

Figure S1. SDS-PAGE analysis of purified glutamate racemases from *B. subtilis* (\*), *B. anthracis* (\*), and *F. tularensis* (\*\*). Two isozymes exist in the case of *B. anthracis*. Molecular weight of glutamate racemases from *B. subtilis* and *B. anthracis* is ~31,500 and *F. tularensis* is ~29,300.

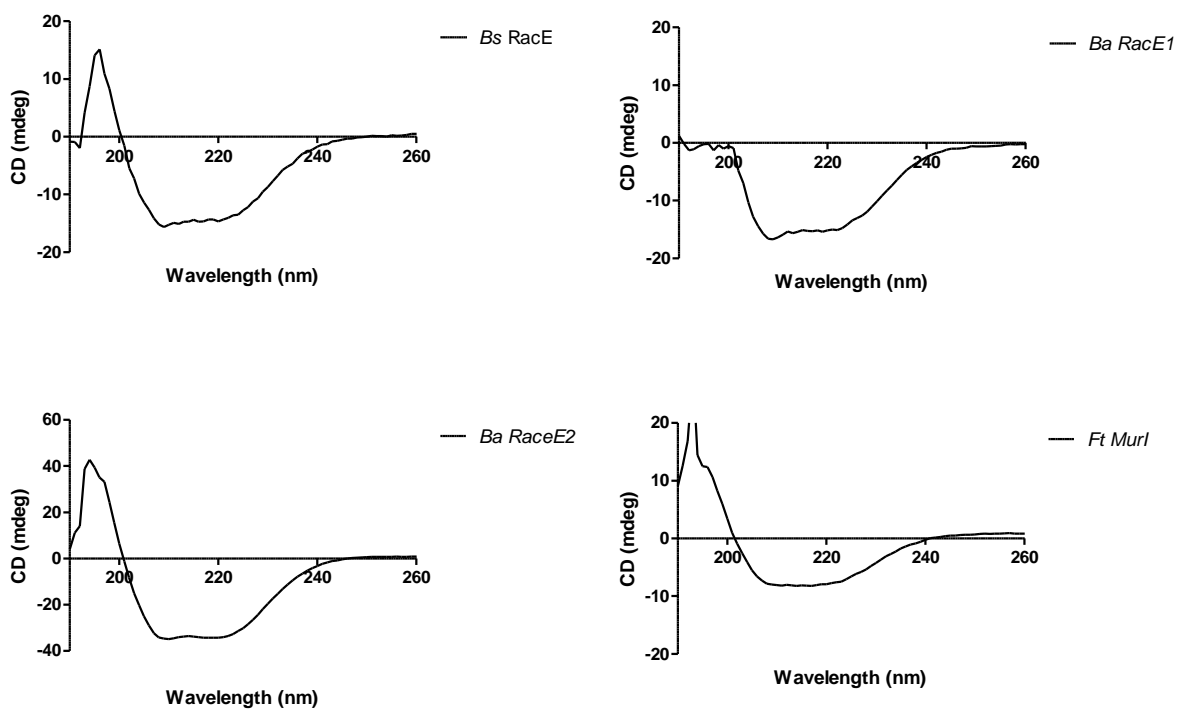


Figure S2. Circular dichroism of purified proteins to assess protein foldedness.

