

Expanded View Figures

Figure EV1. Loss of NAPs results in increases in RNA polymerase occupancy and decreases in overlapping EPODs.

- A Average RNA polymerase occupancy was calculated across intergenic regions within WT EPODs and background. Similar to Fig 2D, the blue dots denote the median and the black line displays the interquartile ranges in each condition. The dashed pink line represents the WT median. (*) indicate the Wilcoxon rank-sum *P* value comparing the change in median versus WT for each condition that has been adjusted using the Benjamini and Hochberg method (against a null hypothesis of no difference in pseudomedians). The gray line denotes the same comparison between the D.S. conditions. *P* value < 0.0005 = ***. Data were averaged across 2 (*hupAB*, *fis*, *stpA*, *ihf*, *dps*), 3 (WT, *hfq*, WT DS, *dps* DS), or 4 (*hns*, *stpA/hns*) biological replicates.
- B Similar to Fig 1D, reading left to right: overlap of the relaxed set of EPOD calls (left) over the stringent set of EPOD calls (bottom).

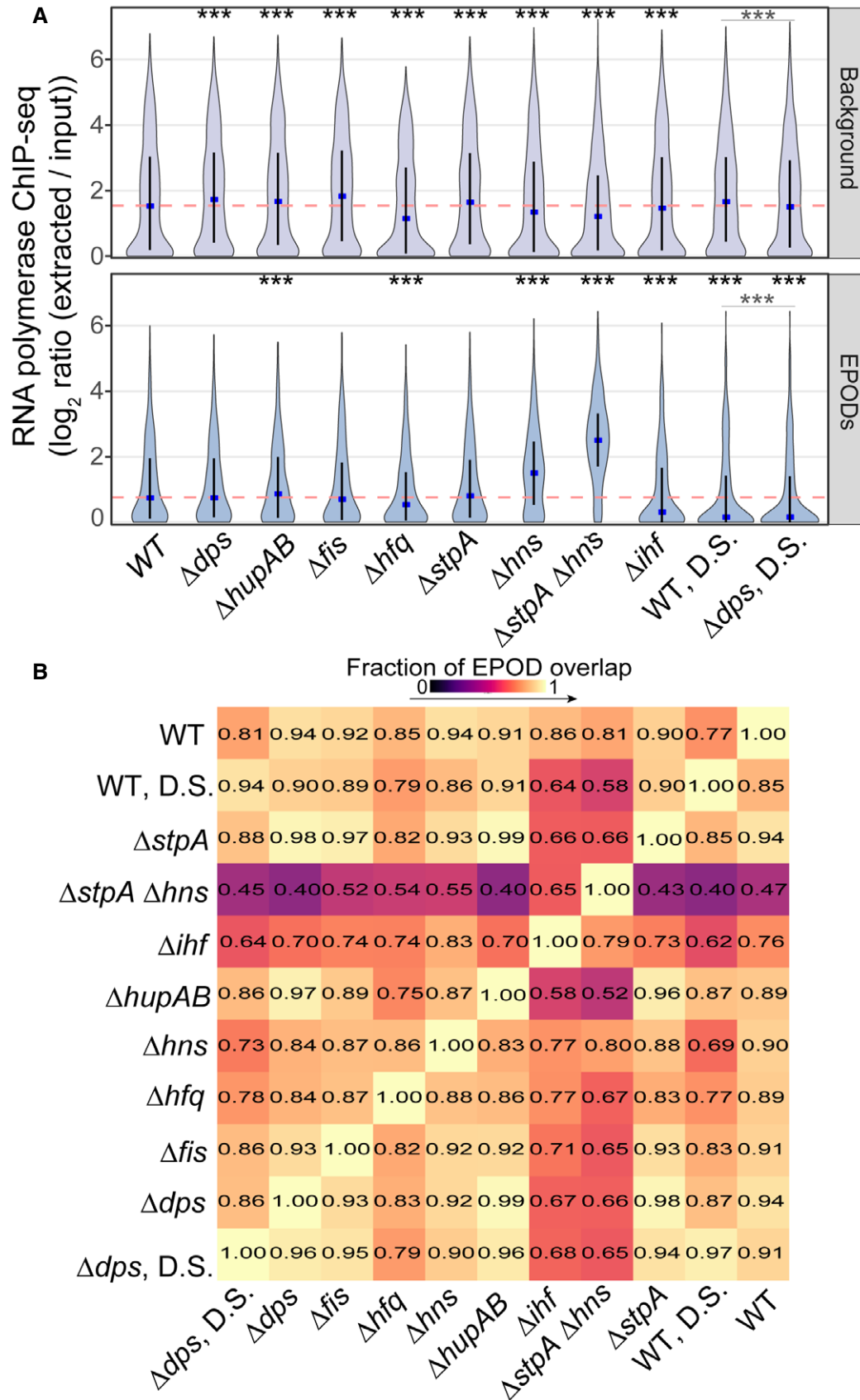


Figure EV1.

