Moriya et al 2010

Supplementary Information

Table S1: Incorporation of ^{3}H-thymine into *B. subtilis* DNA

Strain	Condition	cpm[a]
SU8	48°C 90 min	4919 ± 235
SU8	33°C 180 min	14822 ± 153
SU8	48°C 90 min, + HPUra (100 μM)	222 ± 39
SU472 (*dnaF133*)	48°C 90 min	268 ± 46
SU472 (*dnaF133*)	48°C 90 min, + HPUra (100 μM)	140 ± 25
SU472 (*dnaF133*)	33°C 180 min	19316 ± 860
SU46 (*dna-1*)	48°C 90 min	115 ± 20
SU46 (*dna-1*)	33°C 180 min	18904 ± 288

All cells are outgrown spores in PAB with the addition of thymine (20 μg/ml) and [methyl-^{3}H]thymidine. All spores were initially allowed to germinate at 33°C for 15 min prior to transfer to the non-permissive temperature (48°C). At both temperatures the wild type parent strain (SU8) is able to incorporate significant radiolabel into DNA. Likewise both SU472 (*dnaF133*) and SU46 (*dna-1*) are able to incorporate significant radiolabel at 33°C. However both these strains at 48°C essentially show no thymine incorporation, indicating a complete block to DNA chain elongation of the first round of replication following spore germination. ^{3}H-thymine incorporation values between spores germinated and grown out at the two different temperatures cannot be directly compared due to possible changes in cellular uptake of ^{3}H-thymidine and conversion to ^{3}H-thymine in the cytosol at 33°C and 48°C.

[a] cpm- counts per minute; values are ± SEM

Figure S1: *Unreplicated bilobed nucleoids of* dna-1 *and* dnaF133 *at the non-permissive temperature*

DAPI-stained nucleoids were visualized in spores of SU46 (*dna-1*) and SU472 (*dnaF133*). Spores were allowed to germinate at 34°C for 15-20 min prior to initiation of replication. They were then transferred to the non-permissive temperature (48°C) in PAB + T for 75 minutes.

(A to E) DAPI-stained bilobed nucleoids in outgrown cells of SU46 (*dna-1*). **(F to J)** DAPI-stained bilobed nucleoids in outgrown cells of SU472 (*dnaF133*). Images shown are phase-contrast (left) and DAPI (right). Scale bars are 1 μm.

Figure S2*: Co-visualization of the Z ring and the nucleoid in cells with an unreplicated bilobed nucleoid in* dnaF133 *and* dna-1.

Spores of SU624 (*dna-1 ftsZ-yfp*) (n = 260) or SU625 (*dnaF133 ftsZ-yfp*) (n = 257) were allowed to germinate at 34°C for 15-20 min prior to initiation of replication. They were then transferred to the non-permissive temperature (48°C) in PAB + T containing 0.01% xylose for 75 min at the non-permissive temperature (48^{o}C). DAPI was added, and the nucleoid and Z ring were visualized in live cells using agarose pads containing 0.01% xylose. **(A to D)** Acentral Z rings forming beside the unreplicated bilobed nucleoids in SU624 (*dna-1 ftsZ-yfp*). **(E to F)** Midcell Z rings forming over the unreplicated bilobed nuceloid in SU625 (*dnaF133 ftsZ-yfp*). Images shown are DAPI pseudocoloured in red (left), YFP pseudocoloured in green (middle), and overlay (right). Scale bars are 1 μm.

Figure S1

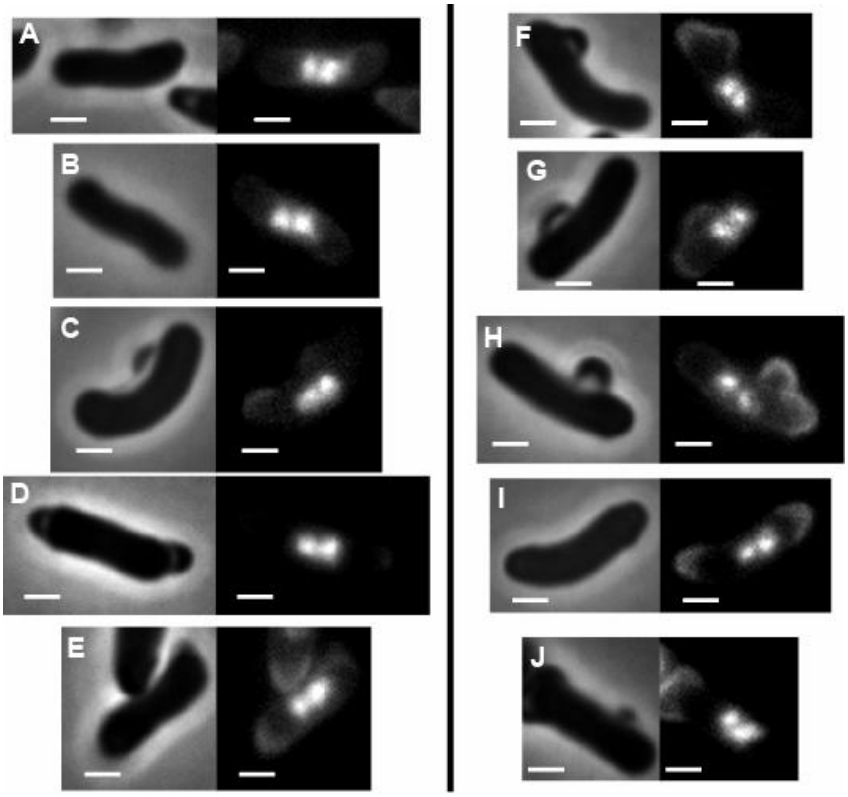

Figure S2