Supporting Information

Defeu Soufo et al. 10.1073/pnas.0911979107

SI Materials and Methods

***Bacillus* Strains and Plasmids.** All plasmid constructions (also plasmids for proteins expression and purification) are indicated in Table S1, and all strains are listed in Table S2. A *B. subtilis* strain that expresses TAP-tag-MreB or EF-Tu-TAP-tag at the original locus was obtained by transforming wild-type strain with pJS55 or pJS58 plasmid generating strain JS82 (*Pxyl-TAP-tag-mreB*) or JS86 (*tufA-TAP-tag*), respectively. The negative control strain (JS83; *Pxyl-TAP-tag*::*amy*) was also constructed bearing only *TAP-tag* at *amy* site using pHJDS6 plasmid to transform wild-type strain. Cell extracts from the above strains were subjected to TAP procedure (see TAP method) and the eluates were resolved by SDS-polyacrylamide gel. The bands were analyzed by LC-MS/MS. An EF-Tu-CFP fusion at original locus was generated by transforming wild-type strain with pJS59 plasmid generating strain JS86 (*tufA-Bscfp*). For dual visualization of MreB and EF-Tu, pJS59 was used to transform competent cells of strain JS36 (*Pxyl-yfp-mreB::amy*)(1) resulting in JS87 strain (*Pxyl-yfp-mreB::amy, tufA-Bscfp*). Transcription of YFP-MreB was induced by the addition of 0.5% xylose.

For BiFC, a strain was generated by transforming JS71(1) expressing YN-MreB (N-terminal BiFC fragment) at the *amy* locus with the plasmid pJS61 (containing EF-Tu fused to the C-terminal fragment of BiFC) resulting in JS94 strain (*Phyper-SPANK-yn-mreB::amy, tufA-yc*), in which EF-Tu-YC is expressed from the original gene locus. Likewise, the plasmid pJS61 was used to transform JS72 (*1*) (expressing YN-Mbl at the *amy* locus) or JS77 (expressing YN-MreC at the *amy* locus) resulting in strains JS95 (*Phyper-SPANK-yn-mbl::amy, tufA-yc*) or JS96 (*Phyper-SPANK-yn-mreC::amy, tufA-yc*) respectively. BiFC was induced by the addition of 1 mM IPTG, for the transcription of the gene fusion at the *amy* site. The stable expression of all BiFC fusions was verified by Western blotting using MreB or EF-Tu antibodies, or GFP antibodies (for the Mbl or MreC fusions).

To visualize EF-Tu-CFP in the absence of MreB, the plasmid pJS60 was used to transform *ΔmreB* (2) competent cells, to obtain JS97 strain (*trpC2 Ωneo3427ΔmreB, tufA-Bscfp*). To generate strains with *tufA* under the control of xylose promoter, plasmid pJS61 was used to transform wild type or JS17, generating strains JS91 (*Pxyl-tufA*) or JS98 (*Pxyl-gfp-mreB::amy, pxyl-tufA*).

TAP Method. One liter of *TAP-tag-mreB* (JS82) or *tufA-TAP-tag* (JS86) or cells expressing TAP-tag only (JS83) were grown at room temperature in LB [(supplemented with appropriate antibiotics (5 μg/mL chloramphenicol for JS82 and JS86 or 50 μg/mL spectinomycin for JS83) and inducer (0.5% xylose for JS82 and JS83)] and harvested (6,000 rpm at 4 °C for 15 min) at OD_{600}: 1.5–3.0. The cell pellet was frozen in liquid nitrogen and stored at −80 °C until use. The cells were disrupted using French press in 20 mL of Nonidet P-40 buffer (6 mM Na_2HPO_4, 4 mM $NaH_2PO_4xH_2O$, 1% NONIDET P-40, 150 mM NaCl, 2 mM EDTA pH 8.0, 50 mM NaF, 4 μg/mL leupeptin) containing the following protease inhibitors per 50 mL of Nonidet P-40 buffer: 1 complete mixture tablets, EDTA free, 130 μL of 0.5 M benzamidine prepared fresh in 100% ethanol, 500 μL of 0.1 M PMSF prepared fresh in 100% ethanol. The lysate was cleared by centrifugation at 16,000 rpm at 4 °C for 30 min, and TAP procedure was carried out as described by Newo S. et al. (3). Half the volume of bed volume of IgG-Sepharose beads and calmodulin resin was used because the experiments were run with 20 mL of lysate instead of 50 mL as mentioned in the cited reference. The eluted protein complexes were separated on SDS/PAGE and visualized by Coomassie blue staining. The bands of interest were excised out from the gel and the proteins were identified by mass spectrometry.