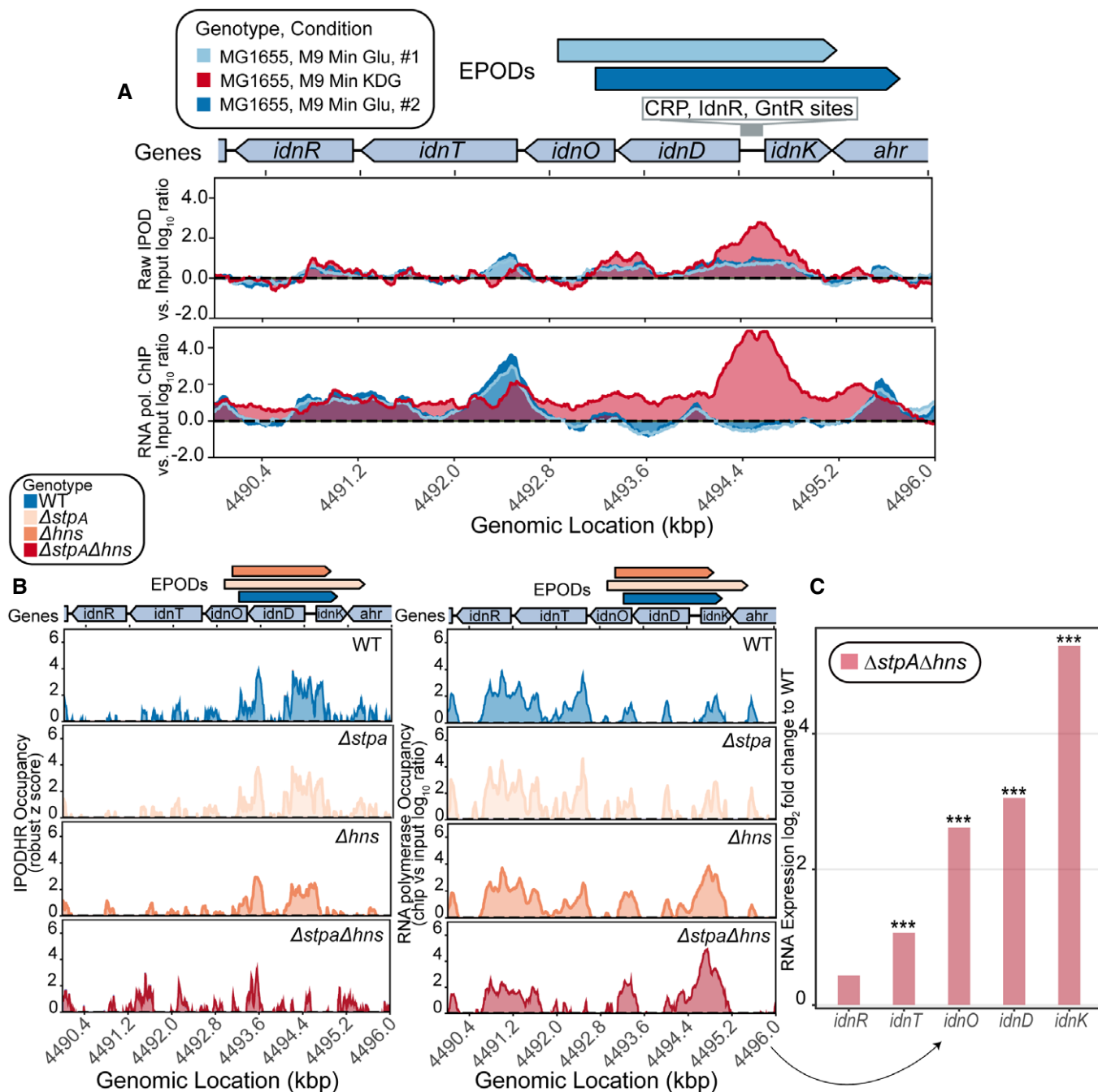


Figure EV2. H-NS and StpA mediate silencing of the *idn* operon.

