

Figure S1. (A) Growth of wild-type (closed symbols) and *fakA* mutant (open symbols) in TSB + 14 mM glucose with 1:3.3 media to flask ratio (circles) and 1:1.66 flask to media ratio (squares). **(B)** Growth of wild-type (closed circle) and *fakA* mutant (open circle) in TSB without glucose. **(C)** Growth of wild-type (closed circle), *fakA* mutant (empty circle) and *fakA* complement (filled triangle) grown in TSB buffered with 50 mM MOPS. Data is the average (n=3) with standard deviation of a representative experiment. All points have error bars and may be smaller than symbols. *denote significant difference ($p < 0.05$) from wildtype using a t-test.

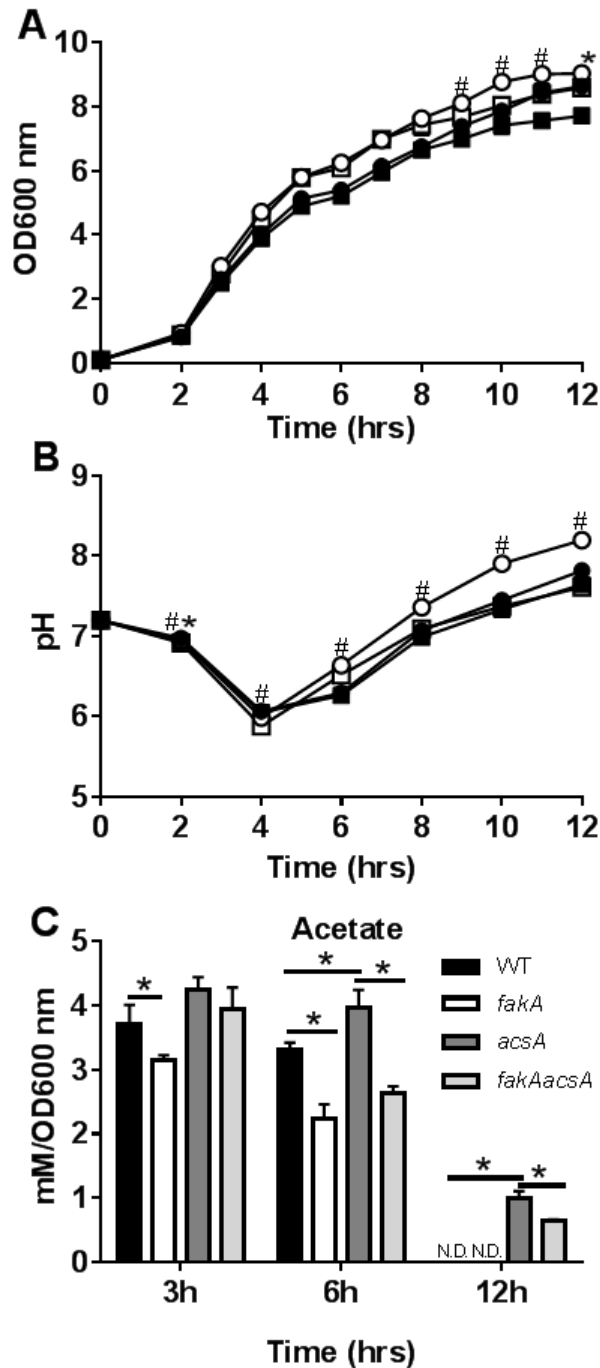


Figure S2. (A) Growth of wild-type (closed circle), *fakA* mutant (open circle), *acsA* mutant (closed square), and *fakA acsA* mutant (open square) **(B)** pH of culture from panel A. **(C)** Quantification of acetate in the culture media at indicated time points. Data is the average (n=3) with standard deviation of a representative experiment. For panels A and B, *denotes significant difference ($p < 0.05$) using a t-test for wild-type v.s. *acsA* mutant and “#” for *fakA* mutant v.s. *fakA acsA* mutant. For panel C, *indicates significant differences ($p < 0.05$) as indicated. ND indicates “not detected”.