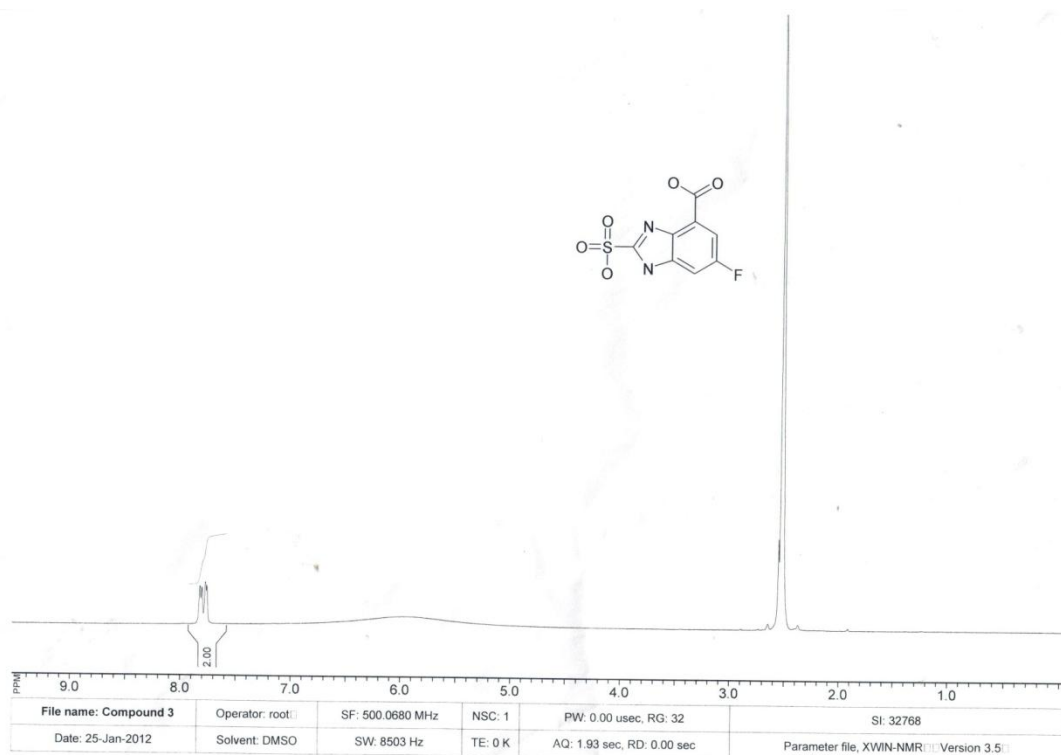
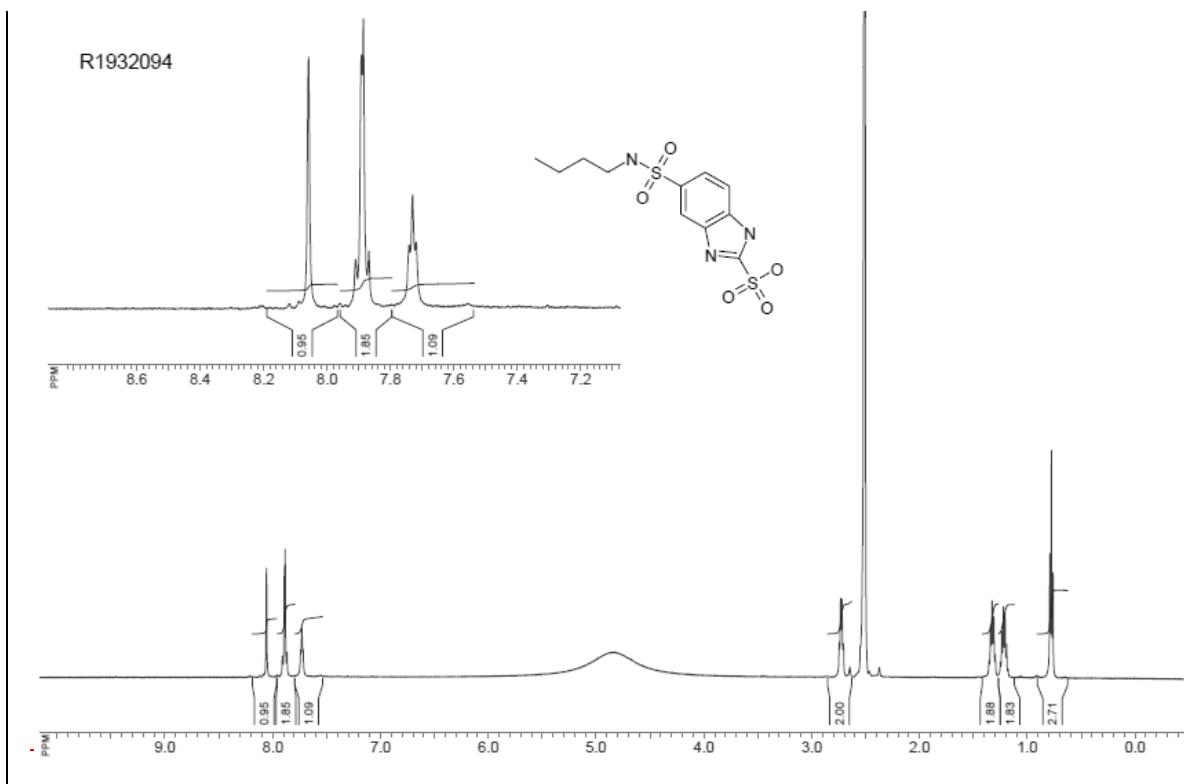


Figure S3. <sup>1</sup>H NMR spectra for compound **4**. Experiment conducted at ambient temperature. Peak at 2.50 corresponds to the employed solvent, DMSO.



