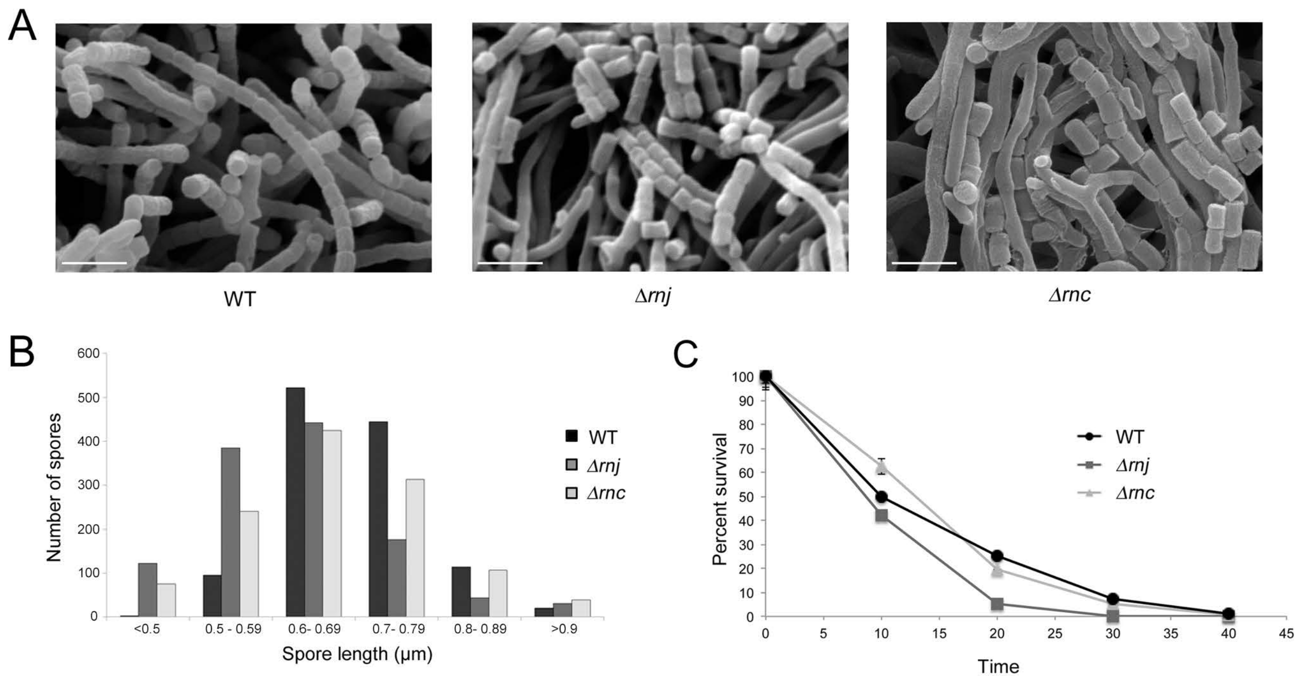
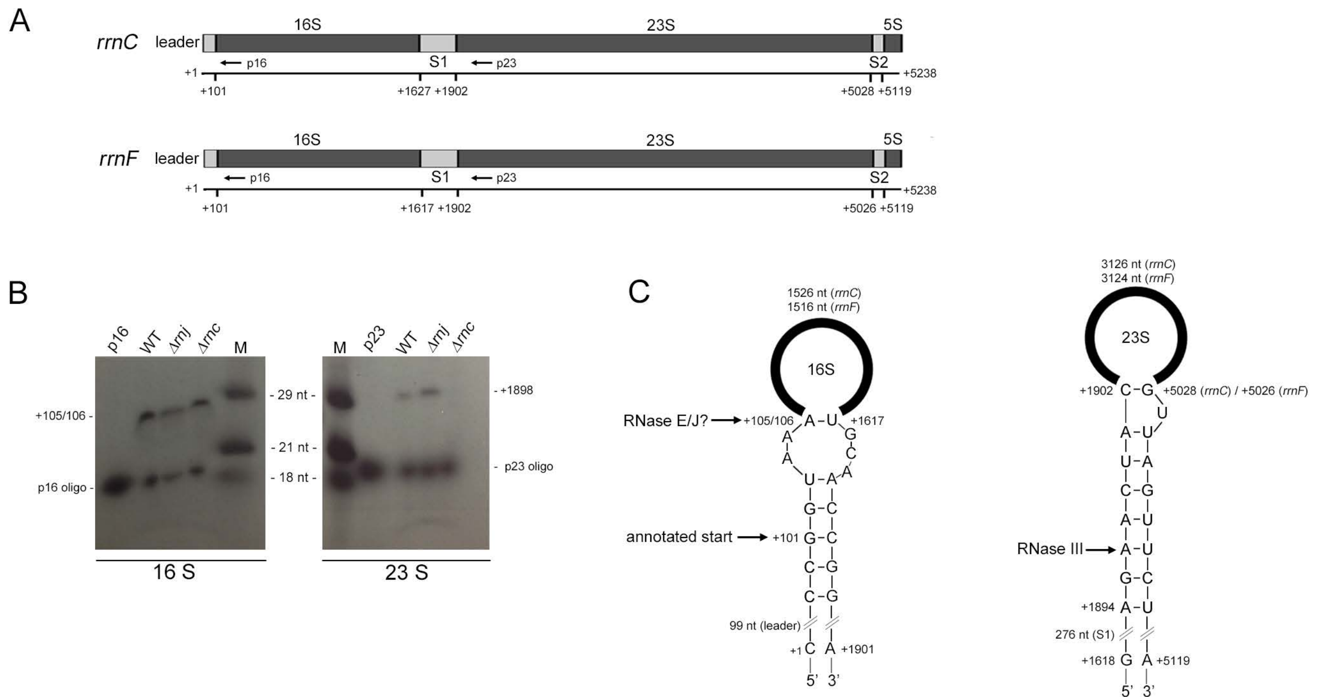
Table S1. Oligonucleotides used in this study