Coexpression and Coelution of Strep-MreB and EF-Tu-6His from Streptavidin Columns. *E. coli* BL21(DE3) cells simultaneously transformed with pCDFDuet plasmid containing *strep-mreB* and pETDuet-1 plasmid containing *tufA-6his* were inoculated in 400 mL of LB supplemented with ampicillin (100 μg/mL) and grown at 37 °C until OD_{600}: 0.6–0.8. The expression of proteins was induced by adding 1 mM IPTG to the culture, which was left to grow for an additional 4 h. Cells were spun down at 6,000 rpm at 4 °C for 15 min and the pellet was quickly frozen in liquid nitrogen and stored at −80 °C for until use. Buffers and columns [two Streptavidin columns (1-mL bed volume), one for the test and the other for the control] for the following steps were purchased from IBA. Cells were disrupted with French press in W buffer (100 mM Tris·HCl pH 8.0, 150 mM NaCl, 1 mM EDTA) and the lysate was cleared by centrifugation at 16,000 rpm and 4 °C for 30 min. For the control experiment, *E. coli* cells expressing only EF-Tu-6His were grown, induced and the cell lysate obtained in the same conditions as above. Lysate from the cells expressing both Strep-tag-MreB and EF-Tu-6His was loaded in one Streptavidin column "test" while the lysate from cells expressing only EF-Tu-6His was loaded in the other "control" (to discriminate the fact that the elution of EF-Tu-6His from the "test" column could be due to its non specific retention to the Streptavidin column). The columns were washed with W buffer followed by elution with E Buffer (100 mM Tris·HCl pH 8.0, 150 mM NaCl, 1 mM EDTA, 2.5 mM desthiobiotin). Proteins in eluted fractions were separated by SDS/PAGE and visualized by Coomassie blue stain.

The experiment with ScpB/EF-Tu or Mbl/EF-Tu followed the same protocol as above, except that pCDFDuet plasmids containing *scpB-strep* or *strep-mbl* were used.

Coelution of Purified EF-Tu-6His from Streptavidin Columns Preloaded with Purified Strep-MreB. For this experiment, Strep-tag-MreB and EF-Tu-6His were purified in their most stable condition [low salt concentration and ATP (see below) for MreB or GDP and Mg^{+2} for EF-Tu)]. *E. coli* cells expressing each tagged protein were grown and induced as mentioned in the previous experiments. Different buffers were used to purify each protein.

Lysates from cells expressing Strep-tag-MreB were obtained with lysis/wash1 buffer (100 mM Tris·HCl pH 8.0, 150 mM NaCl, 5 mM DTT, 0.02% NaN_3, 0.1 mM $CaCl_2$, 0.2 mM ATP). After loading the cleared lysate, the column (1-mL Streptavidin column) was washed with decreasing concentrations of Tris and NaCl buffers [l mL of lysis/wash1, 1 mL of wash2 (50 mM Tris·HCl pH 8.0, 75 mM NaCl, 5 mM DTT, 0.02% NaN_3, 0.1 mM $CaCl_2$, 0.2 mM ATP), 1 mL of wash3 (25 mM Tris-HCl pH 8.0, 30 mM NaCl, 5 mM DTT, 0.02% NaN_3, 0.1 mM $CaCl_2$, 0.2 mM ATP), 1 mL of wash4 (10 mM Tris·HCl, 10 mM NaCl, 5 mM DTT, 0.02% NaN_3, 0.1 mM $CaCl_2$, 0.2 mM ATP), 1 mL of wash5 (5 mM Tris-HCl pH 8.0, 5 mM DTT, 0.02% NaN_3, 0.1 mM $CaCl_2$, 0.2 mM ATP)]. Protein fractions were then eluted with elution buffer (5 mM Tris·HCl pH 8.0, 5 mM DTT, 0.02% NaN_3, 0.1 mM $CaCl_2$, 0.2 mM ATP, 2.5 mM desthiobiotin) and analyzed on SDS/PAGE. Strep-MreB containing fractions were collected in a single sample and dialyzed overnight against wash5 buffer to remove desthiobiotin.