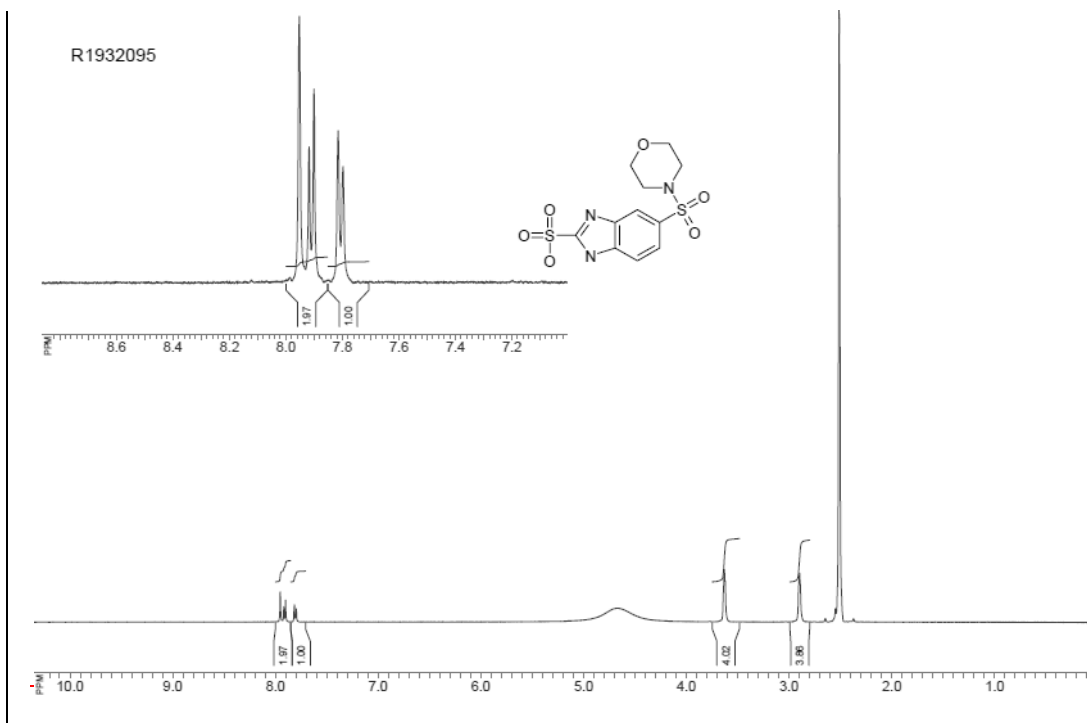


Figure S5.  $^1\text{H}$  NMR spectra for compounds **29**. Experiment conducted at ambient temperature. Peak at 2.50 corresponds to the employed solvent, DMSO.

