

Table S1: Plasmids

Plasmid	Genotype	
pAH25	<i>amyE::spec amp</i>	(gift from Amy Camp, Mount Holyoke College)
pAH54	<i>spec amp</i>	(gift from Amy Camp, Mount Holyoke College)
pMarA	<i>TnYLB-1 amp mls ori^{BsTs}</i>	(Le Breton <i>et al.</i> , 2006)
pMiniMAD	<i>ori^{BsTs} amp mls</i>	(Patrick & Kearns, 2008)
pDG1515	<i>tet amp</i>	(Guérout-Fleury <i>et al.</i> , 1995)
pDG268	<i>amyE::lacZ cat amp</i>	(Antoniewski <i>et al.</i> , 1990)
pDP374	$\Omega\Delta$ <i>fliT mls amp</i>	
pDR111	<i>amyE::P_{hyspank} spec amp</i>	(Ben Yehuda <i>et al.</i> , 2003)
pEX44	<i>lacZ cat amp</i>	(gift from Patrick Eichenberger, NYU)
pEC1	$\Omega\Delta$ <i>motA mls amp</i>	
pEC6	<i>amyE::P_{motA}-motA motB spec amp</i>	
pEC7	<i>amyE::P_{motA}-motA motB^{D24N} spec amp</i>	
pEC8	<i>amyE::P_{motA}-motA motB^{D24E} spec amp</i>	
pEC10	<i>amyE::P_{hyspank}-motA motB spec amp</i>	
pEC12	<i>pgsEΩlacZ cat amp</i>	
pEC13	$\Omega\Delta$ <i>mfd mls amp</i>	
pEC14	$\Omega\Delta$ <i>comQ mls amp</i>	
pEC15	$\Omega\Delta$ <i>flgL mls amp</i>	
pEC16	<i>amyE::P_{motA}-motA^{P155T} motB spec amp</i>	
pEC17	<i>amyE::P_{motA}-motA^{P155R} motB spec amp</i>	
pEC18	<i>amyE::P_{motA}-motA^{R90H} motB spec amp</i>	
pEC19	<i>amyE::P_{ywtD}-lacZ cat amp</i>	
pEC20	<i>amyE::P_{hyspank}-degQ spec amp</i>	
pEC24	<i>amyE::P_{motA}-motA^{R90G} motB spec amp</i>	
pEC25	$\Omega\Delta$ <i>pgsB mls amp</i>	
pEC27	<i>amyE::P_{motA}-motA^{P203R} motB spec amp</i>	
pEC30	<i>amyE::P_{motA}-motA^{P203T} motB spec amp</i>	
pEC32	$\Omega\Delta$ <i>comX mls amp</i>	
pEC33	$\Omega\Delta$ <i>comP mls amp</i>	
pMP196	$\Omega\Delta$ <i>degS mls amp</i>	

Table S2: Primers

Primer	Sequence
93	ACCCGAATGTAAAACCGACAACGA
94	GAACAACCTGCACCATTGCAAGAAACAGGTGTAAGATAATCAAAACGTTT
95	TTGATCCTTTTTTTATAACAGGAATTCAGGAAAATCGGCGTGTGGAAATAG
96	ACGGACGATTACATTCAGGCCGT
695	GCTTGTA AATTCTATCATAATTG
696	AGGGAATCATTGGAAGGTTGG
822	TGAACAAATTCACAATGTCCCTAAAG
823	TGCGTCAATCGAGCTGCTCGC
1008	TGCAGCCGCTGAAGAATATGGCA
1535	CGGCACTGATCCATTCTCCGTCA
1536	CAATTCGCCCTATAGTGAGTTCGTCGCCGCTTTTCACCTCCTCTGA
1537	CCAGCTTTTGTTCCTTTAGTGAGGCGTTTACCCTCCCCCTTTTCTCT
1538	GTGGCCCATGATCACCAGGCAA
2401	AGGAGGAATTCTTATCCGACTCGATATGAGCGAATA
2402	CTCCTCTCGAGAATACCGATTAACGAAGTTTATCCAT
2403	AGGAGCTCGAGAAGGACCGCTTGAAATTTGCAGAAC
2404	CTCCTGGATCCGCATCGGTCTTTTGAACGATATTTTC
2465	CTCCTCTCGAGTTTTTCATTTGTTTCCGCTGCGCC
2471	CTCCTGGATCCTTTGCTATTTTTCATTTGTTTCCGCTG
2472	AGGAGGTCGACAACCATCCAAGAATGGGTTGAGGA
2547	AGGAGCTCGAGAACCCTTATAACAACATTACGAT
2548	CTCCTGGATCCGTCAAAATAGCGCTCCAGCT
2571	CGTTCCTTACGCCAACATCCTTACTCTTCTC
2572	GAGAAGAGTAAGGATGTTGGCGTAAGGAACG
2623	GGACAAGCACCGAAAGTCATCGA
2630	CGTTCCTTACGCCGAGATCCTTACTCTTC
2631	GAAGAGTAAGGATCTCGGCGTAAGGAACG
2641	AGGAGGAATTCAGAAGTACTGTGAAAAGTATTG
2642	CTCCTCTCGAGTGACAGCATACTCTTCGTCTC
2773	AGGAGGTCGACGATATTAACCATAGACAAGCTAGTAAA
2774	CTCCTGTAGCCCTGATATGTCACAAGGCTTCCTT
2811	AGGAGGAATTCCATGCGGACATTGTTGTTGTGCAG
2812	CTCCTGGATCCGAATTATTTATTGGCGTTTACCGGTT
2840	AGGAGGAATTCCACGGCATAATGTCGGATTTATGG
2841	CTCCTGGATCCAACCGTTGATGATGGATTTAAAGTCA
2842	AGGAGGGATCCGCTGGGCATGCTGAAGGGCTTA
2843	CTCCTGTGACAATCCAGCTGCTGTTTCAACGGC
2844	AGGAGGAATTCGATCAGGCGCCTTATAGTTTGTTTA
2845	CTCCTGGATCCTGAGTAAAGAAGTTGTGACAAATCCT
2846	AGGAGGGATCCAGGAATGCTTATTAAGTTATACAAAGG
2847	CTCCTGTGACATCAATATCTACTTGTAATAGTCTTGA
2848	AGGAGGGTACCCTGTGTTGGATGTCAATTCTCTGC
2849	CTCCTGTGACCTGTATCATGCCTTGTGTTACTCTC
2850	AGGAGGTCGACACTCTGGCCGTTAATGCGCAAATT
2851	CTCCTGGATCCTATTCTACATCTATATTTAAATCAGGGT
2914	CTCAGCTTGCACACCGCAAGGTCT
2915	AGACCTTCGCGGTGTGCAAGCTGAG
2916	GGAACGTACGCTCGGACACTTGGAGTAC
2917	GTACTCCAAGTGTCCGAGCGTACGTTCC
2918	CAGGAACGTACGCTACGACACTTGGAGT
2919	ACTCCAAGTGTGCTAGCGTACGTTCTCTG
2955	AGGAGGAATTCATATCCATGAAGCGACACCTGCG

