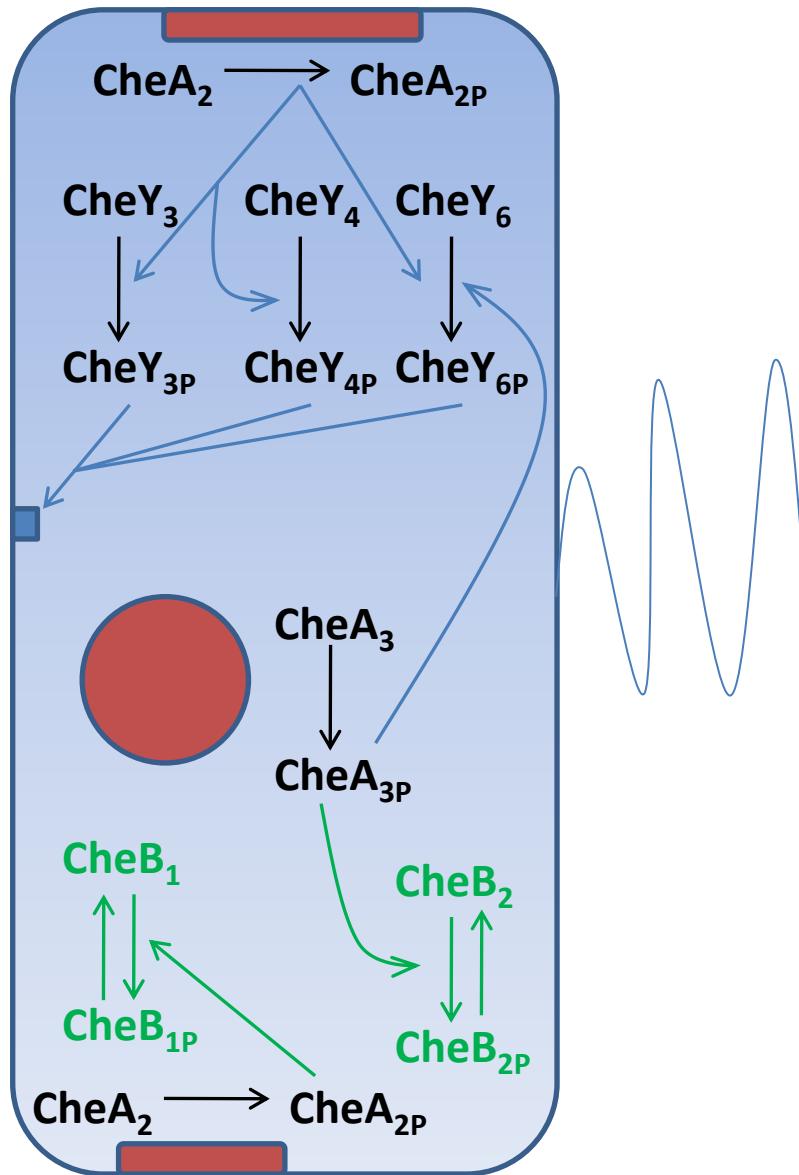
Current Work

- Elucidating adaptation in *R. sphaeroides.*

- Reducing model complexity in respect of protein complex formation.

- Developing spatiotemporal models (incorporating diffusion) of intracellular signalling in *R. sphaeroides.*

Acknowledgements

• Dr Steven Porter, Biological Sciences, University of Exeter.

• Dr George Wadhams, Dept. of Biochemistry, University of Oxford.

• Prof. Judy Armitage, Dept. of Biochemistry, University of Oxford.

• Prof. Philip Maini, Mathematical Institute, University of Oxford.

Publications

Tindall, M.J., Determining model parameters from experimental data: Model reduction, numerical optimisation and asymptotic methods. In preparation.

Tindall, M.J., Gaffney, E., Maini, P.K. and Armitage, J.P. Theoretical insights into bacterial chemotaxis (Invited review).

Tindall, M.J., Porter, S.L., Maini, P.K. and Armitage, J.P. Modelling chemotaxis reveals the role of reversed phosphotransfer and a bi-functional phosphatase. PloS Comput. Biol., 6(8), e1000896, 2010.

Tindall, M.J., Porter, S.L., Wadhams, G., Maini, P.K. and Armitage, J.P. Spatiotemporal modelling of CheY complexes in *Escherichia coli* chemotaxis. *Prog. Biophys. Mol. Biol.,* E-print PMID: 19540260, 2009.

Tindall, M.J., Maini, P.K., Armitage, J.P., Singleton, C. and Mason, A. Intracellular signalling during bacterial chemotaxis in *Practical Systems Biology* (Taylor and Francis), 61, 161-174, 2008.

Tindall, M.J., Porter, S.L., Maini, P.K., Gaglia, G. and Armitage, J.P. Overview of mathematical approaches used to model bacterial chemotaxis I: The single cell. *Bull. Math. Biol.,* 70(6), 1525-69, 2008.

Tindall, M.J., Maini, P.K., Porter, S.L. and Armitage, J.P. Overview of mathematical approaches used to model bacterial chemotaxis II: Bacterial populations. *Bull. Math. Biol.,* 70(6), 1570-607, 2008.