Name	Sequence (5′ to 3′)	Use
Sven5265 Fwd	GGTCCTCTCGGTCTGATTGGGCTGGTGAGAGGC GCAATGATTCCGGGGATCCGTCGACC	Creation of Δ*rnc* strain; confirmation of Δ*rnc* mutation
Sven5265 Rev	TGGGGCGGGGTTCTCGGGAAGCGTGGGGGACG GCCGTCATGTAGGCTGGAGCTGCTTC	Creation of Δ*rnc* strain; confirmation of Δ*rnc* mutation
Sven5265 up	GGGAACCGATGAGAAGGAC	Cloning of *rnc* for mutant complementation; confirmation of Δ*rnc* mutation
Sven5265 in	CGGTACAGCGTGTCCGTGAC	Confirmation of Δ*rnc* mutation
Sven5265 dwn	CTGGGGTGCAGGACCTCG	Cloning of *rnc* for mutant complementation
Sven5394 Fwd	CAAGGGGTACGTGGCGTGCGTGGTGAGGAGAG TCTTTTGAATTCCGGGGATCCGTCGACC	Creation of Δ*rnj* strain; confirmation of Δ*rnj* mutation
Sven5394 Rev	CGCTCCGGTGCTGCGCTGGTCAGACCGTCAGGC CCGTCATGTAGGCTGGAGCTGCTTC	Creation of Δ*rnj* strain; confirmation of Δ*rnj* mutation
Sven5394 up	CCAGAAGCTGCTCCCGGTC	Cloning of Δ*rnj* for mutant complementation; confirmation of Δ*rnj* mutation
Sven5394 in	GTTCCGGCCGATCTCACCG	Confirmation of Δ*rnj* mutation
Sven5394 dwn	GTCGGATTTCCCGCCTCCC	Cloning of *rnj* for mutant complementation; confirmation of Δ*rnj* mutation
M13 FWD	GTAAAACGACGGCCAGT	Cloning and sequencing
16S rRNA	CGGAGAGTTTGATCCTGGC	Reverse transcription for primer extension
23S rRNA	GGACGCGAGCATCTGTGGC	Reverse transcription for primer extension
ladder1	TCAGGTGGGCGACGGCGTCGGCGAGGCTCATGC GGACACTGTAGGCTGGAGCTGCTTC	Primer extension ladder
ladder2	GTGGTGGTGCTCAAGCGGCTCATCGCTACGACTT AGGTGAGCC	Primer extension ladder
ladder3	ATATCATATGTCGATCGACGTCAACAACG	Primer extension ladder
ladder4	CACTGACCAGGAGACTTTCGC	Primer extension ladder
ladder5	ATCGCCTCCGCGTCCACG	Primer extension ladder
HrdBF	CCGTTTCCATCGTTCCGAGA	Semi-quantitative RT-PCR
HrdBR	ATCTGCCCATCAGCCTTTCC	Semi-quantitative RT-PCR
JadAF	AATTCGACGCCTCCGAGATG	Semi-quantitative RT-PCR
JadAR	TCGAAGTCCTGGAGGTGGAA	Semi-quantitative RT-PCR
JadMF	GGGCACCCTCAACTACATCC	Semi-quantitative RT-PCR
JadMR	TCTTGAGCAGCTTCACGGAG	Semi-quantitative RT-PCR
JadJF	CGAAATCGCTGTCCGTGTG	Semi-quantitative RT-PCR
JadJR	GATAACTGGTCGCCGGAGTG	Semi-quantitative RT-PCR