Lysates from cells expressing EF-Tu-6His were obtained with lysis/wash buffer [50 mM Tris·HCl pH 7.6, 60 mM NH_4Cl, 10%

glycerol, 10 μM GDP, 7 mM β-mercaptoethanol, 7 mM $MgCl_2$ (add after cells disruption), 1 mM imidazole]. The cleared lysate was run through 1-mL Histrap HP column attached to Äktaprime plus from Amersham. After washing steps, protein fractions were then eluted with elution buffer (50 mM Tris·HCl pH 7.6, 60 mM NH_4Cl, 10% glycerol, 10 μM GDP, 7 mM β-Mercaptoethanol, 7 mM $MgCl_2$, 200 mM imidazole) in a gradient from 20% to 100%. EF-Tu-6His containing fractions were collected in a single sample and dialyzed overnight against lysis/wash buffer.

Strep-Mbl was purified via the same protocol used to purify Strep-tag-MreB as mentioned in this experiment. ScpB-strep was purified following the Strep-tag purification protocol described in the manual from IBA and was dialyzed against wash5 buffer.

Cell lysates from *E. coli* cultures containing an empty plasmid were also run throughout Strep-tag purification protocol and the dialyzed eluates (control sample) were used for control experiment.

For the experiment, Strep-tag proteins (Strep-MreB, Strep-Mbl and ScpB-Strep) [(1 mL of 500 μg/mL Strep-MreB/Mbl and 1 mL of 1.5 mg/mL ScpB-Strep (ScpB was highly expressed compare with Strep-MreB/Mbl)] were first loaded onto "test" columns (each protein per column) and the control sample was loaded onto the "control" column. EF-Tu-6His (1.5 mL of 700 μg/mL) was added onto all four columns followed by washing steps with washing buffer (50 mM Tris·HCl pH 7.6, 60 mM NH_4Cl, 10% glycerol, 10 μM GDP, 7 mM β-mercaptoethanol, 7 mM $MgCl_2$, 1 mM imidazole). The proteins were finally eluted with the standard elution E buffer from IBA (100 mM Tris·HCl pH 8.0, 150 mM NaCl, 1 mM EDTA, 2.5 mM desthiobiotin). The samples were resolved by SDS/PAGE and visualized by Coomassie blue stain.

35S-Methionine Labeling Experiment. Overnight cultures (WT and *Pxyl-tufA*) grown in LB were diluted to OD_{600}: 0.025 in 20 mL of M9 glucose and M9 fructose + 0.5% xylose for WT cells or M9 fructose + 0.5% xylose supplemented with 0.01% (wt/vol) Tryptophan, 0.01% (wt/vol) phenylalanine, and 0.01% (wt/vol) yeast extract for *Pxyl-tufA* (JS91). Fresh cultures were grown at 37 °C until OD_{600}: 0.3–0.5 and each split into two separate flasks (experiments were carried out in duplicate). 100 μCi of 35S methionine were added to each flask. 1-mL samples were taken as time point zero, added in prechilled 1.5-mL Eppendorf tubes containing 100 μL of stop solution (100 mM Tris·HCl pH 7.5, 1mg/mL chloramphenicol, 10 mM methionine), and kept on ice. The following 1-mL samples were taken, stopped, and kept on ice each minute until 10 min. All samples were then spun down for 10 min at 13,000 rpm and 0 °C. Obtained pellets were resuspended in 1,000 μL of cold wash solution (100 mM Tris·HCl pH 7.5, 0.1 mg/mL chloramphenicol) and spun down again (the pellet could be frozen for later use). The pellets were further resuspended in 100 μL of cold cell lysis buffer (10 mM Tris·HCl pH 7.4, 1mg/mL $MgCl_2 \times 6\ H_2O$, 50 μg/mL RNase, 50 μg/mL DNase, 100 μg/mL lysozyme, 243 μg/mL PMSF) and transferred into Eppendorf tubes containing 120–150 μg of glass beads. Cells were disrupted (6 min at a speed of 30 1/s) using Mixer Mill MM 200 from Retsch. Cells extract were cleared at 13,000 rpm and 4 °C for 15 min. 5 and 10 μL of each sample were transferred on Whatman filter papers and washed with 10 min with cold 10% TCA solution, 10 min with cold 5% TCA solution and twice 10 min with 100% ethanol at room temperature. Whatman filter papers (containing samples) were finally air-dried and placed in a scintillation counter.