MaxPeak: 98.14%  
Ret\_Time: 0.399 min

### Compound 3



#	Time	Area%
1	0.399	98.14
2	0.654	1.86

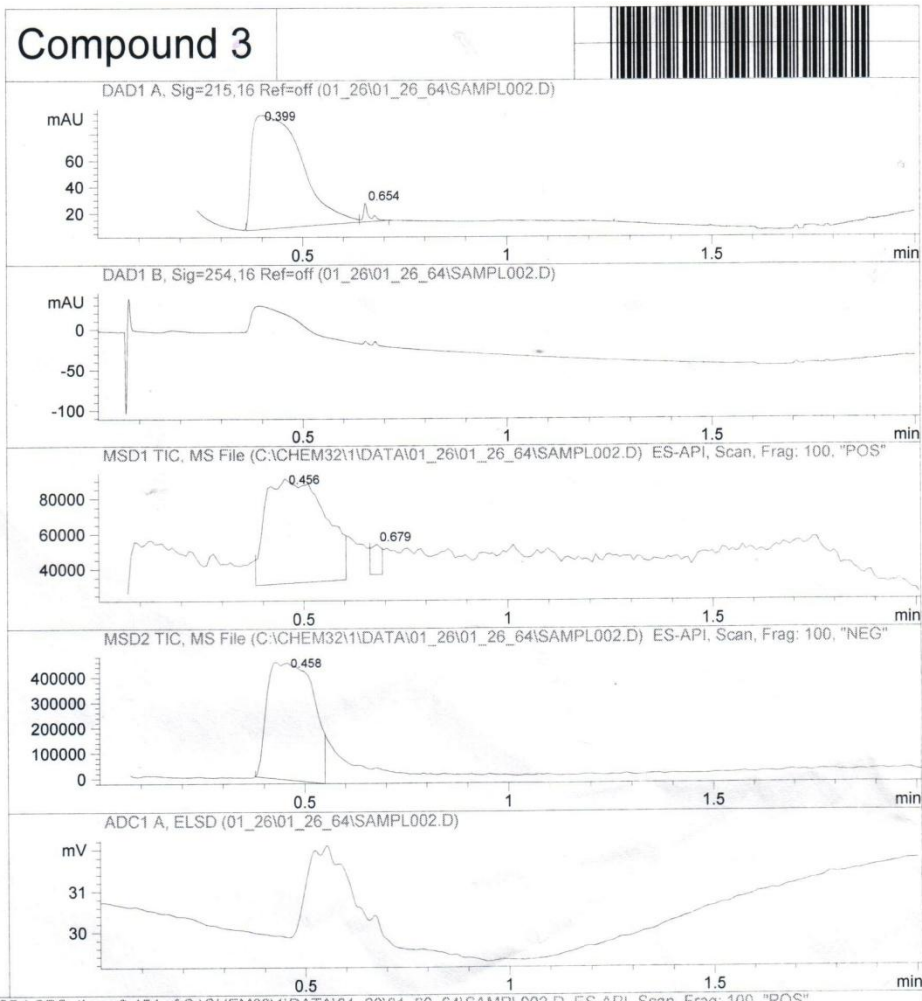


Figure S6. HPLC analysis of compound 4, showing 98.1% purity.

