Mechanism-based pharmacodynamic model of a β-lactam (LAC)/β-lactamase inhibitor (BLI) combination against *Eschericia coli*

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PAGANZ

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β-lactam (LAC)/β-lactamase (BLI)

- β-lactamase-mediated hydrolysis
  - Class A to D based on amino acid sequence homology
- ESBL-producing *E. coli*
  - ↑ morbidity, mortality, healthcare-related costs

Metallo-β-lactamase?
Extended-spectrum-β-lactamase?

Objective

- To develop a mechanism-based model that characterizes the pharmacodynamics of LAC/BLI against different β-lactamase producing *E. coli* strains

Methods

- 4 strains of *E. coli*

STRAIN	β-LACTAMASE		LAC MIC (mg/L)				
	Type	Ambler Class	BLI 0 mg/L	BLI 1 mg/L	BLI 4 mg/L	BLI 16 mg/L	BLI 64 mg/L
#LAPD1	None	-	**0.25**	-	-	-	-
#LAPD2	AmpC	C	**4**	4	2	0.5	0.5
#LAPD3	CMY-10	C	**8**	4	4	1	0.5
#LAPD4	CTXM-15	A	**>128**	>128	2	2	1

Methods

- Time-kill studies
  - ~10^6 CFU/mL; 48 h
  - Log_{10}CFU/mL *versus* time

		LAC (mg/L)											
		LAC 0		LAC 1		LAC 4		LAC 16		LAC 64		LAC 256	
BLI (mg/L)	BLI 0	BLI 0	LAC 0	BLI 0	LAC 1	BLI 0	LAC 4	BLI 0	LAC 16	BLI 0	LAC 64	BLI 0	LAC 256
	BLI 1	BLI 1	LAC 0	BLI 1	LAC 1	BLI 1	LAC 4	BLI 1	LAC 16	BLI 1	LAC 64	BLI 1	LAC 256
	BLI 4	BLI 4	LAC 0	BLI 4	LAC 1	BLI 4	LAC 4	BLI 4	LAC 16	BLI 4	LAC 64	BLI 4	LAC 256
	BLI 16	BLI 16	LAC 0	BLI 16	LAC 1	BLI 16	LAC 4	BLI 16	LAC 16	BLI 16	LAC 64	BLI 16	LAC 256
	BLI 64	BLI 64	LAC 0	BLI 64	LAC 1	BLI 64	LAC 4	BLI 64	LAC 16	BLI 64	LAC 64	BLI 64	LAC 256

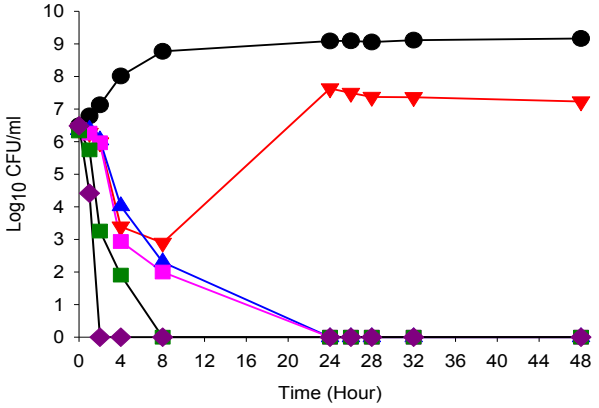
Methods

- PD structural model development
  - Time-kill data arranged according to fixed BLI concentrations
    - 5 data sets per strain
  - Best-fit curves obtained for each data set using a maximum likelihood method (ADAPT V5.0.42)
  - Model discrimination
    - Rule of parsimony
    - Akaike's Information Criterion (AIC)
  - Variance model
    - SD^2

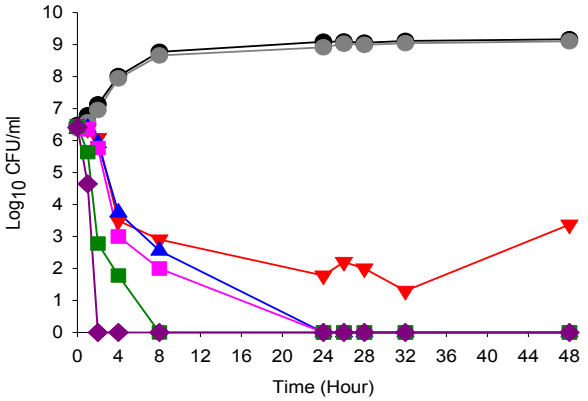
Results: Time-kill

#LAPD1
(LAC MIC 0.25 mg/L)

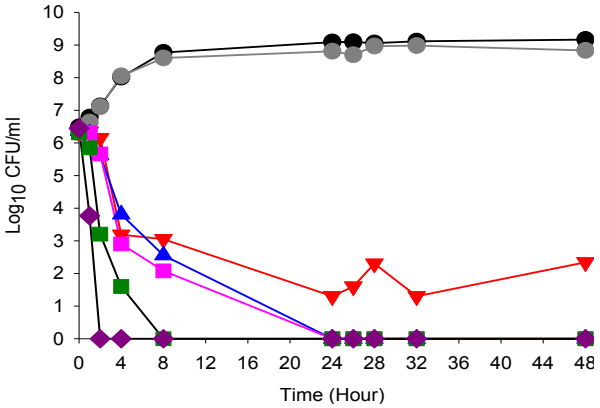
A. BLI 0 mg/L



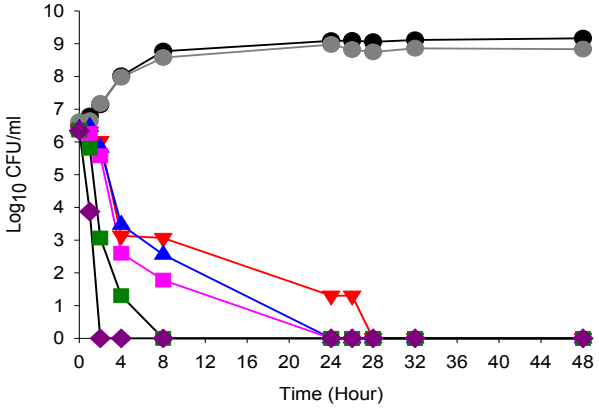
B. BLI 1 mg/L



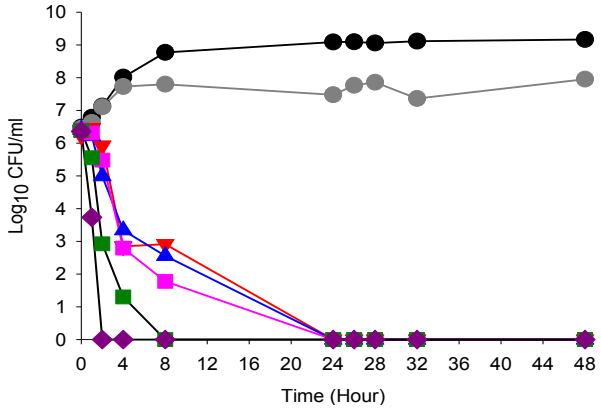
C. BLI 4 mg/L

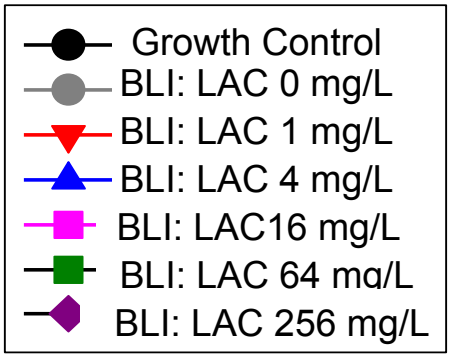


D. BLI 16 mg/L



E. BLI 64 mg/L



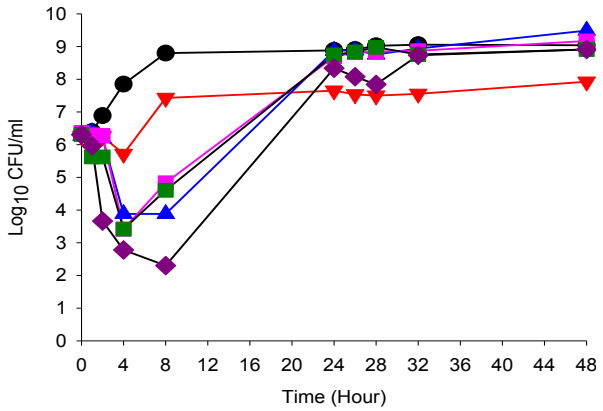


Results: Time-kill

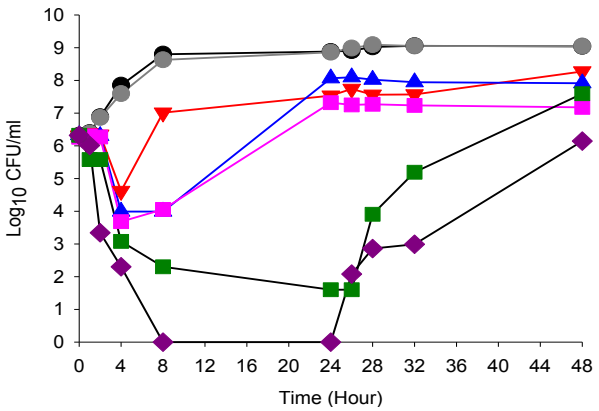
#LAPD2

(LAC MIC 4 mg/L)