Immunofluorescence. Cells were fixed with glutaraldehyde as described by Pogliano et al. (4). Cells walls were made permeable with lysozyme, and cells were treated with 1:5,000 diluted EF-Tu antiserum from rabbit, and with secondary Alexa Fluor 488-coupled secondary antibody (Molecular Probes/Invitrogen). A standard FITC/GFP filter was used for the detection of fluorescence; outlines of cells were captured using bright field illumination. EF-Tu antiserum (provided by Gramsch Laboratories) was obtained from rabbit which has being immunized with a specific EF-Tu peptide from NMI Technologie Transfer.

Microscopy. Fluorescence microscopy was performed using an Olympus AX70 microscope or a Zeiss Axioimager equipped with a digital CCD camera and a 100× TIRF objective with an aperture of 1.45. 2D and 3D deconvolution was performed using Auto Deblur ×1.4.1. For 3D reconstruction, 100–130 images (spacing ~0.08 μm) were taken through the focal plane and were processed in Auto Deblur ×1.4.1 and MetaMorph 6.3r6 programs (Universal Imaging). For FRAP studies, a 50-mW argon laser was mirrored onto the specimen from above the filter plane, using the side port of the Zeiss microscope. The size of the laser beam (50 μm) was generated by a pinhole inserted into the laser beam within the module that incorporates the optical wire into the side port (custom made by A&S).

Strains from Movies S1–S3 were cultured in $S7_{50}$ minimal media at room temperature; late-exponentially growing cells (OD 1.5) were used for time-lapse microscopy. Strains for Movies S4–S6 were grown in M9 medium, grown to OD of 0.5.

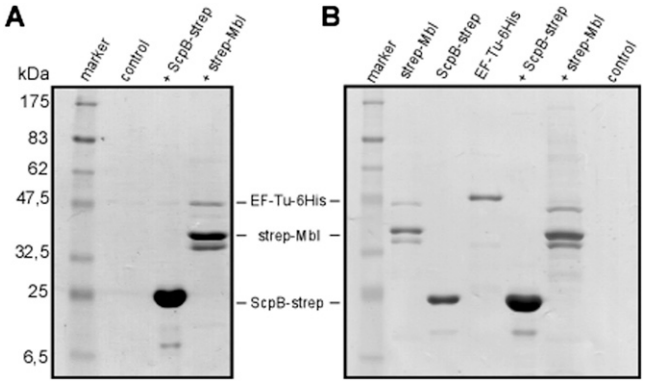


Fig. S1. Control experiments for the interaction studies in Fig. 1. (*A*) Coexpression control experiments. EF-Tu-6His was expressed alone "control" or coexpressed with ScpB-strep "+ ScpB-strep" or strep-Mbl "+strep-Mbl" in *E. coli* cells. Shown are main elutions from streptavidin columns. Note that EF-Tu-6his did not significantly coeluted with ScpB-Strep, and little coelution was observed with strep-Mbl, compared with the ≈1:1 ratio observed in the strep-MreB/EF-Tu-6His coelution (Fig. 1*C*). (*B*) Control experiments with purified proteins (lanes 2–4). Purified EF-Tu-6His was loaded onto streptavidin columns preloaded with elution fractions from Streptavidin columns from *E. coli* cell extract ("control") or with purified ScpB-strep ("+ ScpB-strep") or purified strep-Mbl ("+strep-Mbl"). Shown are main elution fractions. Note that EF-Tu-6His did not coelute with purified strep-Mbl or purified ScpB-strep.