A Raw IPOD and RNA polymerase ChIP-seq (both versus input log₁₀ ratios) over the *idn* operon.

B IPOD-HR was performed in rich defined medium (RDM) supplemented with glucose and exhibits a loss of occupancy in the deletion of both *hns* and *stpA*, as well as increased RNA polymerase occupancy.

C RNA-seq was performed in parallel and shows log₂ fold change compared to WT of *idn* operon expression upon deletion of *hns* and *stpA*. (*) indicate adjusted *P*-value < 0.0005 = *** (calculated using DeSeq2 as described in Methods).

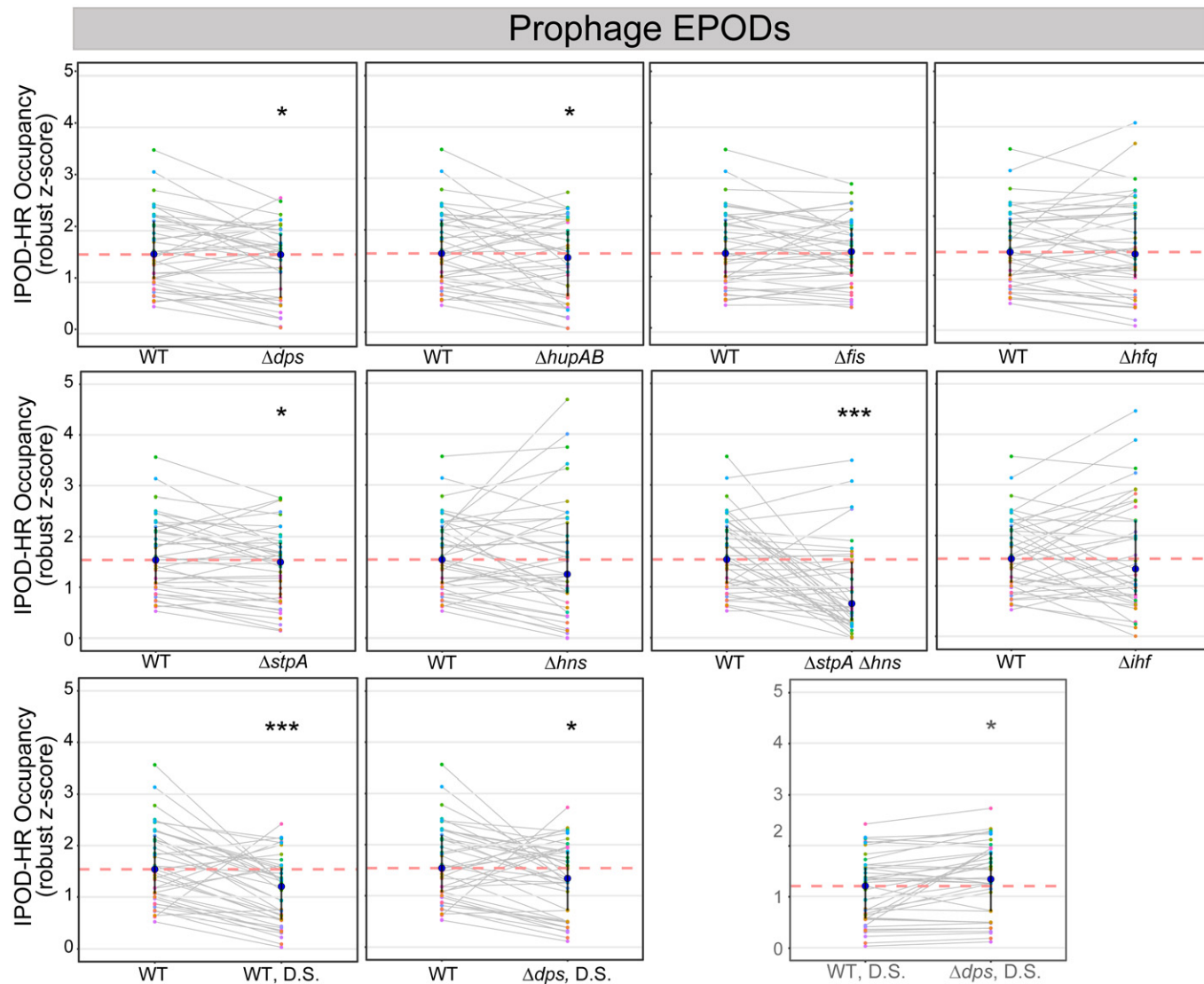


Figure EV3. Global protein occupancy across prophage EPODs changes with deletion of NAPs.