MaxPeak: 99.03%  
Ret\_Time: 0.645 min

# Compound 17



#	Time	Area%
1	0.645	99.03
2	0.752	0.97

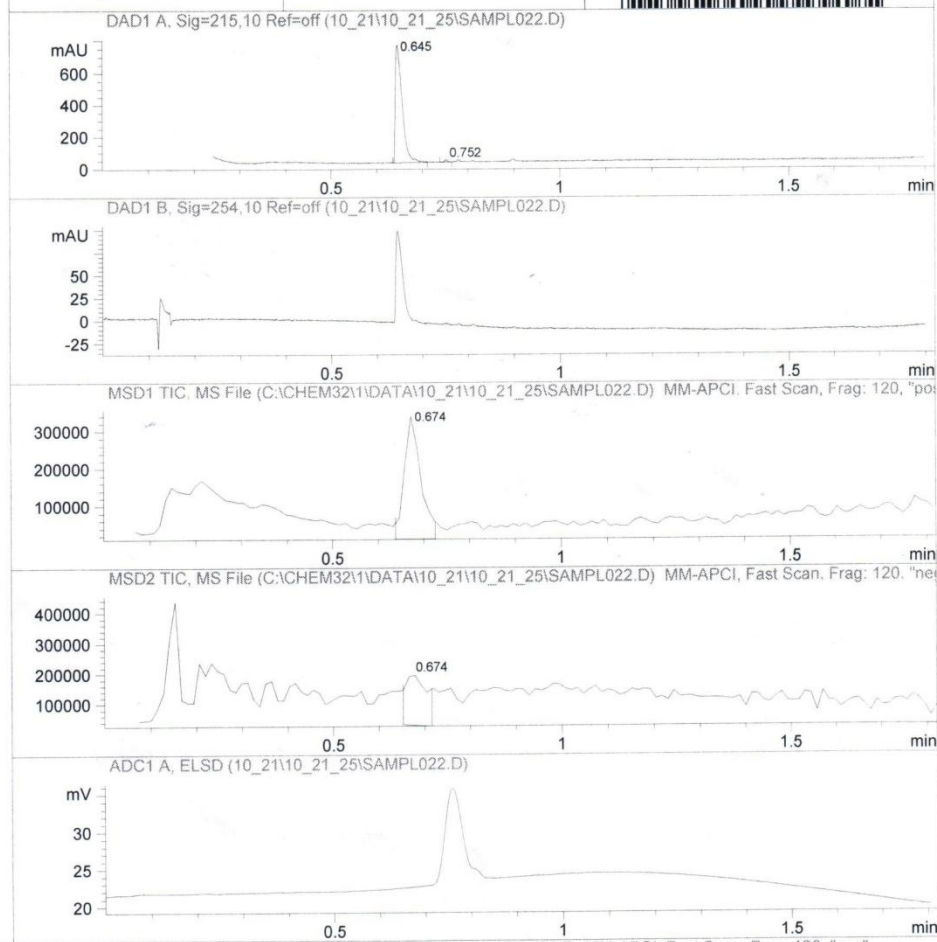


Figure S7. HPLC analysis of compound **18**, showing 99.0% purity.

MaxPeak: 100.00%  
Ret\_Time: 0.475 min

# Compound 29



#	Time	Area%
1	0.475	100.00

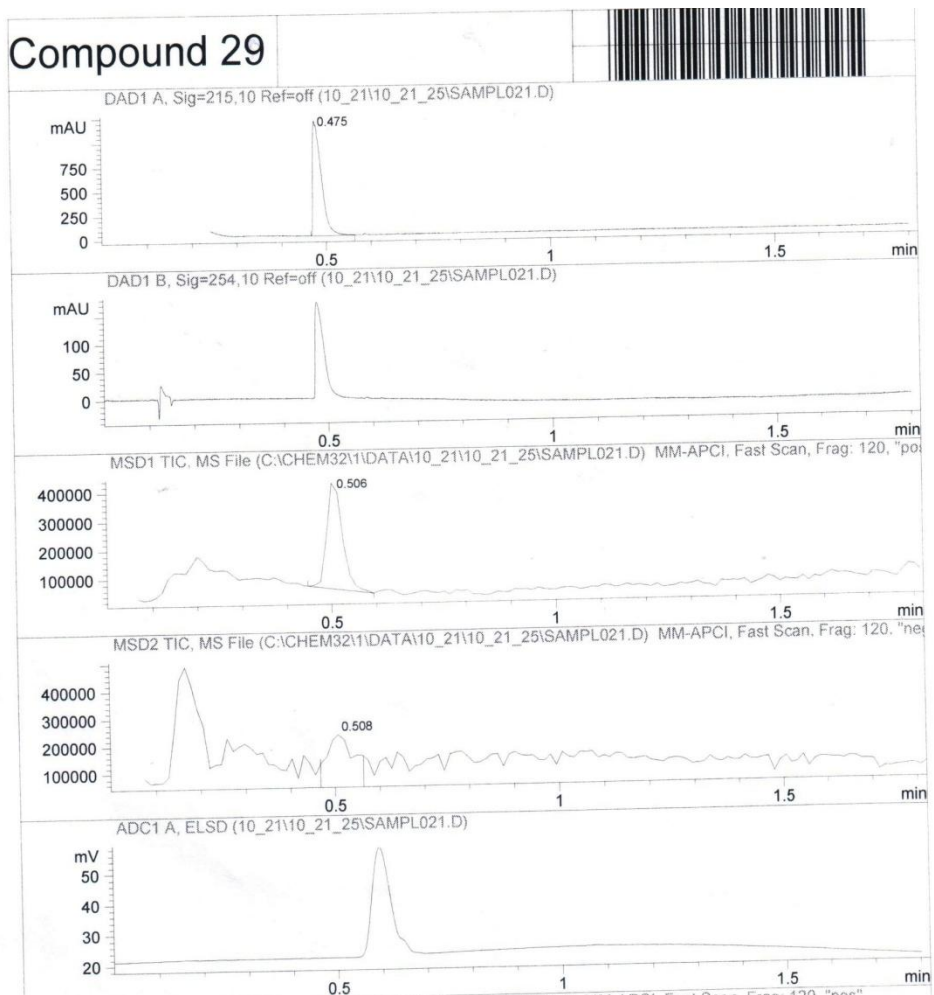


Figure S8. HPLC analysis of compound **29**, showing 100.0% purity.