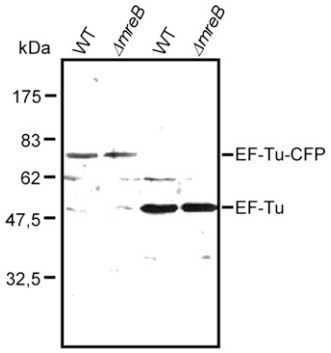


Fig. S2. Western blot showing the level of EF-Tu or of EF-Tu-CFP in wild-type (WT) and Δ*mreB* (3725) cells. Cell extracts from late exponentially growing WT cells (OD 3.5) or Δ*mreB* cells in SMM/PAB medium at room temperature were separated by SDS/PAGE and immunoblotted using α-EF-Tu antiserum. Equal amounts of protein were loaded on the first and second or in the third and fourth lane. Note that the antiserum reacts better with wild-type EF-Tu than with the CFP fusion.

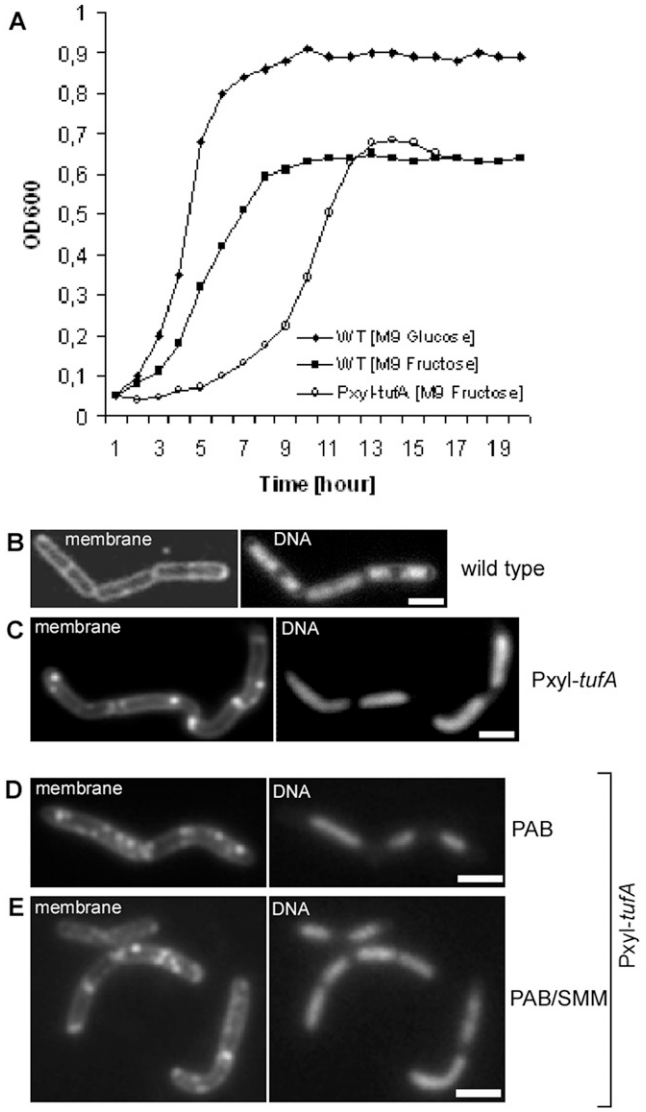


Fig. S3. Effect of the depletion of EF-Tu on growth of *B. subtilis* cells. (*A*) To compare the translation efficiency of *B. subtilis* wild type vs. *Pxyl-tufA* (JS91), the strains were grown in the conditions in which they both exhibit a similar growth rate. These conditions were given when cells were grown in M9 medium with fructose instead of glucose, between an OD of 0.2 and 0.5 (doubling time for wild-type cells was 98 min and for pxyl-tufA cells was 93 min). Overnight cultures (grown in LB) were diluted to OD_{600}: 0.025 into a fresh M9 media and grown at 37 °C. Samples were taken each hour for OD measurement at 600 nm. Incorporation of radioactive methionine (Fig. 4*C*) was measured at an OD_{600} of 0.5. (*B–E*) Fluorescence microscopy of exponentially growing *B. subtilis* cells. (*B*) Wild-type cells. (*C*) *Pxyl-tufA* cells (JS91, EF-Tu depletion). Cells were grown in LB (wild-type strain) or LB plus xylose (EF-Tu depletion strain) until early exponential phase (OD_{600} of 0.5–0.8). Note that the phenotype (lost of rod shape of *Pxyl-tufA* strain) observed here is the same as the one shown in Fig. 4 *D* and *E* with cells grown in M9. (*D* and *E*) *Pxyl-tufA* cells (JS91, EF-Tu depletion) grown in PAB plus xylose (*D*) or or in PAB/SMM plus xylose (*E*). (Scale bars: 2 μm.)