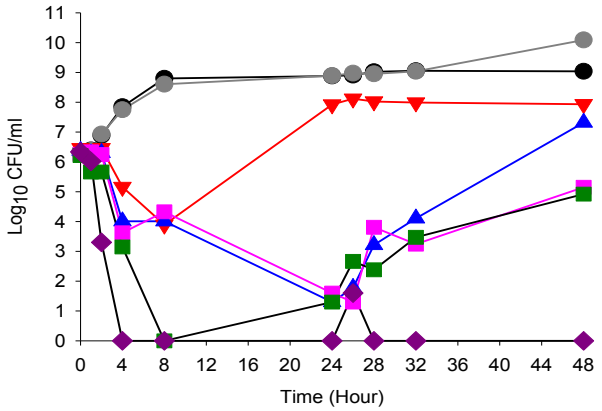
A. BLI 0 mg/L



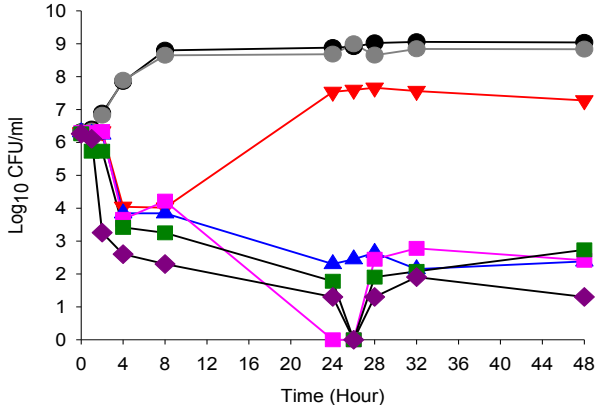
B. BLI 1 mg/L



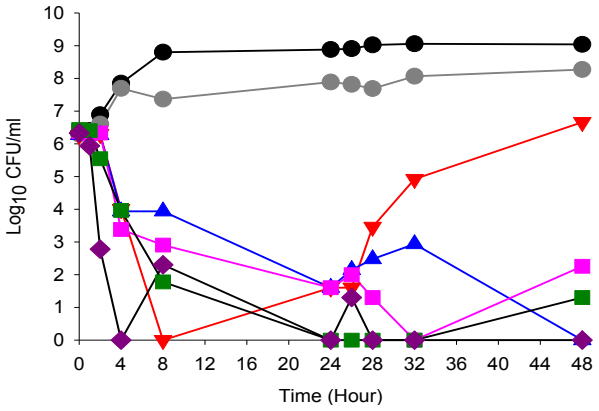
C. BLI 4 mg/L

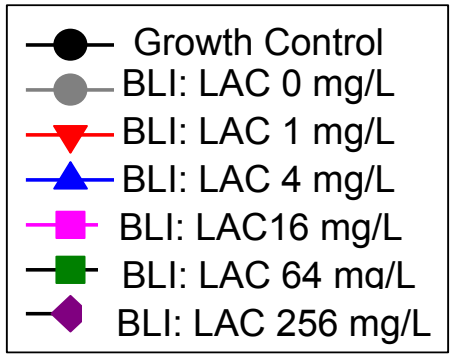


D. BLI 16 mg/L



E. BLI 64 mg/L

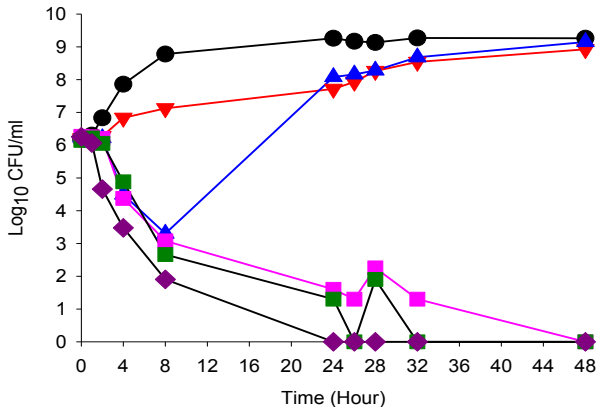




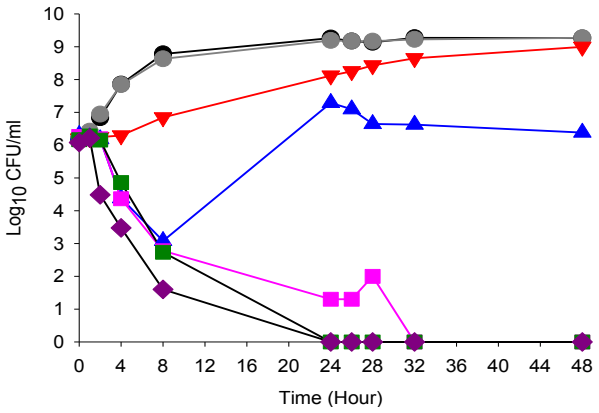
Results: Time-kill

#LAPD3
(LAC MIC 8 mg/L)

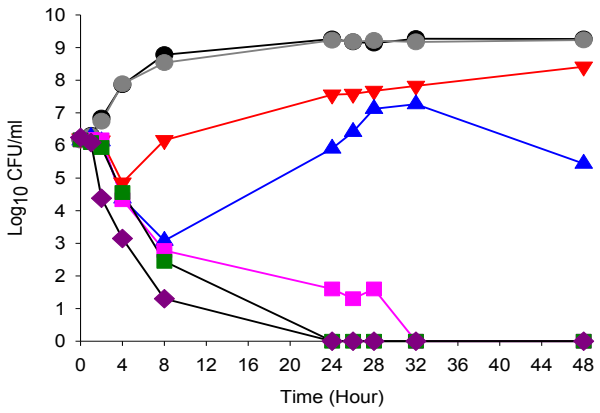
A. BLI 0 mg/L



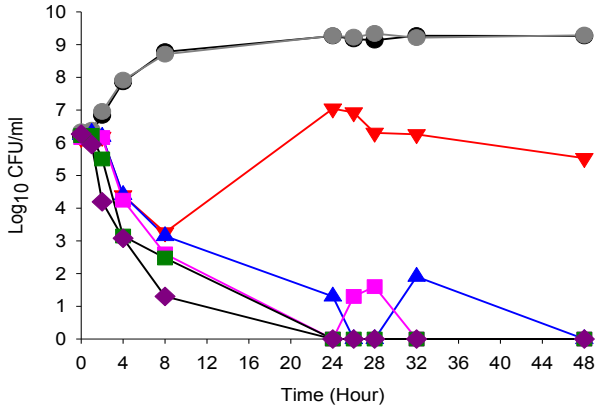
B. BLI 1 mg/L



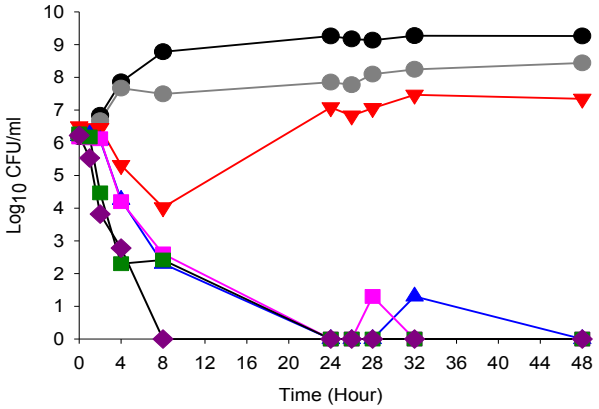
C. BLI 4 mg/L

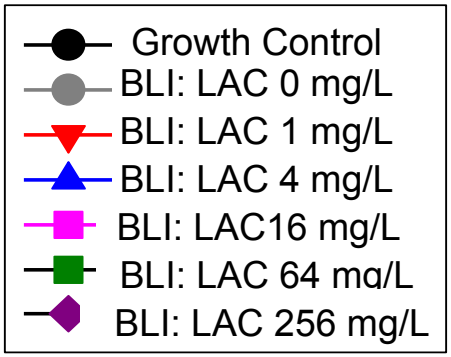


D. BLI 16 mg/L



E. BLI 64 mg/L



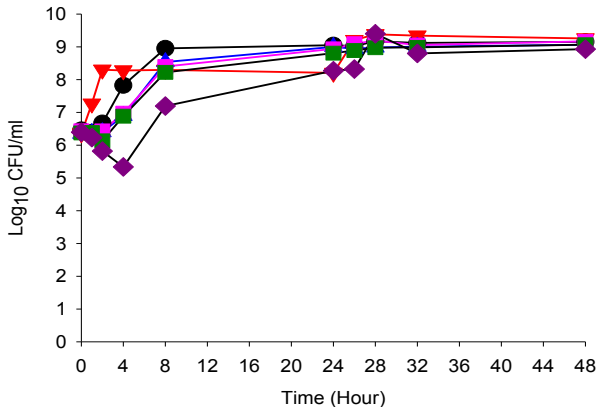


Results: Time-kill

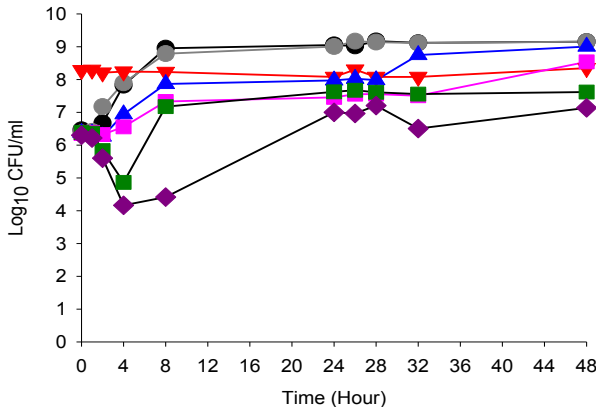
#LAPD4

(LAC MIC > 128 mg/L)