The average IPOD-HR occupancy (robust z scores) across each of the 41 prophage EPODs of WT and NAP deletions. Each colored dot represents an individual EPOD. Gray lines connect the same EPOD in WT (or WT D.S.) versus NAP deletion to appreciate the change in occupancy for that particular EPOD. The pink dashed line indicates the median of the WT comparison. The dark blue dot indicates the median of the genotype and transparent lines display the interquartile ranges. The summary of these data is displayed as violins in Fig 5B. (*) indicate the Wilcoxon rank-sum *P* value comparing the change in pseudomedian versus WT for each condition that has been adjusted using the Benjamini and Hochberg method (against a null hypothesis of no difference in pseudomedians). The gray line denotes the same comparison between the D.S. conditions. *P* value < 0.05 = *, < 0.0005 = ***.

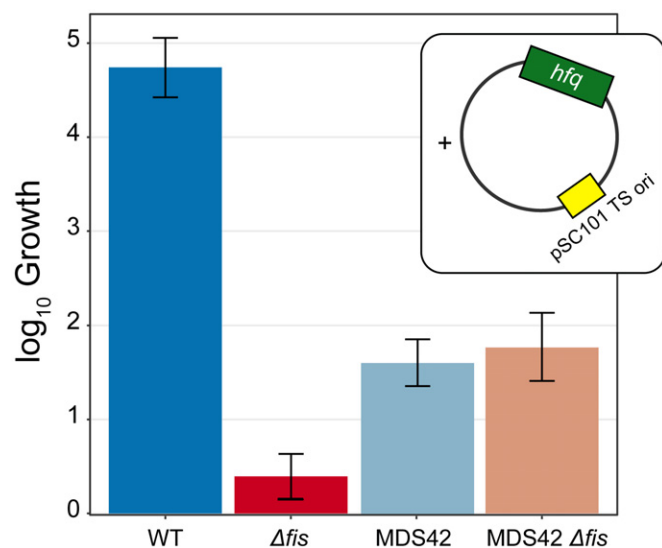


Figure EV4. Growth deficiency of $\Delta fis \Delta hfq$ cells.

WT, MDS42, Δfis , and MDS42 Δfis cells containing a temperature sensitive plasmid with *hfq* had their genomic copy of *hfq* deleted. Cells were grown in a permissive temperature (30°C), and then shifted to a non-permissive temperature for plasmid replication (42°C), thus removing *hfq* as the plasmid is dropped. The log₁₀ fold change in CFU is displayed. Δfis cells were unable to grow with the loss of the temperature sensitive *hfq* plasmid, however, upon deletion of mobile elements and prophages from the genome (MDS42 strain background), viability was restored. Data show fitted values plus 95% credible intervals for a Bayesian analysis, in which the plate counts themselves were treated as Poisson random variables, with rate parameters arising from an initial cell concentration in cells/ml (at the permissive temperature) and then a post-treatment scaling factor applied to that initial concentration. Both parameters were fitted on a log₁₀ scale, with a Uniform (0,15) prior on the initial cell concentrations and Uniform (-10,10) prior on the treatment effect. The treatment effect parameter is plotted as "log₁₀ growth", reflecting the observed growth at 42°C. Fits were based on three biological replicates for each genotype and were performed using pymc3 (Salvatier et al, 2016).