Hpf UTRF	GACAGGAGTTCAGGGCGAAT	Semi-quantitative RT-PCR
Hpf UTRR	ATTACCGGAGCTCGGACTTG	Semi-quantitative RT-PCR
Hpf codingF	GAAAGCCTGCCGTTCCCA	Semi-quantitative RT-PCR
Hpf codingR	GACCTCGCCAGCGACAAG	Semi-quantitative RT-PCR



Supplementary Figure 1. Spore morphology and dormancy characteristics.
A. Scanning electron micrographs comparing wild-type strain (left), the Δ*rnj* strain (middle), and the Δ*rnc* strain (right) after growth for 6 days on MYM agar medium at 30°C. Bars = 2 µm.
B. Distribution of spore lengths for wild-type and mutant strains as determined using light microscopy images. Spore lengths were measured using ImageJ. For each strain, >1200 spores were measured, and calculated lengths were rounded to the nearest 0.01 µm.
C. Heat sensitivity of wild-type *S. venezuelae* and the RNase mutant strains. Spores were tested for the ability to survive heat shock at 50°C for the times indicated. Approximately 250 heat-shocked spores were spread on MYM agar plates and incubated at 30°C for 4 days. Their survival rates were then calculated as percentages. Each value is an average of three replicates, and the standard error was calculated for the percent survival at each time point.



Supplementary Figure 2. Analysis of the 5' end maturation of pre-16S rRNA and pre-23S rRNA.

A. Schematic representation of the rrnC and rrnF operons in *S. venezuelae*. The mature 16S, 23S and 5S rRNA sequences are indicated in dark grey, while in light grey are shown the leader, spacer 1 (S1) and spacer 2 (S2) sequences. The figure is to scale with respect to the length of each component of the rRNA operon. The scale bar below the rRNA operon shows the predicted start and stop coordinates of each mature rRNA species. The relative locations of oligonucleotides used for primer extension are shown (p16 for 16S and p23 for 23S).

B. Primer extension analysis of 16S and 23S processing. Primer p16 was extended to detect 16S 5' processing sites, while primer p23 was extended to detect 23S 5' processing sites. The extension products were separated on a 6% (w/v) denaturing polyacrylamide gel and radiolabelled oligonucleotides were used to create a ladder (M) with the expected sizes of 18 nt, 21 nt, and 29 nt. The location of the primary 16S and 23S processing sites are indicated on the left (16S) and right (23S) of the gels, relative to the promoter (+1) of the *rrnF* operon.

C. Secondary structure models of *S. venezuelae* pre-16S and pre-23S rRNA. Structures are based on M-fold analyses with modifications. Nucleotide numbers are as per the scale bar in (A). Predicted and experimentally determined processing sites are indicated with arrows, along with the RNase(s) responsible for cleavage. Black circles at the top of the 16S and 23S schematics represent the mature rRNA sequences. For the 16S structure, the sequence at the 5' end corresponds to the operon leader sequence, while the region at the 3' end corresponds to spacer 1 (S1). Similarly for the 23S structure, the 5' end sequence corresponds to spacer 1 (S1), and that at the 3' end corresponds to spacer 2 (S2), as indicated in (A).