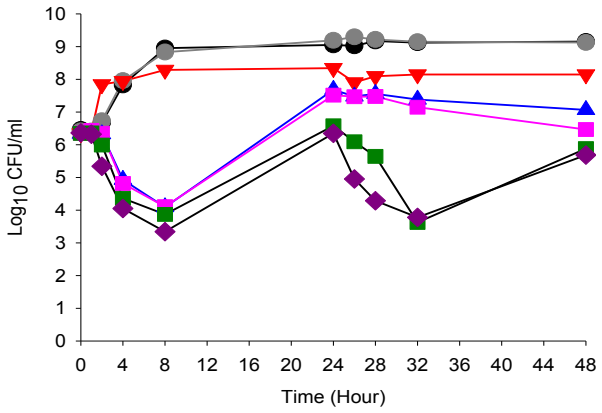
A. BLI 0 mg/L



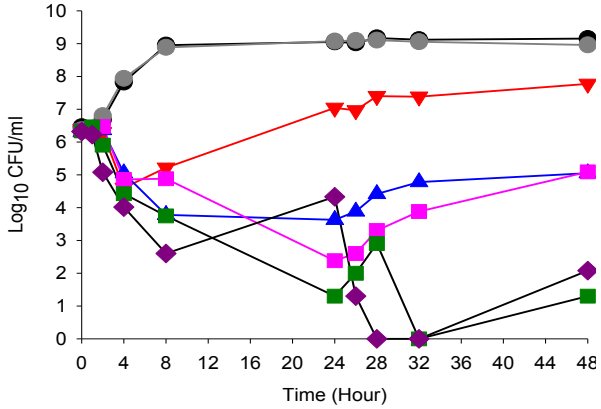
B. BLI 1 mg/L



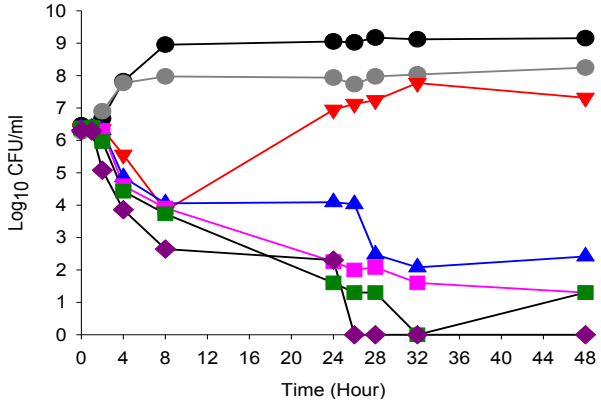
C. BLI 4 mg/L

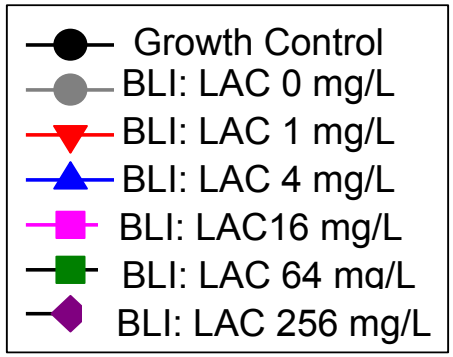


D. BLI 16 mg/L

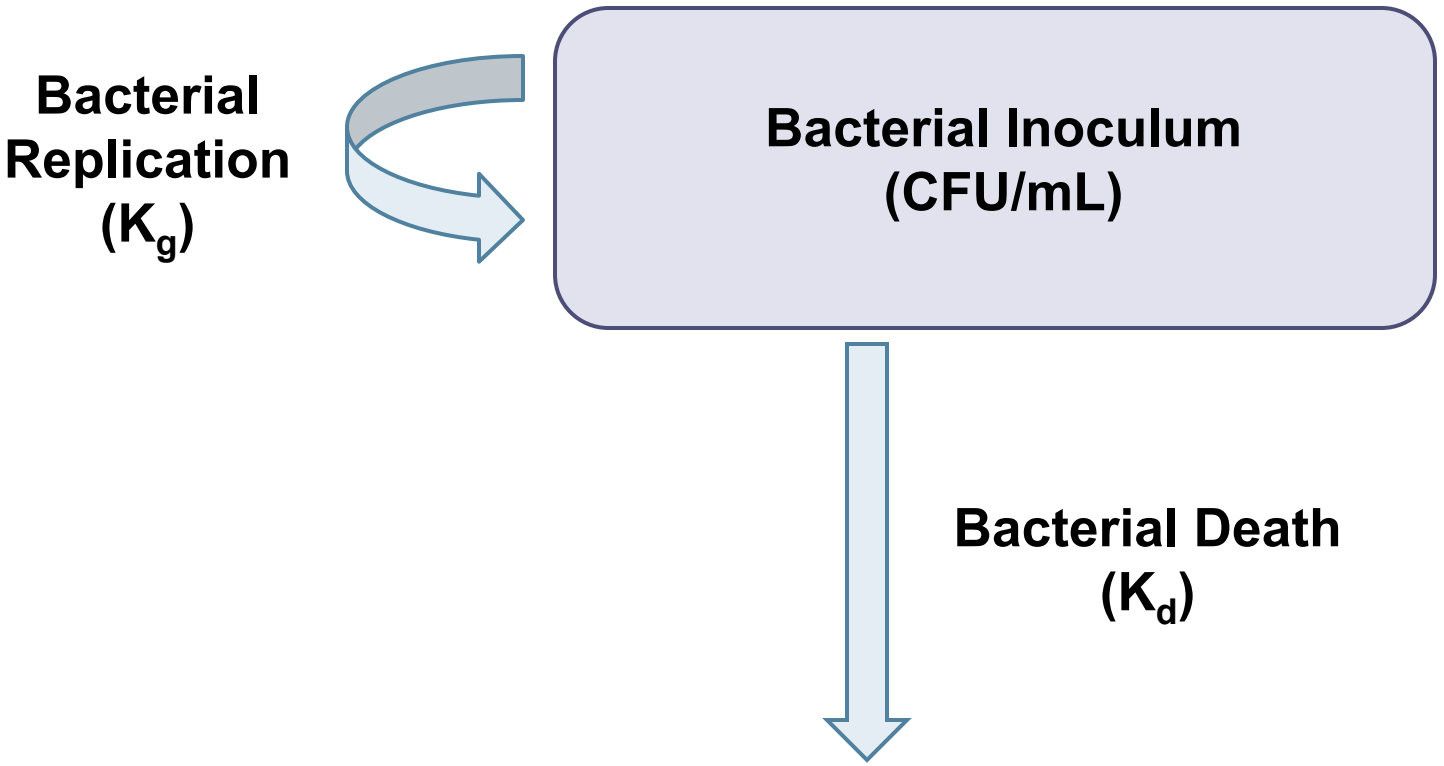


E. BLI 64 mg/L





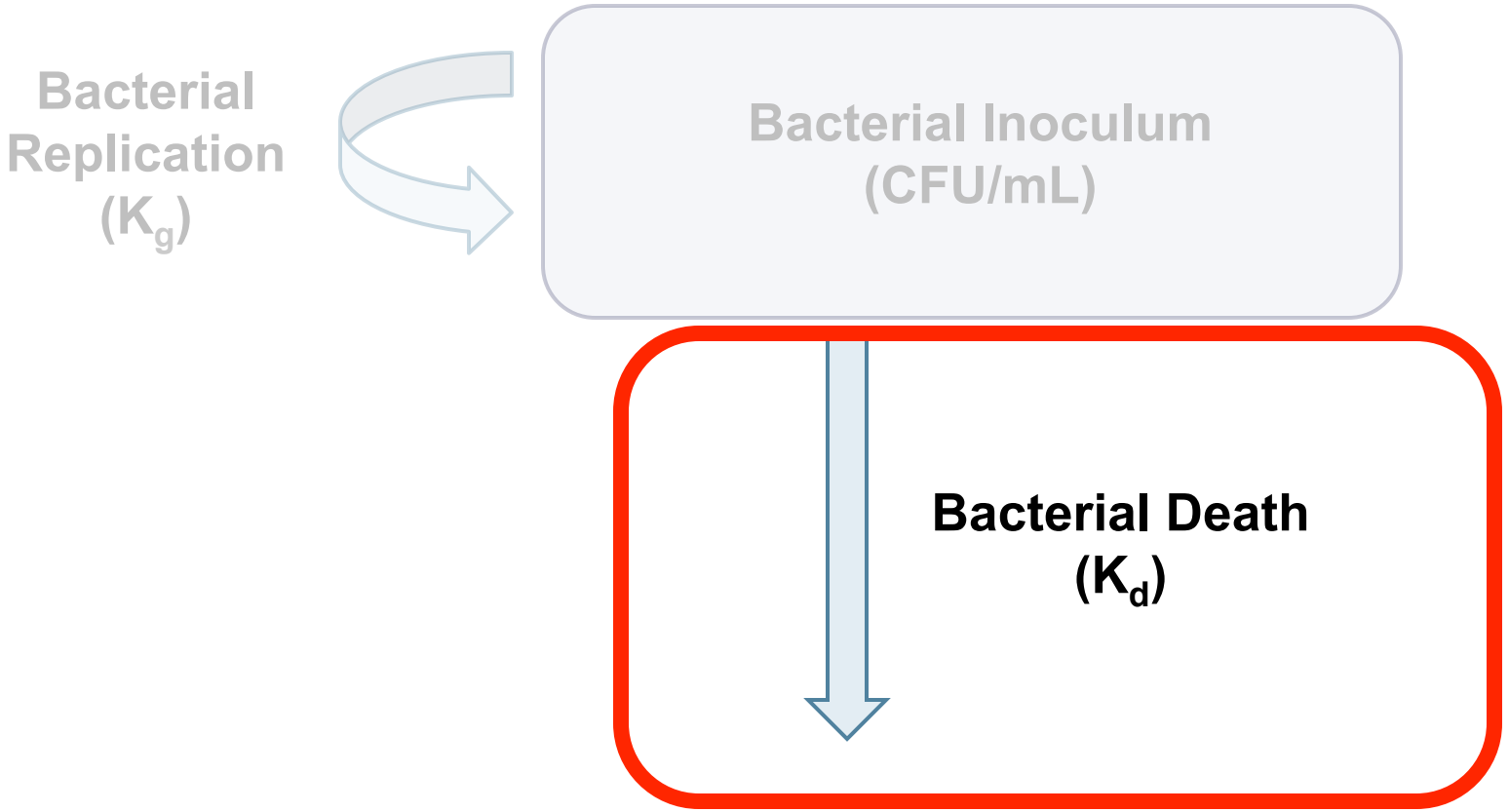
Results – PD Model



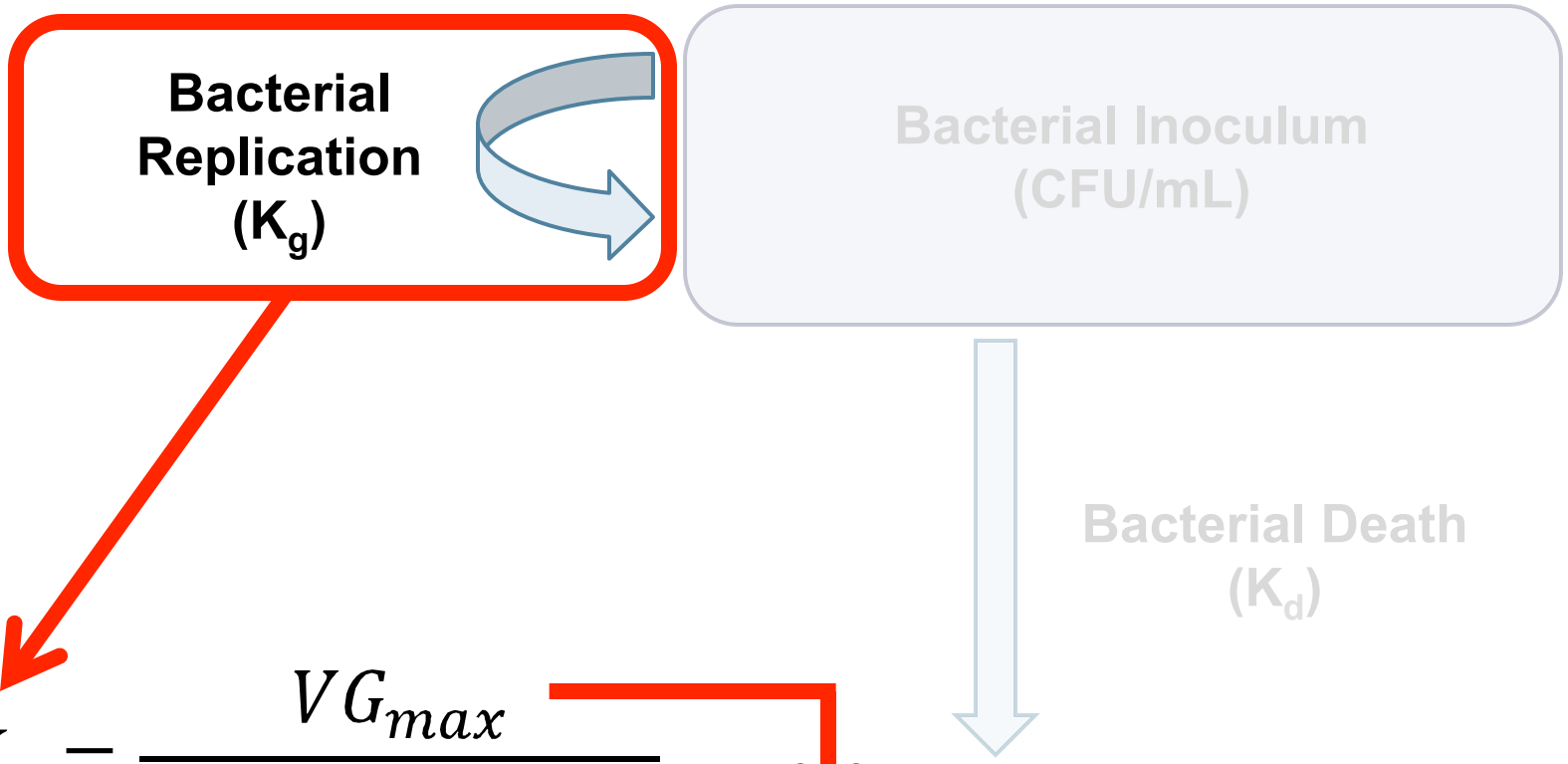
Meagher AK et al., Antimicrob Agents Chemother. **2004** Jun; 48(6):2061-8