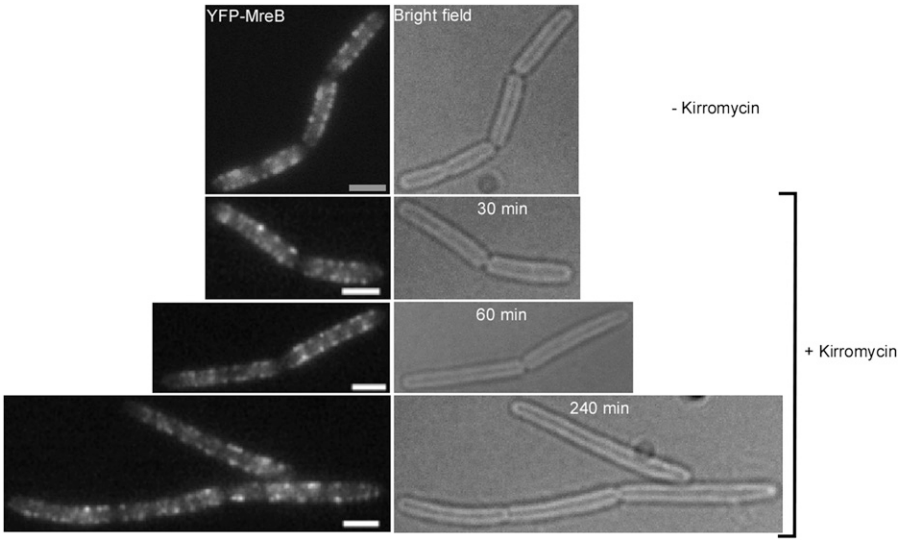


Fig. S4. Effect of the addition of kirromycin on the localization of YFP-MreB (JS36) in *B. subtilis*. Cells were grown in $S7_{50}$ minimal media at 37 °C until an OD_{600} of 1.5 (late exponential phase). 100 μg of kirromycin were added and samples were collected every 30 min for microscopy. Note that growth has ceased after 60 min and no effect on YFP-MreB localization was observed. (Scale bars: 2 μm.)

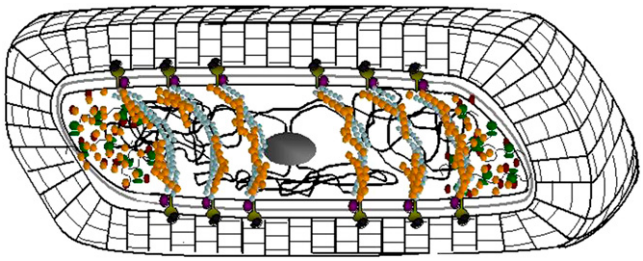


Fig. S5. Model of the dual function of EF-Tu in *B. subtilis*. MreB filaments (gray dots) polymerize and depolymerize along a helical track underneath the cell membrane, and EF-Tu (yellow dots) colocalizes with these structures but itself forms rather static extended structures. EF-Tu affects the positioning and dynamics of MreB filaments and could thus serve as tracks for the dynamic filaments. EF-Tu is also associated with ribosomes (green dots) that are mostly located at polar zones. MreB also interacts with the MreCD membrane proteins (dark green and purple dots along the membrane), which in turn interact with cell wall synthesizing proteins (black dots). The DNA polymerase is depicted as a gray oval in the cell center, and the chromosome as black lines.