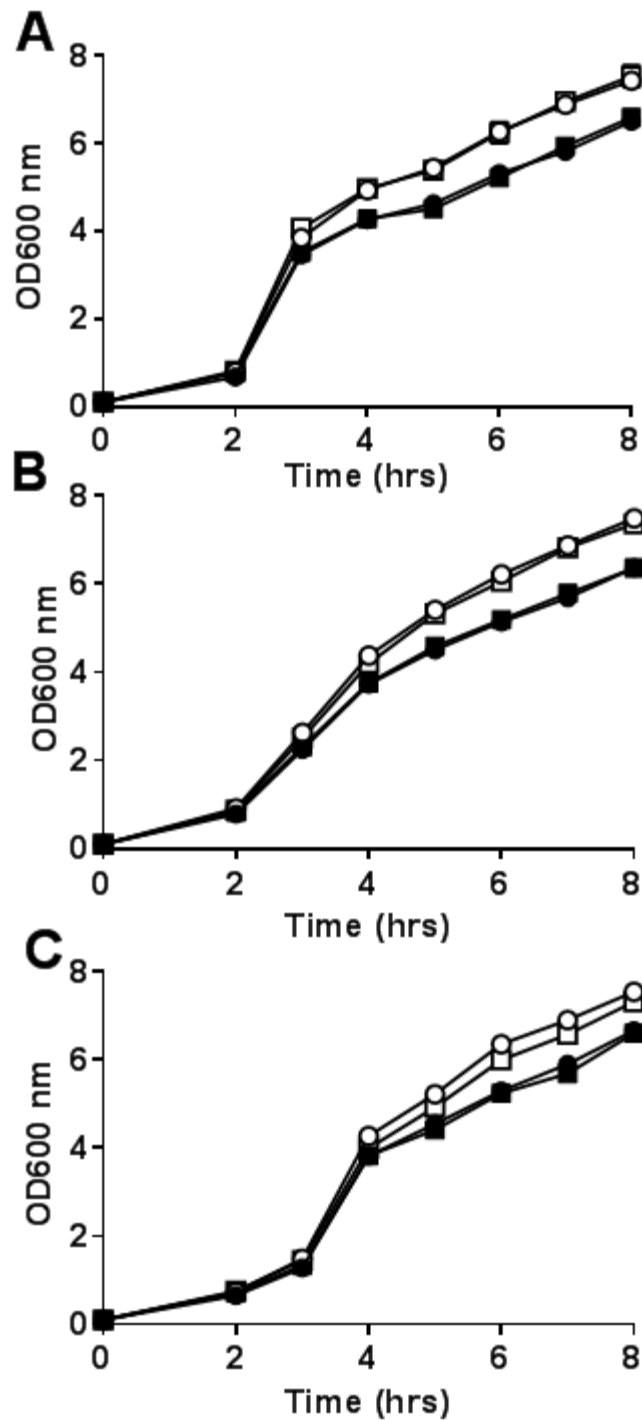


Figure S3. (A) Growth of wildtype (closed circles), *fakA* mutant (open circles), *ldh1* mutant (closed squares), and *fakA ldh1* mutant (open squares). (B) Growth of wildtype (closed circles), *fakA* mutant (open circles), *ldh2* mutant (closed squares), and *fakA ldh2* mutant (open squares) (C) Growth of wild-type (closed circles), *fakA* mutant (open circles), *alsS* mutant (closed squares), and *fakA alsS* mutant (open squares).

Table S1. Quantification of organic acids.

Organic Acid ^a	Media	Strain	Cells ^b			
			3h	P-value	6h	P-value
α-ketoglutarate	3.22	WT	1.62	<u>0.029</u>	0.45	<u>0.011</u>
		Δ <i>fakA</i>	1.19		0.66	
β-hydroxybutyrate	80.70	WT	2.42	<u>0.037</u>	4.12	0.061
		Δ <i>fakA</i>	1.72		3.16	
acetyl-CoA	n.d. ^c	WT	546.77	0.830	314.84	0.540
		Δ <i>fakA</i>	560.95		406.66	
citrate	16.00	WT	1.71	0.053	0.85	<u>0.032</u>
		Δ <i>fakA</i>	1.42		1.47	
fumarate	6.56	WT	BLOQ		0.37	0.456
		Δ <i>fakA</i>	BLOQ		0.35	
lactate	77.30	WT	343.36	<u>0.014</u>	27.54	0.278
		Δ <i>fakA</i>	474.73		24.86	
malate	702.44	WT	1.15		BLOQ	
		Δ <i>fakA</i>	BLOQ		0.42	
malonyl-CoA	n.d. ^c	WT	18.74	0.408	2.60	0.345
		Δ <i>fakA</i>	15.99		3.22	
Succinate	80.67	WT	58.22	0.388	191.81	0.142
		Δ <i>fakA</i>	48.18		247.46	

^aQuantification of selected organic acids by LC/MS/MS of sterile media before inoculation or cells after three and six hours of growth in TSB + 14 mM glucose.

^bvalues are in nM per mg protein. Statistically-significant differences are marked by underline. BLOQ indicates below limit of quantification.