Bulitta JB et al., Antimicrob Agents Chemother. **2009** Jan; 53(1):46-56

Results – PD Model



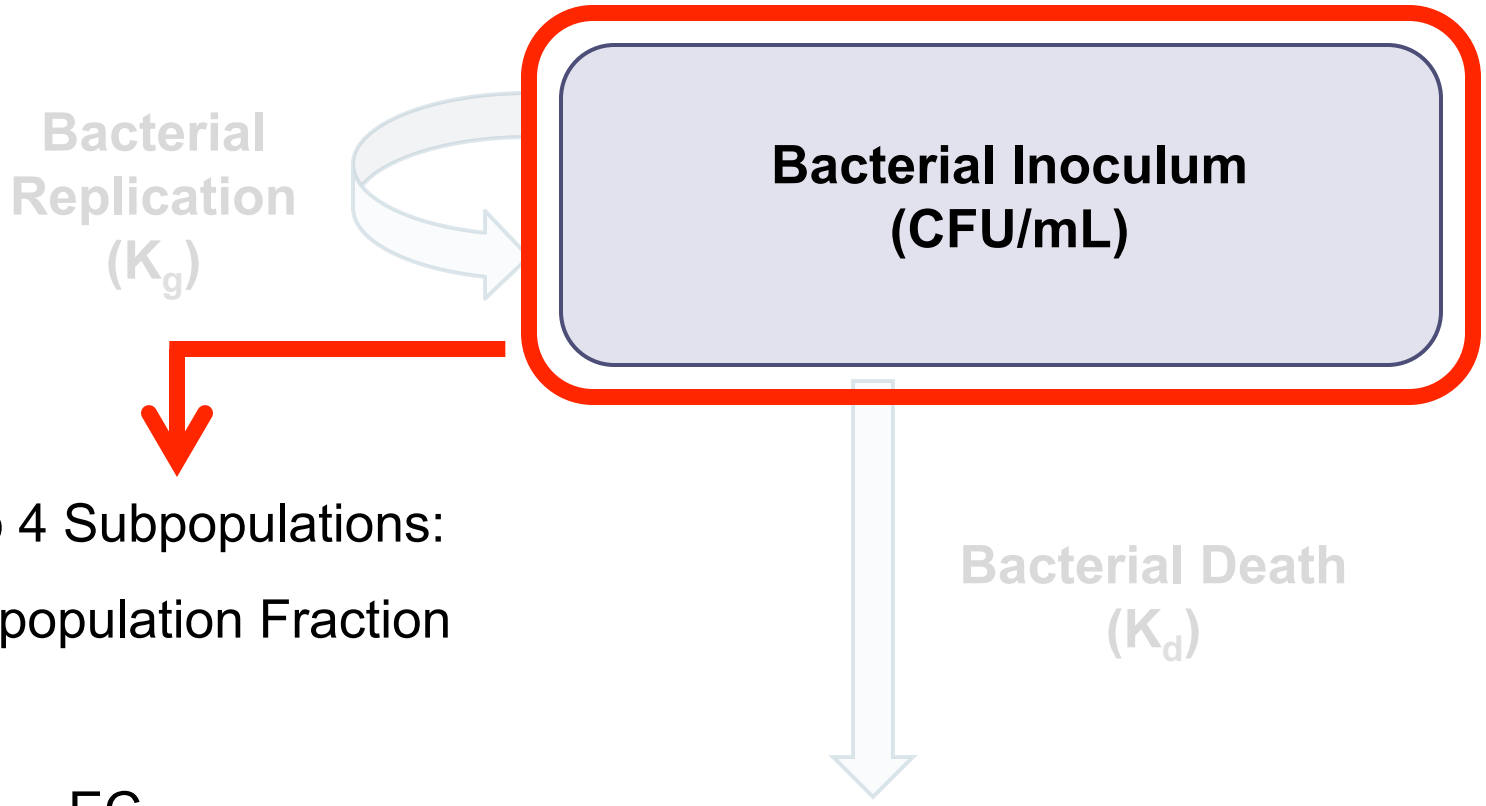
Results - PD Model



$$K_g = \frac{VG_{max}}{CFU_m + CFU_{tot}}$$

$$VG_{max} = K_d \times (CFU_m + CFU_{max})$$

Results – PD Model



Up to 4 Subpopulations:

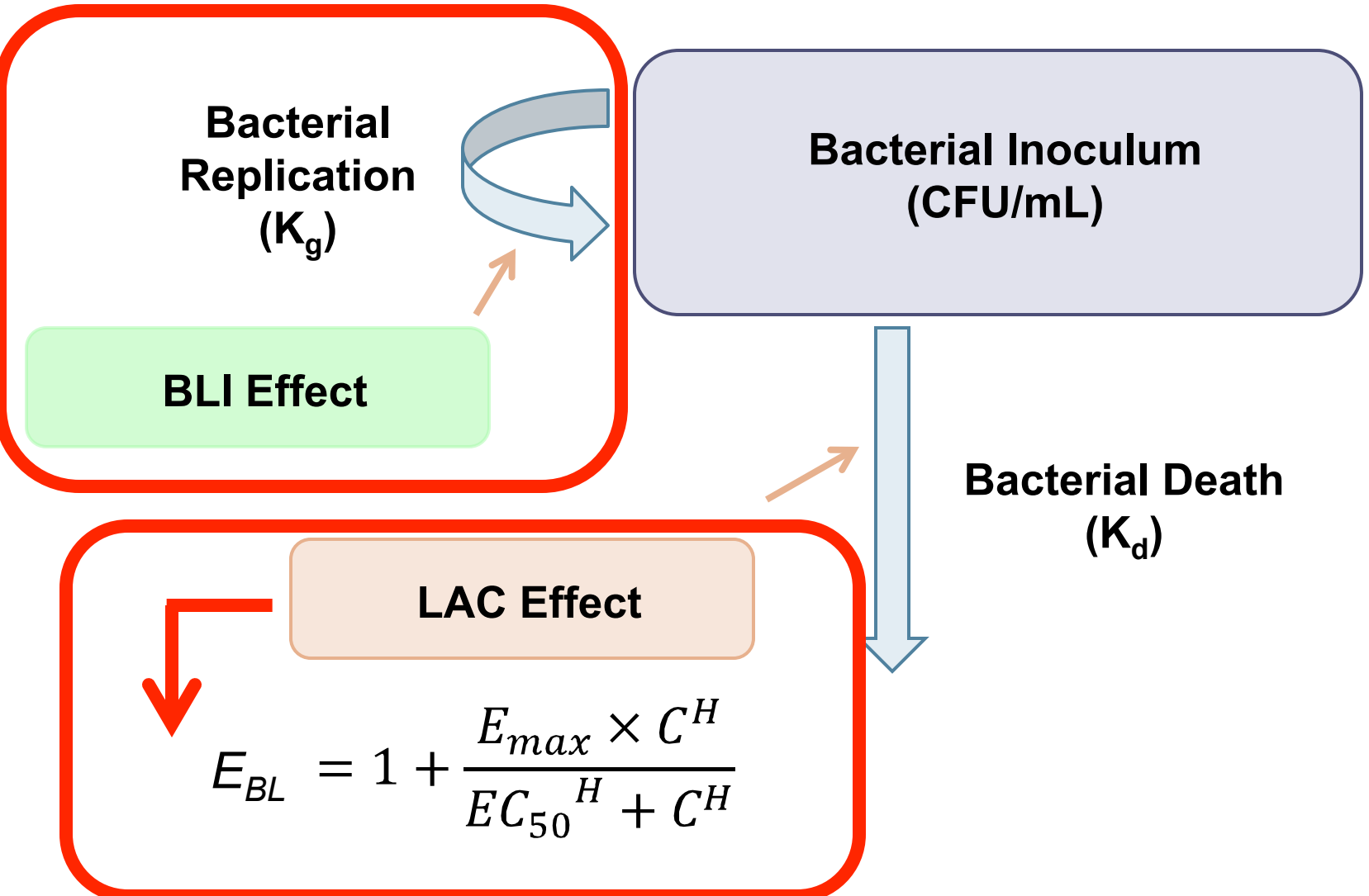
'Subpopulation Fraction (SF)'