2956	CTCCTGGATCCCCTCTTTTTGCAGTTTTAACATCGG
2957	AGGAGAAGCTTGTTACCATAACAATTCATTGATCT
2958	CTCCTGCTAGCATCCATTTATTGTATCGGTAGAACGA
3001	CTCCTCTCGAGGACAGCACAGGCTATAATGAGTAAC
3002	AGGAGCTCGAGCTGCTATTGAGCGGAGCGACCT
3003	CTCCTGAATTCGGTAAAGCCAAGCGTCGCAATCG
3008	GGCTCAGCTTGCAGGCCGCGAAGGTCTG
3009	CAGACCTTCGCGGCCTGCAAGCTGAGCC
3010	CAGGGTATGTGTTATGGCATCGTTTTCGCAAATAAAATAAAAC
3011	GTTTTAATTTATTTGCGAAACGATGCCATAACACATACCCTG
3012	CAGGGTATGTGTTATGGCATACTTTGCAAATAAAATAAA
3013	TTTAATTTATTTGCGAAAGTATGCCATAACACATACCCTG
3043	AGGAGGGATCCGACAAAGTCTTCACAAATATGGATGA
3126	AGGAGGAATTCGATCTTGAAGATGAAGATAATCACCA
3127	CTCCTCTCGAGTTAATTAGGTCTTGCATCTTGTATCC
3128	AGGAGCTCGAGCTCAATGGGGTGATTAATAGGTG
3129	CTCCTGGATCCAATACCAAGATTAAATATTGGTATGATAT
3130	AGGAGGAATTCTCTGATTGCACTAGCAAATGTAGCA
3131	CTCCTCTCGAGTATTAAGTCTTCAATATTAATCCACCT
3132	AGGAGCTCGAGGCTGATATTGAAATCGAATTGTAATGG
3133	CTCCTGGATCCGAAGAATCAGGCATTCTCTAGGTG
3899	AGGAGGGATCCCACGTATGTTTCGAATTCAACAT
3900	CTCCTGCGGCCCGCCTTTGTTTTATTTCATAATTTCCCTC
3901	AGGAGGCCGCCGCGTTCCGTTATCTCTTTGACTATG
3902	CTCCTGTGACACGGCTTGCCTCCGGTCGTTTACA

SUPPLEMENTAL REFERENCES

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Le Breton Y, Mohapatra NP, Haldenwang WG. 2006. *In vivo* random mutagenesis of *Bacillus subtilis* by use of TnYLB-1, a mariner-based transposon. *Appl. Environ. Microbiol.* **72**:327–333.

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SUPPLEMENTAL FIGURE LEGEND

Figure Legend S1. MotA/MotB are required for swimming motility but MotP/MotS are not. Swim plates of LB containing (+NaCl) or lacking (-NaCl) added NaCl and fortified with 0.3% agar were toothpick inoculated in the center of the plate and incubated at 37°C for 8 hours. Plates were filmed against a black background such that zones of colonization appear white and uncolonized agar appears black. Ancestral strain derivatives of *B. subtilis* readily swarm atop solid surfaces even surfaces solidified with 0.3% agar that normally supports swimming motility. To discourage swarming and encourage swimming motility, cells were mutated for surfactin synthesis (*urfAA*) and EPS biosynthesis (*epsH*). The following strains were used to generate this figure: wild type (DK1484), *motAB* (DK1492), and *motPS* (DK1493).

Figure S1