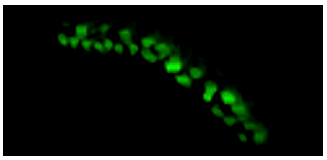
Table S1. Plasmids

Plasmid (genotype)	Construction
pJS55 (*bla Pxyl-TAP-mreB cat*)	*gfp* was replaced with TAP-tag sequence in pJS12(5) plasmid (originally constructed for GFP-MreB fusion at the original locus) using PstI and ApaI.
pHJDS6 (*bla amyE::Pxyl-TAP-tag spec*)	*gfp* was replaced with *TAP-tag* sequence in pSG1729(6) plasmid using KpnI and ApaI.
pJS58 (*bla tufA-TAP-tag cat*)	*gfp* was exchanged with *TAP-tag* sequence in pSG1164(6) using PstI and SpeI, and ≈500 bp of C-terminal part of *tufA* inserted between ApaI and ClaI restrictions sites.
pJS60 (*bla tufA-Bscfp cat*)	*gfp* was exchanged with *Bscfp* in pSG1164(6) using EcoRI and PstI and ≈500 bp of C-terminal part of *tufA* inserted between ApaI and EcoRI restrictions sites.
pJS61 (*bla tufA-yc cat*)	*Bscfp* was exchanged with *yc* (the last 86 amino acids of *yfp* from Clontech) in pJS59 (this work) using EcoRI and PstI restrictions sites.
pJS62 (*bla Pxyl-tufA cat*)	*gfp* in pHJDS1(5) was exchanged with ≈500 bp of N-terminal part of *tufA* inserted between PstI and EcoRI restrictions sites.
pJS63 (*sm Strep-mreB*)	*Strep-tag-mreB* sequence was amplified from *B. subtilis* (wild type) chromosomal DNA using an upstream primer containing *Strep-tag* sequence. The resulting PCR product was inserted between NdeI and XhoI in pCDFDuet (Novagen) vector.
pJS67 (*bla tufA-6His*)	*tufA-His-tag* sequence was amplified from *B. subtilis* (wild type) chromosomal DNA using a downstream primer containing *His-tag* sequence. The resulting PCR product was inserted between NdeI and XhoI in pETDuet (Novagen) vector.
pCDFDuet(*Strep-mbl*) (*sm Strep-mbl*)	*Strep-tag-mbl* sequence was amplified from *B. subtilis* (wild type) chromosomal DNA using an upstream primer containing *Strep-tag* sequence. The resulting PCR product was inserted between NdeI and XhoI in pCDFDuet (Novagen) vector.
pETDuet(*ScpB-Strep*) (*bla scpB-Strep*)	*scpB-Strep-tag* sequence was amplified from *B. subtilis* (wild type) chromosomal DNA using an downstream primer containing *Strep-tag* sequence. The resulting PCR product was inserted between NdeI and XhoI in pETDuet (Novagen) vector.

Table S2. Strains used in this study

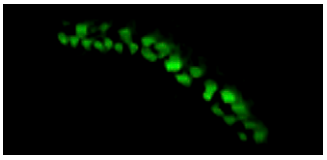
Strain	Genotype	Reference
PY79	Wild-type *Bacillus subtilis* subsp. *subtilis*	
JS12	*Pxyl-gfp-mreB*	5
JS17	*Pxyl-gfp-mreB::amy*	5
JS36	*Pxyl-yfp-mreB::amy*	1
JS69	*Pxyl-yn::amy*	1
JS71	*Phyper-SPANK-yn-mreB::amy*	1
JS72	*Phyper-SPANK-yn-mbl::amy*	1
JS77	*Phyper-SPANK-yn-mreC::amy*	This work
JS82	*Pxyl-TAP-MreB*	This work
JS83	*Pxyl-TAP-tag::amy*	This work
JS86	*tufA-TAP*	This work
JS88	*tufA-Bscfp*	This work
JS89	*Pxyl-yfp-mreB::amy, tufA-Bscfp*	This work
JS90	*tufA-yc*	This work
JS91	*Pxyl-tufA*	This work
JS92	*Pxyl-yn::amy, tufA-yc*	This work
JS93	*Phyper-SPANK-yn-mreB::amy, Pxyl-yc*	This work
JS94	*Phyper-SPANK-yn-mreB::amy, tufA-yc*	This work
JS95	*Phyper-SPANK-yn-mbl::amy, tufA-yc*	This work
JS96	*Phyper-SPANK-yn-mreC::amy, tufA-yc*	This work
3725	*trpC2 Ωneo3427 ΔmreB*	2
JS97	*trpC2 Ωneo3427 ΔmreB, tufA-Bscfp*	This work
JS98	*Pxyl-gfp-mreB::amy, Pxyl-tufA*	This work