^cnot determined

Table S2. Quantification of cellular nucleotides

Nucleotide ^a	3 hrs				6 hrs			
	WT ^b	$\Delta fakA$ ^b	% change ^c	P-value	WT ^b	$\Delta fakA$ ^b	% change ^c	P-value
NAM	1.42	3.88			1.13	1.18		
NMN	0.38	0.12	-68.6	0.001	0.20	0.21		
NAD	46.84	28.18	-39.8	0.005	40.99	52.39		
NADP	2.86	2.04			1.96	3.13	60.1	0.002
AMP	70.26	71.47			32.67	59.03	80.7	0.004
ADP	18.25	17.20			7.97	12.20	53.0	0.017
ADPR	11.97	18.55	55.0	0.005	5.57	2.55	-54.2	0.011
ATP	6.52	6.09			1.92	3.22	67.2	0.015
GMP	14.58	10.27	-29.5	0.007	9.40	18.57	97.5	0.005
GDP	15.39	10.44	-32.1	0.001	6.34	9.89	56.1	0.013
GTP	3.61	2.19	-39.2	0.002	1.32	1.67		
CMP	7.55	6.18	-18.1	0.015	6.46	12.79	97.9	0.006
CDP	1.66	1.03	-37.9	0.032	1.07	1.78	66.6	0.018
CTP	0.87	0.47	-45.2	0.001	0.30	0.47	56.1	0.029
UMP	15.80	19.20	21.5	0.014	8.46	20.08	137.3	0.009
UDP	18.36	19.73			8.57	13.73	60.2	0.009
TMP	5.17	5.37			2.92	14.77	406.6	0.021
TDP	3.57	4.91	37.3	0.002	1.43	4.64	224.9	0.004
TTP	0.80	0.85			0.21	0.76	253.4	0.002
IMP	10.58	8.22	-22.4	0.033	1.38	4.95	258.3	0.005
IDP	0.36	0.16	-57.2	0.003	0.04	0.03		
ITP	0.54	0.23	-58.0	0.008	0.06	0.05		
NADH	36.50	42.00			19.60	14.32	-26.9	0.007
NADPH	1.16	0.77	-33.6	0.021	0.69	0.63		
NAD/NADH	1.37	0.67	-51.0	0.020	2.12	3.70	74.2	0.013
NADP/NADPH	2.47	2.66			2.84	5.01	76.3	0.001

^aQuantified using LC/MS/MS from cells collected after three or six hours of growth in TSB + 14 mM glucose.

^bShown as nM per mg protein, except for NAD/NADH and NADP/NADPH (shown as ratio).

^cChange in mutant from wild-type (WT) strain, red box denotes a decrease in mutant while green identifies an increase in mutant. Only those with significant differences ($p < 0.05$) are shown.

Table S3. Quantification of extracellular amino acids

Amino acid	Media ^a	Strain	Supernatant ^b			
			3h	P-value	6h	P-value
Alanine	2692.08	WT	594.03	0.256	382.43	<u>0.002</u>
		$\Delta fakA$	988.13		72.20	
Arginine	1616.45	WT	901.83	0.528	916.62	0.547
		$\Delta fakA$	1114.91		604.18	
Asparagine	870.91	WT	39.85	<u>0.036</u>	22.43	0.313
		$\Delta fakA$	75.96		19.39	
Aspartate	602.03	WT	162.84	0.567	193.21	<u>0.007</u>
		$\Delta fakA$	198.94		3.66	
Citruline	20.01	WT	19.46	0.428	26.68	0.283
		$\Delta fakA$	12.39		19.50	
Glutamate	2387.40	WT	489.98	0.265	257.68	0.085
		$\Delta fakA$	842.71		19.87	
Glutamine	542.46	WT	BLOQ		BLOQ	
		$\Delta fakA$	2.47		BLOQ	
Glycine	758.59	WT	111.32	0.729	25.32	0.369
		$\Delta fakA$	87.15		7.65	
Histidine	567.36	WT	246.92	0.253	218.62	0.591
		$\Delta fakA$	344.32		143.46	
Isoleucine	1539.47	WT	575.56	0.167	433.75	0.901
		$\Delta fakA$	895.23		403.93	
Leucine	7071.18	WT	2870.56	0.329	2322.54	0.576
		$\Delta fakA$	3832.42		1672.32	
Lysine	6442.79	WT	1662.38	0.352	1381.47	0.531
		$\Delta fakA$	2189.76		923.10	
Ornithine	51.92	WT	30.25	0.151	43.70	0.569
		$\Delta fakA$	23.79		49.01	
Methionine	1318.28	WT	443.97	0.659	385.42	0.383
		$\Delta fakA$	679.39		261.61	
Phenylalanine	1910.52	WT	1004.86	0.744	1012.08	0.494
		$\Delta fakA$	1122.32		673.73	
Proline	398.74	WT	183.10	0.446	193.10	0.670
		$\Delta fakA$	248.13		232.58	
Serine	1544.93	WT	56.55	<u>0.005</u>	15.82	0.174
		$\Delta fakA$	171.51		4.17	
Threonine	1453.77	WT	119.45	<u>0.049</u>	26.46	0.365
		$\Delta fakA$	473.13		0.97	
Tryptophan	551.52	WT	248.25	0.617	253.99	0.681
		$\Delta fakA$	293.05		196.59	
Tyrosine	563.59	WT	207.77	0.218	213.68	0.724
		$\Delta fakA$	317.66		167.61	
Valine	2717.28	WT	1184.58	0.430	1104.39	0.742
		$\Delta fakA$	1545.04		883.73	

^aµM concentration of sterile media determined by LC/MS/MS.

^bµM concentration determined by LC/MS/MS and normalized to relative optical density. Statistically-significant differences are marked with an underline. BLOQ indicates below limit of quantification.