EC_{50_1}

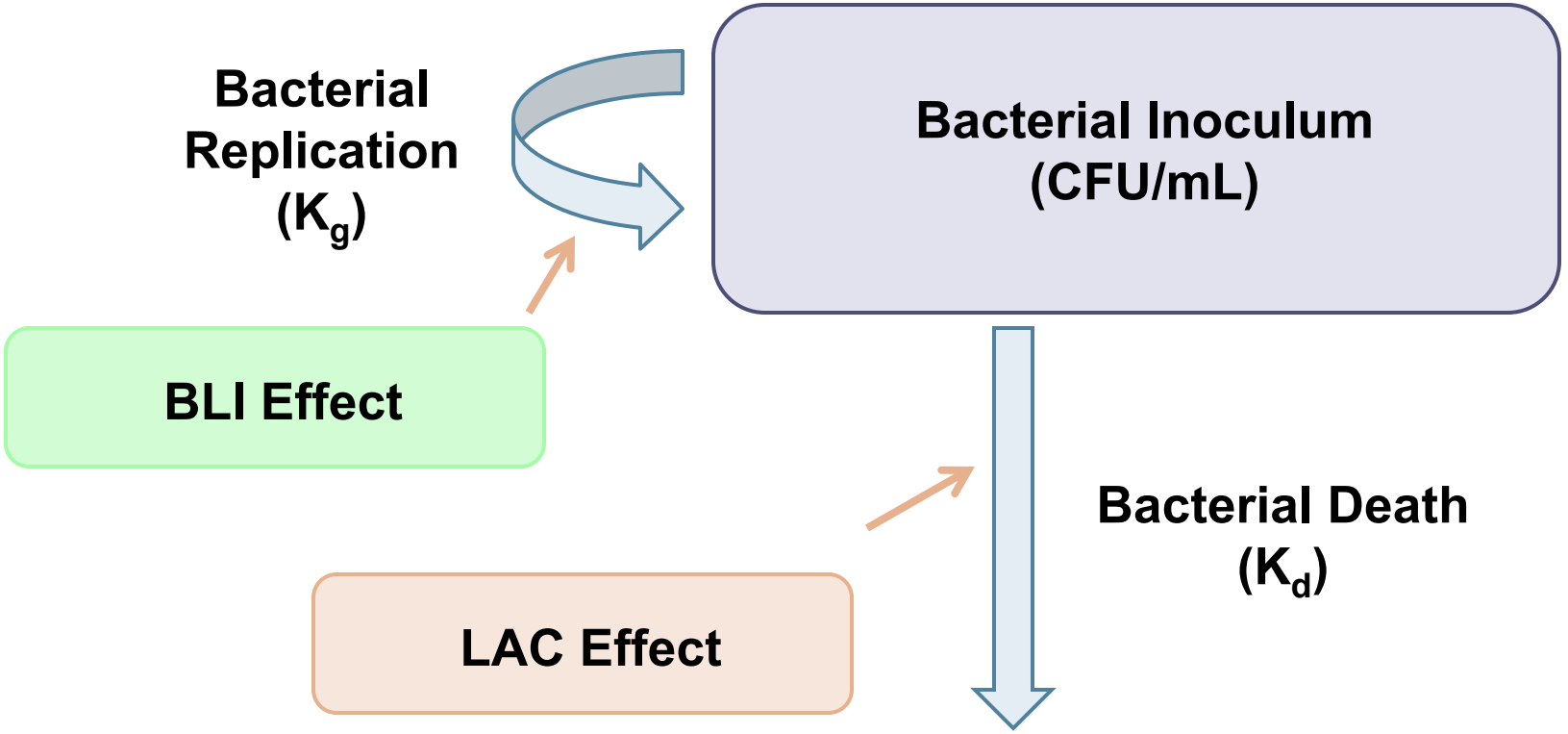
EC_{50_2}

EC_{50_3}

Results - PD Model



Results – PD Model



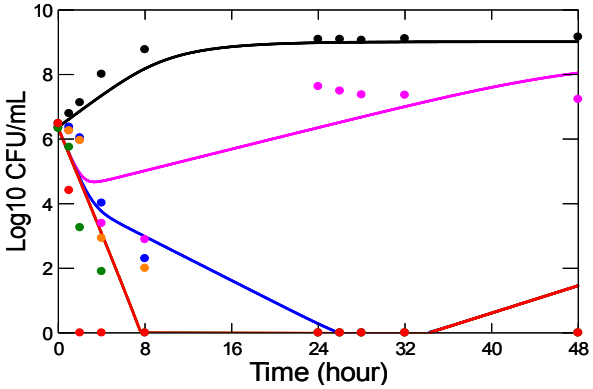
$$\frac{dB}{dt} = CFU_{subpopulation\ 1,2\ or\ 3} \times (K_g \times E_{BLI} - K_d \times E_{BL})$$

Results – PD Model

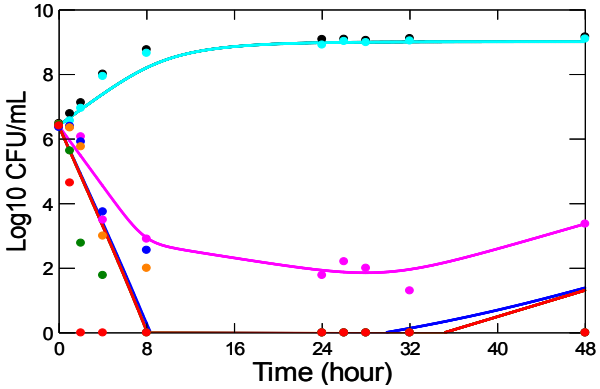
#LAPD1

(LAC MIC 0.25 mg/L)

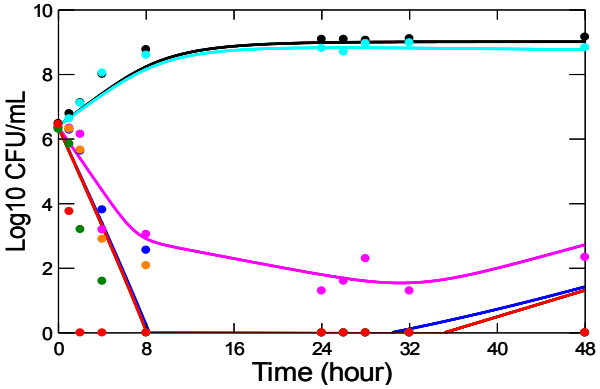
A. BLI 0 mg/L *(R=0.945)*



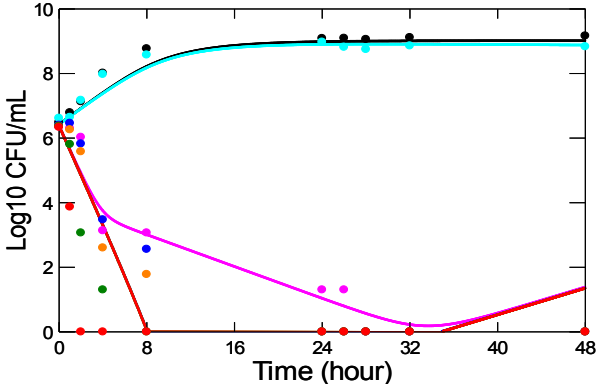
B. BLI 1 mg/L *(R=0.957)*



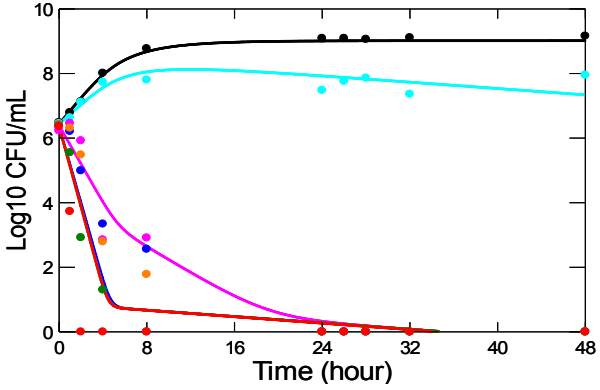
C. BLI 4 mg/L *(R=0.955)*



D. BLI 16 mg/L *(R=0.953)*



E. BLI 64 mg/L *(R=0.975)*



- Growth Control
- LAC 0 mg/L
- LAC 1 mg/L
- LAC 4 mg/L
- LAC 16 mg/L
- LAC 64 mg/L
- LAC 256 mg/L

Results – PD Model

#LAPD1
(LAC MIC 0.25 mg/L)

Parameter	BLI concentration (mg/L)				
	0	1	4	16	64
$Log_{10}CFU_0$ (CFU/mL)	6.35	6.40	6.40	6.40	6.40
$Log_{10}IC1$ (CFU/mL)	6.29	Bacterial Density			6.35
$Log_{10}IC2$ (CFU/mL)	0.0572				0.0466
$Log_{10}IC3$ (CFU/mL)	0.0240	1.13E-07	4.40E-08	9.91E-11	1.82E-05
$Log_{10}IC4$ (CFU/mL)	6.35E-10	6.40E-10	6.40E-10	6.40E-10	6.40E-10
$Log_{10}CFU_{max}$ (CFU/mL)	9.00	Replication Function			9.00
$Log_{10}CFU_m$ (CFU/mL)	8.72	8.72	8.70	8.71	8.49
$Log_{10}Vg_{max}$ (CFU/mL/h)	8.66	8.66	8.65	8.66	8.59
E_{max}	8.13	7.79	7.74	7.75	11.9
EC_{50_1}	0.0862	Pharmacodynamics			0.841
EC_{50_2}	6.70	2.55	2.20	1.87	2.74
EC_{50_3}	6.72	15.6	18.7	26.9	1.55E+04
E_{BLI}	1.000	1.000	0.972	0.940	0.811

Results – PD Model

#LAPD1
(LAC MIC 0.25 mg/L)

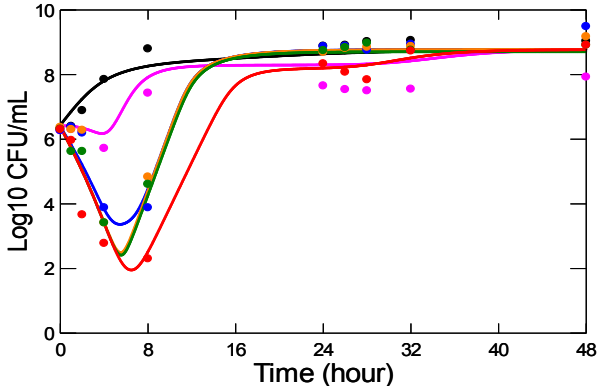
Parameter	BLI concentration (mg/L)				
	0	1	4	16	64
$Log_{10}CFU_0$ (CFU/mL)	6.35	6.40	6.40	6.40	6.40
$Log_{10}IC1$ (CFU/mL)	6.29	6.40	6.39	6.38	6.35
$Log_{10}IC2$ (CFU/mL)	0.0372	0.00373	5.65E-03	0.0248	0.0466
$Log_{10}IC3$ (CFU/mL)	0.0240	1.13E-07	4.40E-08	9.91E-11	1.82E-05
$Log_{10}IC4$ (CFU/mL)	6.35E-10	6.40E-10	6.40E-10	6.40E-10	6.40E-10
$Log_{10}CFU_{max}$ (CFU/mL)	9.00	9.00	9.00	9.00	9.00
$Log_{10}CFU_m$ (CFU/mL)	8.72	8.72	8.70	8.71	8.49
$Log_{10}Vg_{max}$ (CFU/mL/h)	8.66	8.66	8.65	8.66	8.59
E_{max}	8.13	7.79	7.74	7.75	11.9
EC_{50_1}	0.0862	0.659	0.606	0.148	0.841
EC_{50_2}	6.70	2.33	2.28	1.87	2.74
EC_{50_3}	6.72	15.6	18.7	26.9	1.55E+04
E_{BLI}	1.000	1.000	0.972	0.940	0.811

Results – PD Model

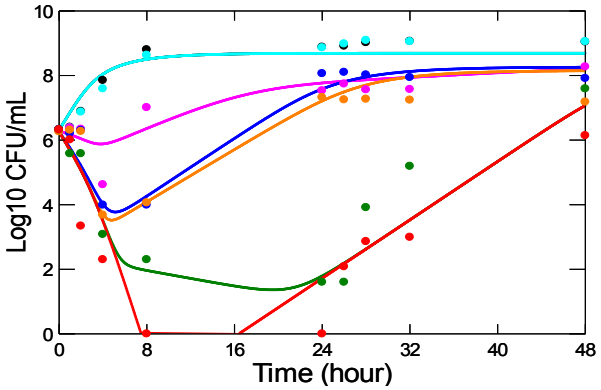
#LAPD2

(LAC MIC 4 mg/L)