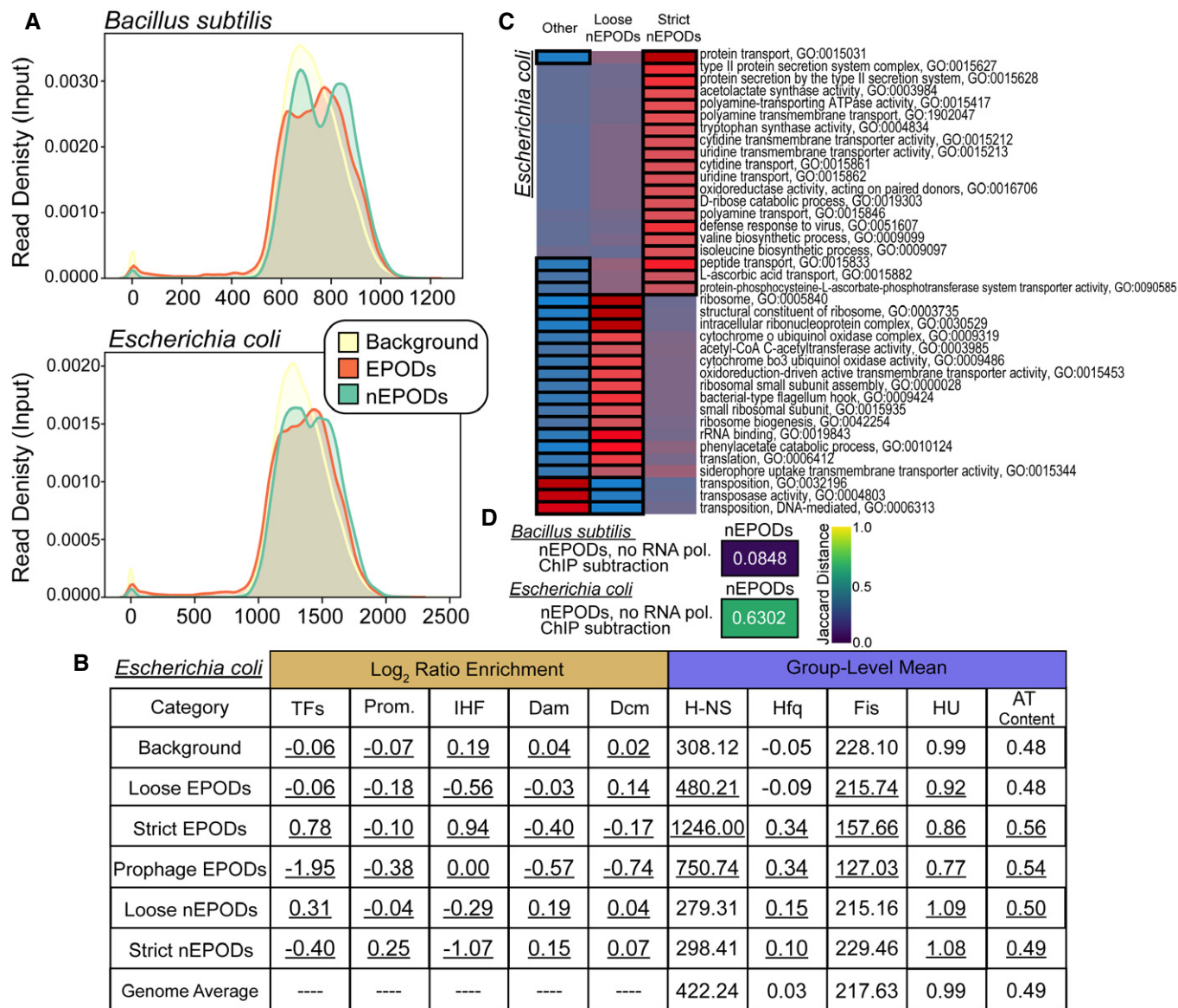


Figure EV5. nEPODs are a biological feature in *Bacillus subtilis*.

A Kernel density plots displaying the input read density *Bacillus subtilis* and *Escherichia coli* in regions called as EPODs (orange), nEPODs (green), and Background (yellow).

B Assessment of *Escherichia coli* nEPODs compared to other regions. Log₂ ratio enrichment: The ratio of the number of TFs, promoters, or motifs in a given category to the total number of TFs, promoters, or motifs. A chi-squared test was performed, and all categories were significantly associated with each class; values underlined had a *P*-value < 0.05. Group-level mean: The 500-bp rolling mean for the binding of each NAP was used to calculate the group-level means for across each category and compared with the overall average for the genome. Permutation based *P*-values were calculated comparing each class versus the background. The values underlined had a *P*-value < 0.05.

C *Escherichia coli* nEPODs overlap highly transcribed gene categories.

D Jaccard distance comparing nEPODs called with and without RNA polymerase ChIP-seq subtraction. The high Jaccard distance in *Escherichia coli* indicates that nEPODs change dramatically with and without the inclusion RNA polymerase binding, and therefore RNA polymerase associated. However, locations of nEPODs in *Bacillus subtilis* are unchanged with RNA polymerase binding and are distinct, protein occupied regions.