Inhibitor X with 10mM and 0.1mM Stocks								
Media (uL)	Compound (uM)	Compound (uL)	[Stock] uM	Inoculum (uL)	PBS (uL)	Final Vol	Column	
100	0	0	100	20	80	200	1	
100	0.1	0.2	100	20	79.8	200	2	
100	1	2	100	20	78	200	3	
100	10	20	100	20	60	200	4	
100	50	1	10000	20	79	200	5	
100	100	2	10000	20	78	200	6	
100	500	10	10000	20	70	200	7	
100	1000	20	10000	20	60	200	8	
100	1500	30	10000	20	50	200	9	
100	2000	40	10000	20	40	200	10	
100	2500	50	10000	20	30	200	11	
100	3000	60	10000	20	20	200	12	

Column	1	2	3	4	5	6	7	8	9	10	11	12
Row A	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1
Row B	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1
Row C	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1	Inhibitor 1
Row D	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank
Row E	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank
Row F	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2
Row G	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2
Row H	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2	Inhibitor 2

Figure S9. Example of MIC50 reagent volumes for a single replicate of one inhibitor. Inhibitor concentration varies along the X-axis of the plate. Two compounds could be assayed in triplicate per 96-well plate of bacteria. Blank wells contain 100  $\mu$ L of phosphate-buffered saline (PBS) and 100  $\mu$ L of 2X media.

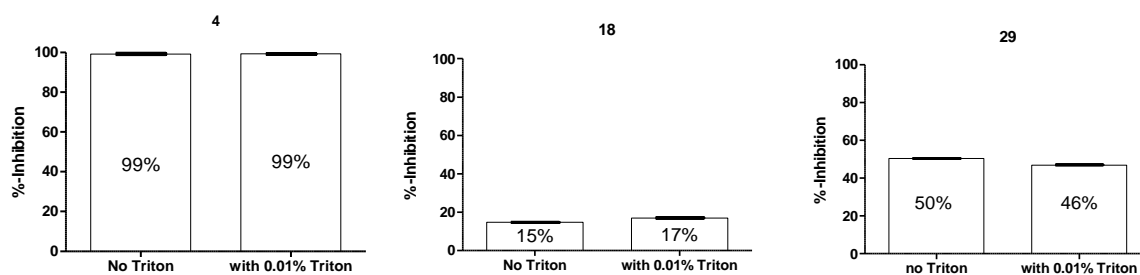


Figure S10. Colloidal aggregate testing for newly synthesized derivatives of 1*H*-benzimidazole-2-sulfonic acid. Racemase activity tested in the presence of a fixed concentration of inhibitor (200  $\mu$ M), 1 mM D-glutamate, and with or without 0.01% Triton X-100 included in the working buffer. %-Inhibition determined in triplicate with standard deviation shown (error bars).

### BISA Derivative MIC<sub>50</sub> vs *Lactococcus lactis*

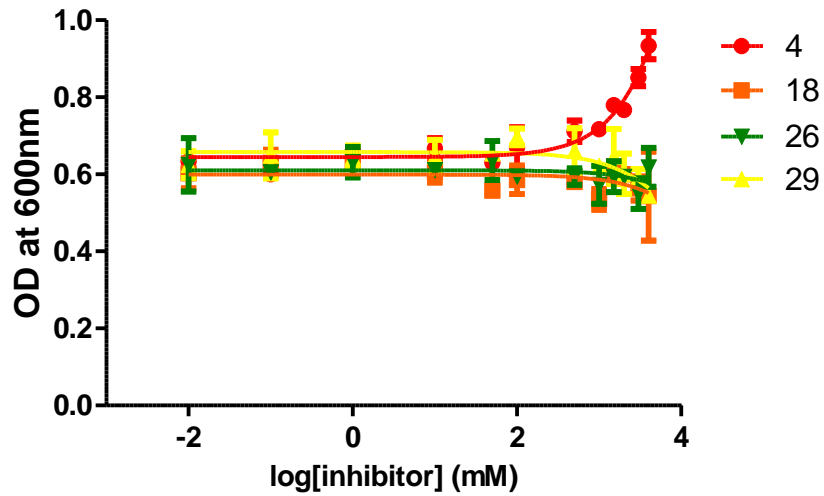


Figure S11. MIC<sub>50</sub> analysis of select derivatives of 1*H*-benzimidazole-2-sulfonic acid against *Lactococcus lactis*. Compounds **18** and **26** show no growth inhibition within the tested range of inhibitor concentrations. Compound **29** starts to show growth inhibition at the highest tested concentration. Surprisingly, compound **4** (a potent growth inhibitor against *B. subtilis*) causes noticeable growth activation compared to untreated cells. This is an unexpected phenomenon, which may be the result of productive metabolism of the inhibitor compound.