1. Defeu Soufo HJ, Graumann PL (2006) Dynamic localization and interaction with other *Bacillus subtilis* actin-like proteins are important for the function of MreB. *Mol Microbiol* 62: 1340–1356.
2. Formstone A, Errington J (2005) A magnesium-dependent *mreB* null mutant: implications for the role of *mreB* in *Bacillus subtilis*. *Mol Microbiol* 55:1646–1657.
3. Newo AN, et al. (2007) Proteomic analysis of the U1 snRNP of *Schizosaccharomyces pombe* reveals three essential organism-specific proteins. *Nucleic Acids Res* 35:1391–1401.
4. Pogliano K, Harry E, Losick R (1995) Visualization of the subcellular location of sporulation proteins in *Bacillus subtilis* using immunofluorescence microscopy. *Mol Microbiol* 18:459–470.
5. Defeu Soufo HJ, Graumann PL (2004) Dynamic movement of actin-like proteins within bacterial cells. *EMBO Rep* 5:789–794.
6. Lewis PJ, Marston AL (1999) GFP vectors for controlled expression and dual labelling of protein fusions in *Bacillus subtilis*. *Gene* 227:101–110.



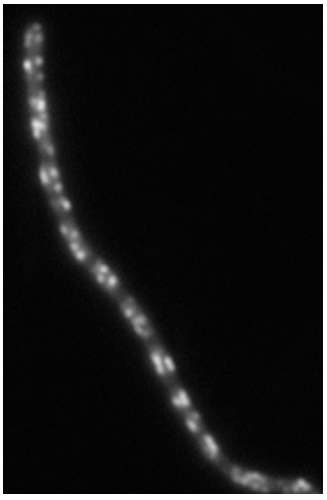
Movie S1. 3D reconstruction of exponentially growing *B. subtilis* cells expressing EF-tu-BsCFP (JS88). Horizontal orientation from −45 to + 45° angle of view.

Movie S1



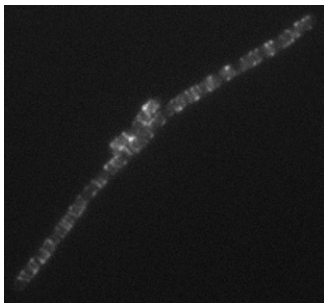
Movie S2. 3D reconstruction of exponentially growing *B. subtilis* cells expressing EF-tu-BsCFP (JS88). Horizontal orientation from −180 to + 180° angle of view.

Movie S2



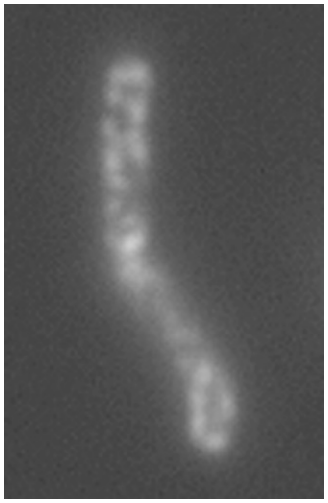
Movie S3. Time-lapse movie of exponentially growing *B. subtilis* cells expressing EF-Tu-BsCFP (JS88); images taken at 60-s intervals; shown are 5 frames/s.

Movie S3



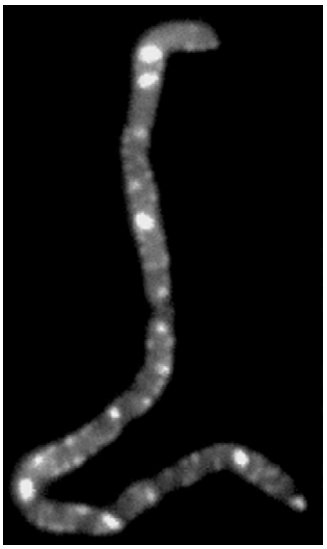
Movie S4. Time-lapse movie of exponentially growing *B. subtilis* cells expressing GFP-MreB (JS36), images taken at 10-s intervals; shown are 5 frames/s.

Movie S4



Movie S5. Time-lapse movie of exponentially growing *B. subtilis Pxyl-tufA* cells expressing GFP-MreB (JS98); images taken at 10-s intervals; shown are 5 frames/s.

Movie S5



Movie S6. Time-lapse movie of exponentially growing *B. subtilis Pxyl-tufA* cells expressing GFP-MreB (JS98); images taken at 10-s intervals; shown are 5 frames/s.

Movie S6