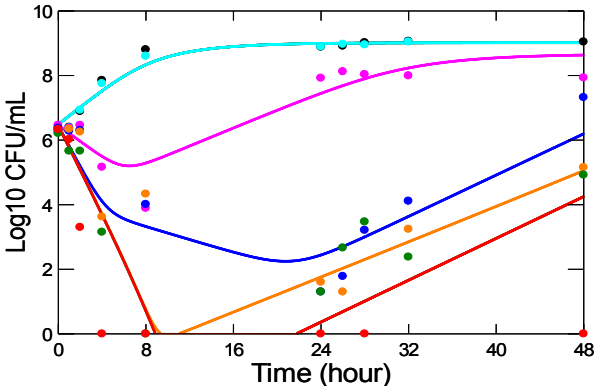
A. BLI 0 mg/L *(R=0.970)*



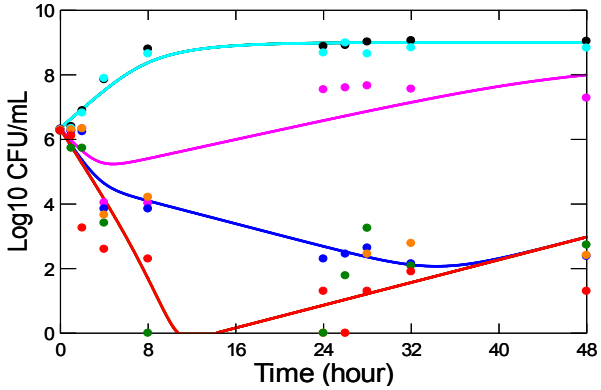
B. BLI 1 mg/L *(R=0.966)*



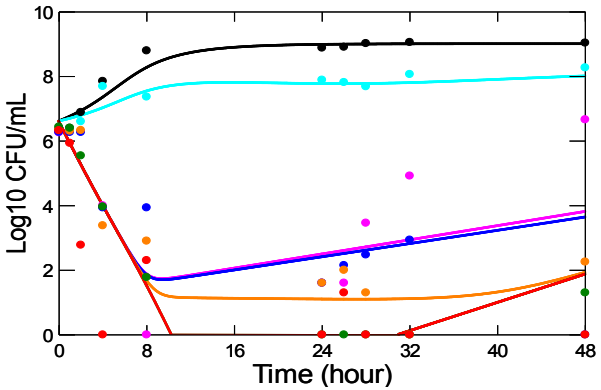
C. BLI 4 mg/L *(R=0.909)*



D. BLI 16 mg/L *(R=0.962)*



E. BLI 64 mg/L *(R=0.936)*



- Growth Control
- LAC 0 mg/L
- LAC 1 mg/L
- LAC 4 mg/L
- LAC 16 mg/L
- LAC 64 mg/L
- LAC 256 mg/L

Results – PD Model

#LAPD2
(LAC MIC 4 mg/L)

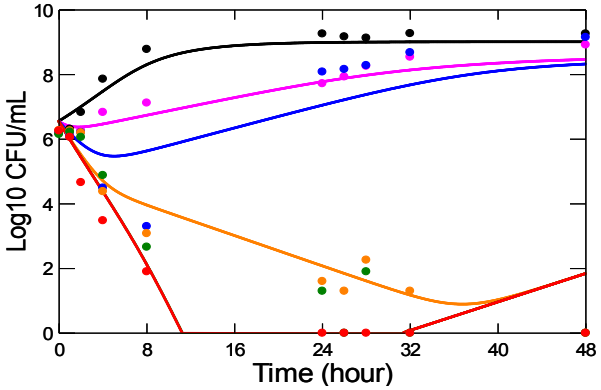
Parameter	BLI concentration (mg/L)				
	0	1	4	16	64
$Log_{10}CFU_0$ (CFU/mL)	6.40	6.25	6.45	6.35	6.50
$Log_{10}IC1$ (CFU/mL)	6.40	5.72	6.42	6.16	0.831
$Log_{10}IC2$ (CFU/mL)	1.35E-03	0.532	0.0293	0.185	5.67
$Log_{10}IC3$ (CFU/mL)	1.71E-09	9.27E-04	6.83E-08	1.17E-09	2.43E-05
$Log_{10}IC4$ (CFU/mL)	6.40E-10	6.25E-10	6.45E-10	1.64E-07	4.65E-08
$Log_{10}CFU_{max}$ (CFU/mL)	8.96	8.98	9.00	8.99	9.00
$Log_{10}CFU_m$ (CFU/mL)	7.90	8.02	8.69	8.62	8.56
$Log_{10}Vg_{max}$ (CFU/mL/h)	8.29	8.24	8.65	8.62	8.61
E_{max}	8.41	8.65	7.28	6.39	5.78
EC_{50_1}	1.21	0.909	0.761	0.469	0.0551
EC_{50_2}	2.20	3.57	6.44	7.69	0.346
EC_{50_3}	480	109	92.4	12.1	54.6
E_{BLI}	1.00	1.00	1.00	0.982	0.772

Results – PD Model

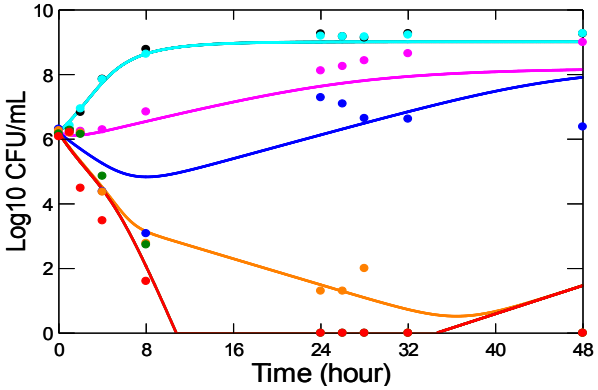
#LAPD3

(LAC MIC 8 mg/L)

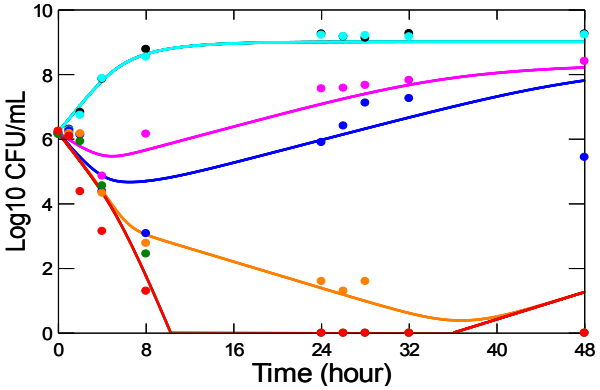
A. BLI 0 mg/L *(R=0.970)*



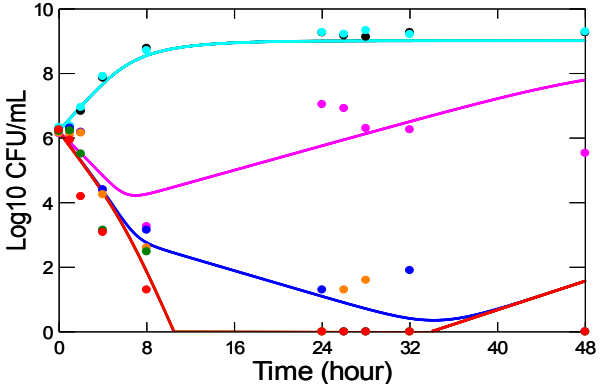
B. BLI 1 mg/L *(R=0.966)*



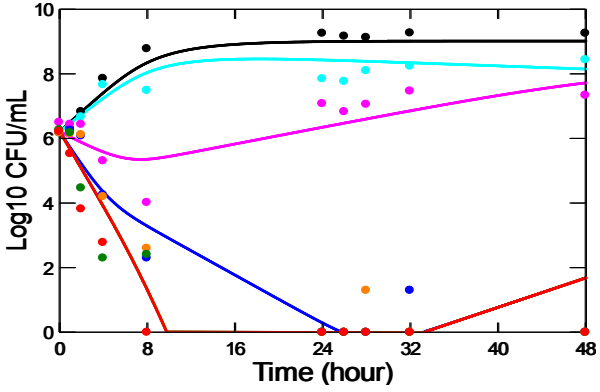
C. BLI 4 mg/L *(R=0.909)*



D. BLI 16 mg/L *(R=0.962)*



E. BLI 64 mg/L *(R=0.936)*



- Growth Control
- LAC 0 mg/L
- LAC 1 mg/L
- LAC 4 mg/L
- LAC 16 mg/L
- LAC 64 mg/L
- LAC 256 mg/L

Results – PD Model

#LAPD3
(LAC MIC 8 mg/L)

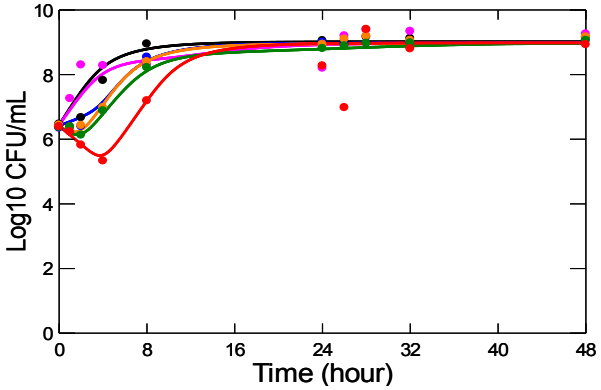
Parameter	BLI concentration (mg/L)				
	0	1	4	16	64
$Log_{10}CFU_0$ (CFU/mL)	6.20	6.20	6.20	6.20	6.20
$Log_{10}IC1$ (CFU/mL)	3.28E-05	3.14	5.87	6.19	5.97
$Log_{10}IC2$ (CFU/mL)	6.20	3.03	0.300	4.29E-11	1.90E-06
$Log_{10}IC3$ (CFU/mL)	7.99E-34	0.0314	0.0266	0.0118	0.228
$Log_{10}IC4$ (CFU/mL)	6.20E-10	6.20E-10	6.20E-10	6.20E-10	6.20E-10
$Log_{10}CFU_{max}$ (CFU/mL)	9.00	9.00	9.00	9.00	9.00
$Log_{10}CFU_m$ (CFU/mL)	8.59	8.43	8.48	8.53	8.62
$Log_{10}Vg_{max}$ (CFU/mL/h)	8.07	8.58	8.59	8.60	8.63
E_{max}	5.92	6.28	6.56	6.40	6.47
EC_{50_1}	1.20	0.0173	0.520	0.408	0.926
EC_{50_2}	1.79	5.21	7.26	0.850	1.16
EC_{50_3}	24.2	27.1	28.2	6.84	5.68
E_{BLI}	1.00	1.00	1.00	1.00	0.904

Results – PD Model

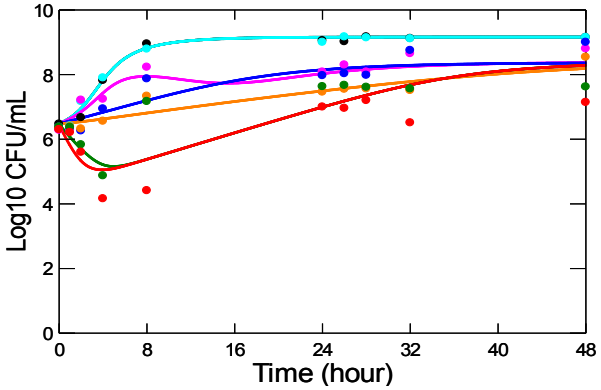
#LAPD4

(LAC MIC > 128 mg/L)

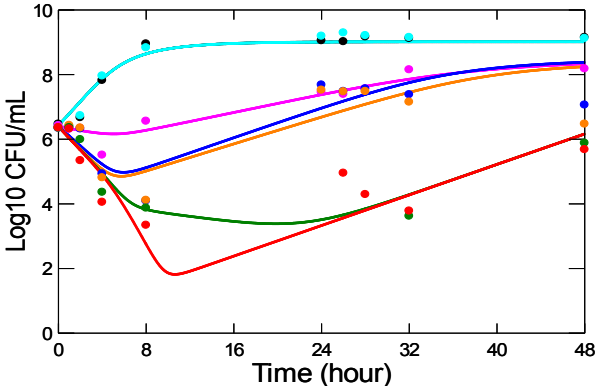
A. BLI 0 mg/L *(R=0.952)*



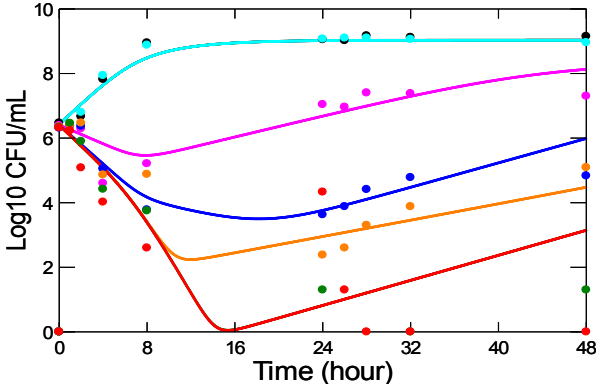
B. BLI 1 mg/L *(R=0.934)*



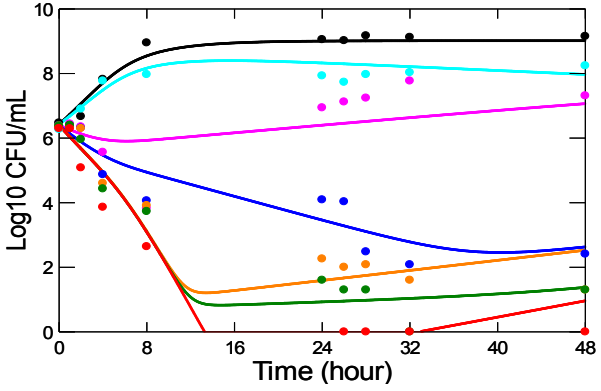
C. BLI 4 mg/L *(R=0.893)*



D. BLI 16 mg/L *(R=0.957)*



E. BLI 64 mg/L *(R=0.980)*



- Growth Control
- LAC 0 mg/L
- LAC 1 mg/L
- LAC 4 mg/L
- LAC 16 mg/L
- LAC 64 mg/L
- LAC 256 mg/L

Results – PD Model

#LAPD4

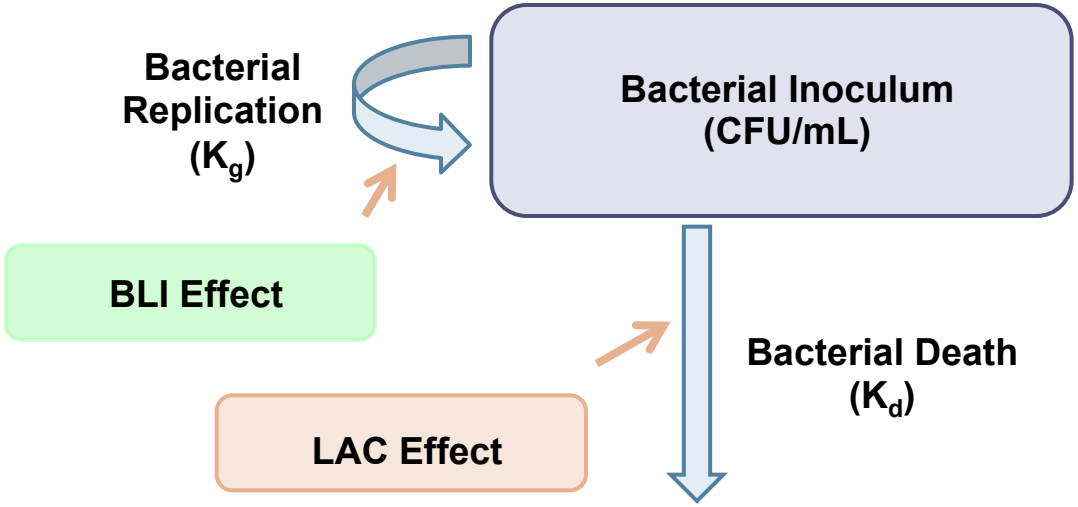
(LAC MIC > 128 mg/L)

Parameter	BLI concentration (mg/L)				
	0	1	4	16	64
$Log_{10}CFU_0$ (CFU/mL)	6.40	6.40	6.40	6.40	6.40
$Log_{10}IC1$ (CFU/mL)	0.0520	0.867	5.09	6.27	5.21
$Log_{10}IC2$ (CFU/mL)	6.05	5.43	1.28	0.130	1.19
$Log_{10}IC3$ (CFU/mL)	0.290	0.0264	0.0362	6.76E-05	1.12E-05
$Log_{10}IC4$ (CFU/mL)	3.57E-03	0.0793	7.59E-06	7.57E-08	2.07E-08
$Log_{10}CFU_{max_1}$ (CFU/mL)	9.00	9.00	9.00	9.00	9.00
$Log_{10}CFU_m$ (CFU/mL)	8.38	8.47	8.50	8.63	8.56
$Log_{10}Vg_{max_1}$ (CFU/mL/h)	8.57	8.59	8.60	8.63	8.61
E_{max}	6.36	6.08	5.65	4.86	4.85
EC_{50_1}	3.33	1.31	0.624	0.597	0.509
EC_{50_2}	3.77	23.6	3.61	6.36	7.70
EC_{50_3}	193	23.7	119	66.4	284
E_{BLI}	1.00	1.00	1.00	1.00	0.970

Summary

- First PD model developed for a ß-lactam/ß-lactamase combination
- Most data sets fit with two subpopulations differing in the degree of susceptibility to LAC (EC_{50})
- Intrinsic effect of BLI was modest
- Combination:
  - Reduced EC_{50} to LAC in comparison to monotherapy
  - Eliminated completely resistant subpopulations

Conclusion



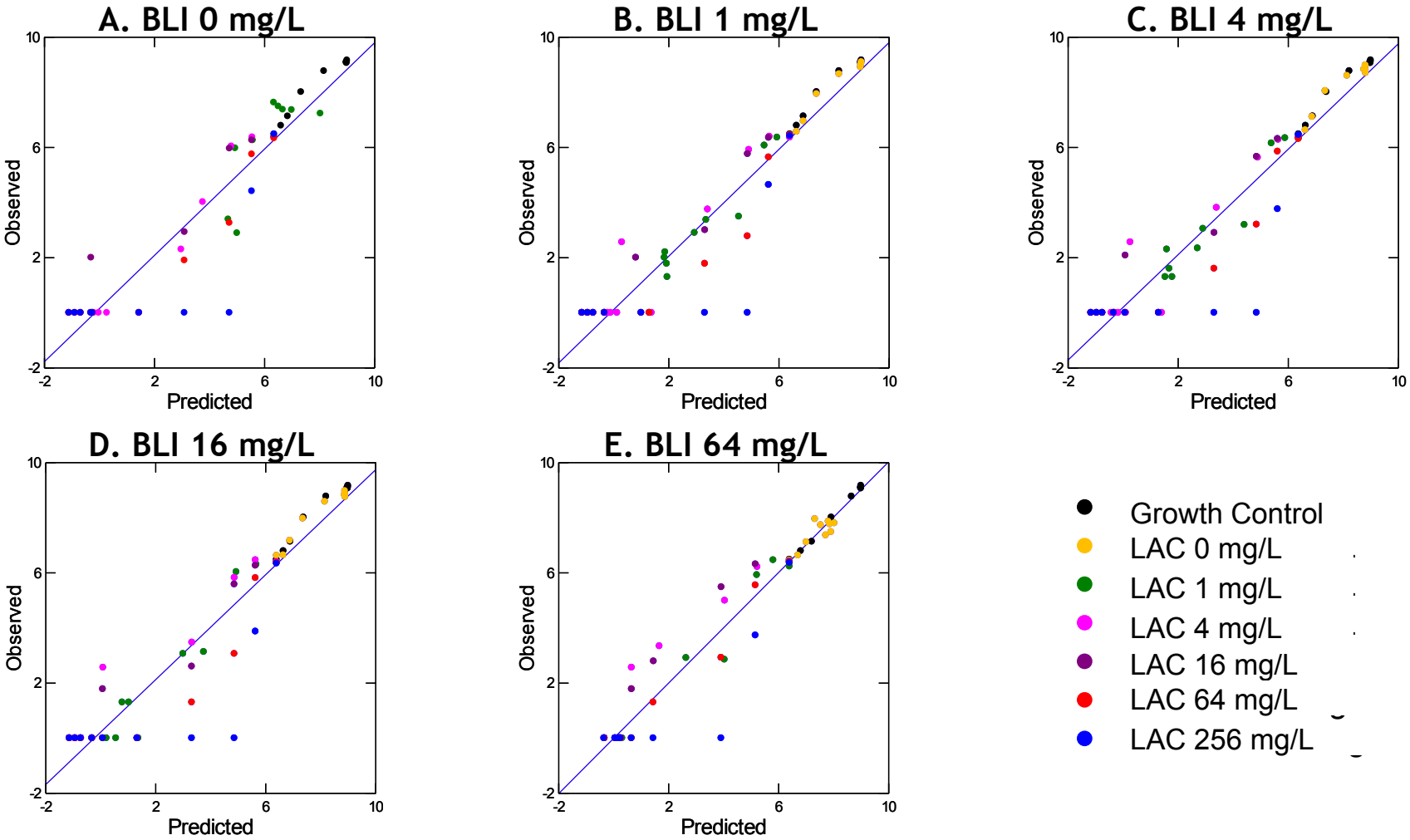
$$\frac{dB}{dt} = CFU_{subpopulation\ 1,2\ or\ 3} \times (K_g \times E_{BLI} - K_d \times E_{BL})$$

- BL and BLI ≥ 4 mg/L demonstrates excellent activity against ß-lactamase producing strains of *E. coli*

Observed *versus* Predicted

#LAPD1

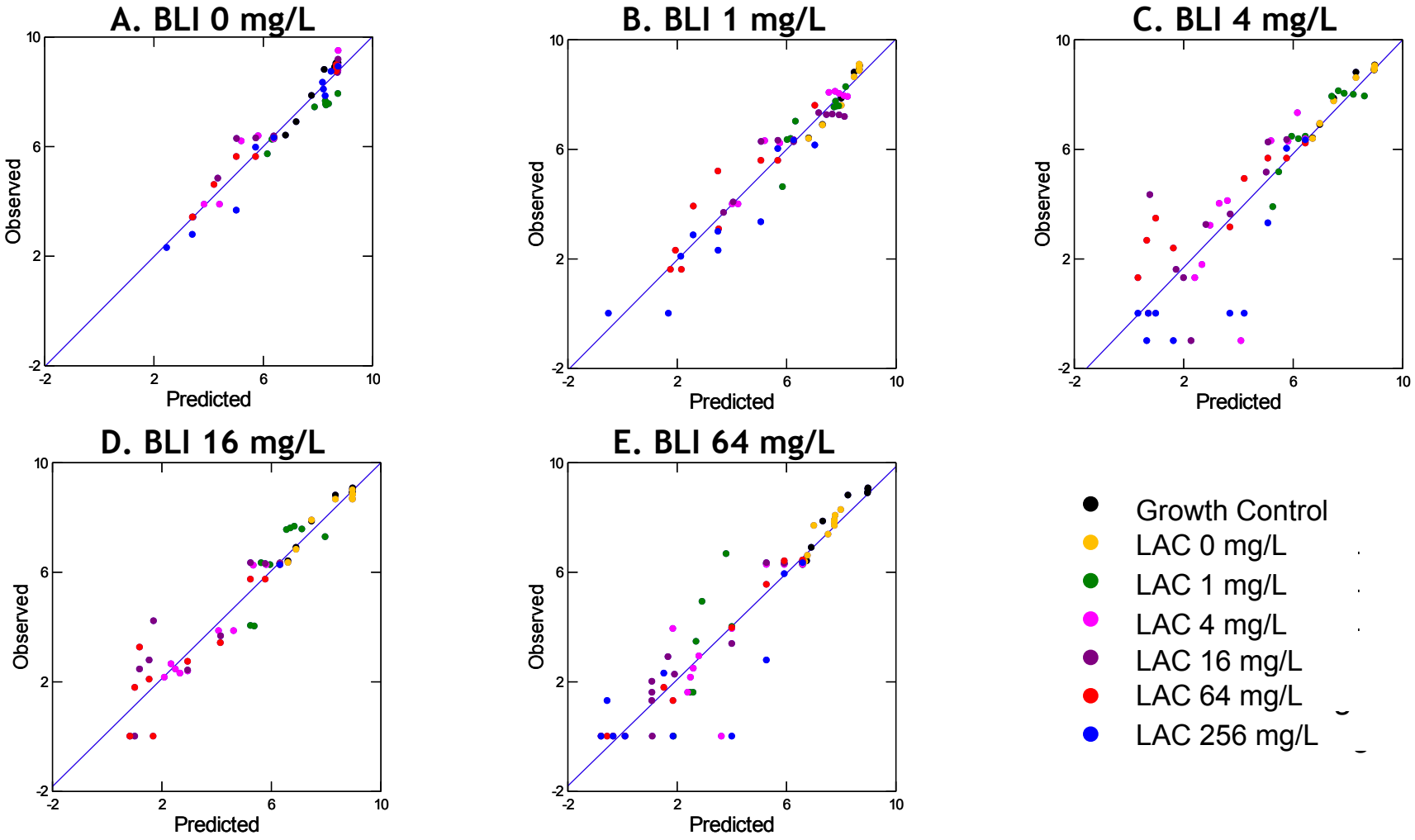
(LAC MIC 0.25 mg/L)



Observed *versus* Predicted

#LAPD2

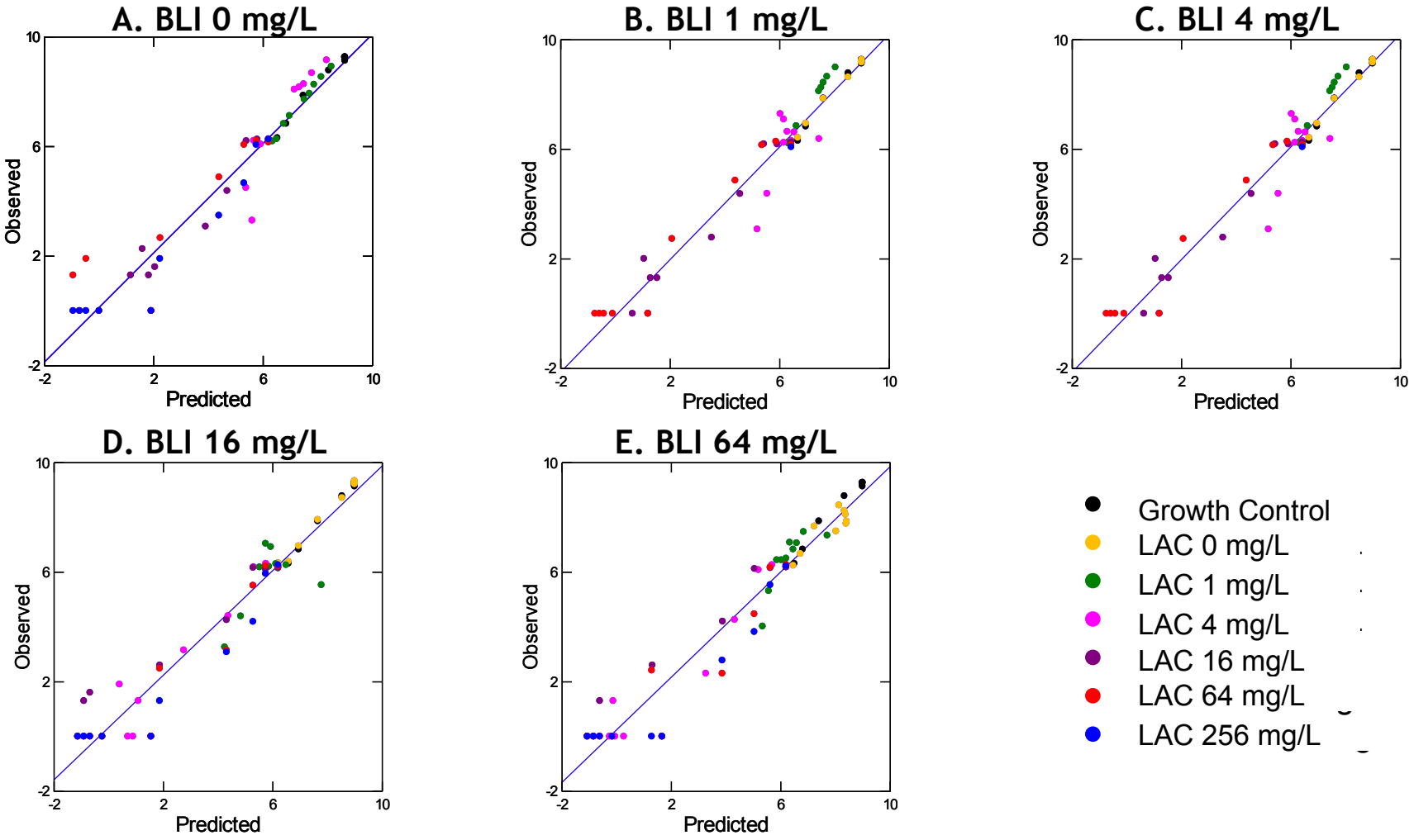
(LAC MIC 4 mg/L)



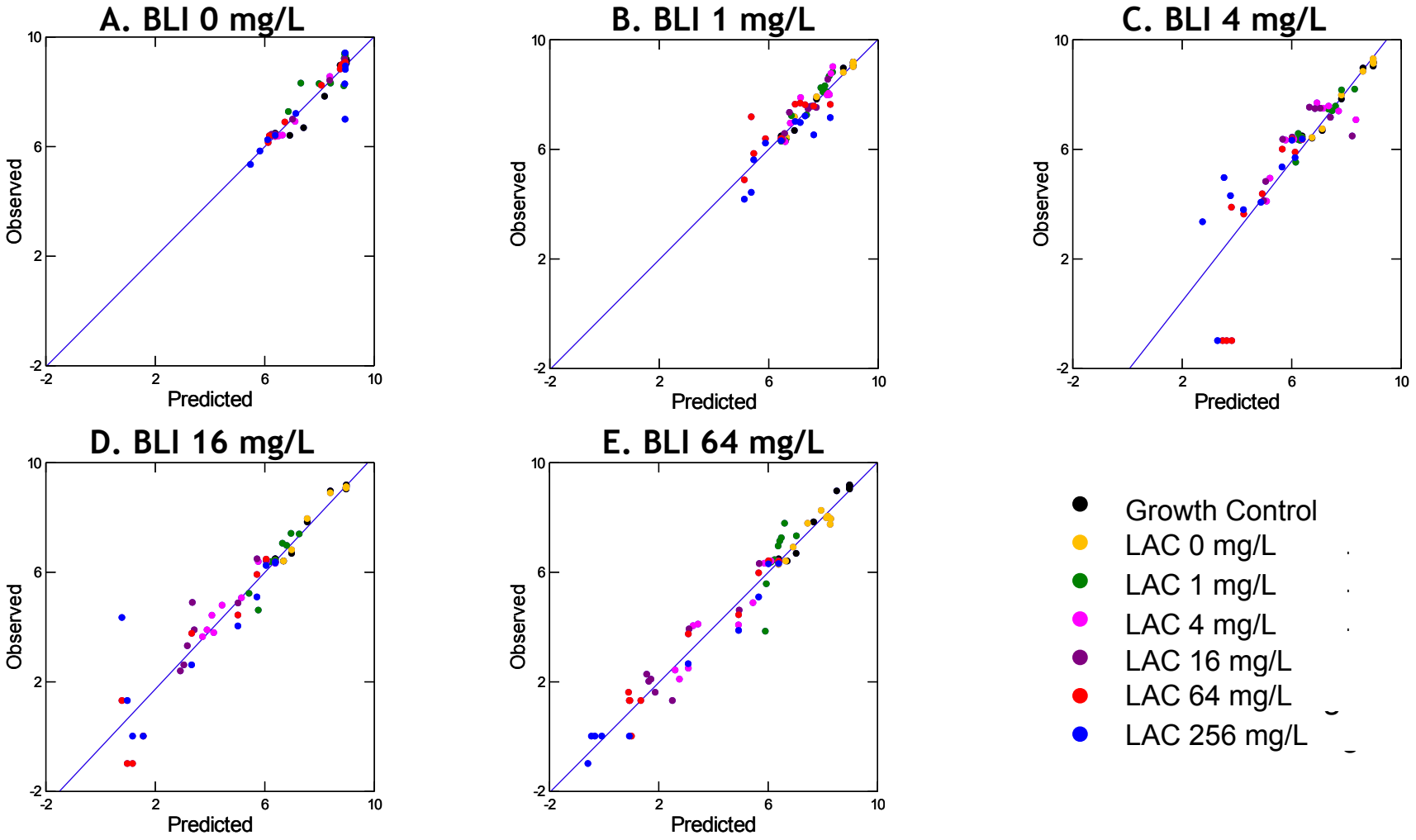
Observed *versus* Predicted

#LAPD3

(LAC MIC 8 mg/L)



Observed *versus* Predicted

#LAPD4

(LAC MIC > 128 mg/L)