

Supplemental data for:

The Bacteriophage ϕ 29 Portal Motor can Package DNA Against a Large Internal Force

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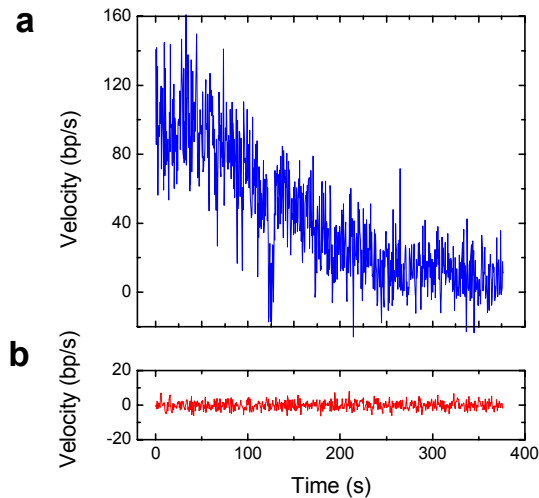
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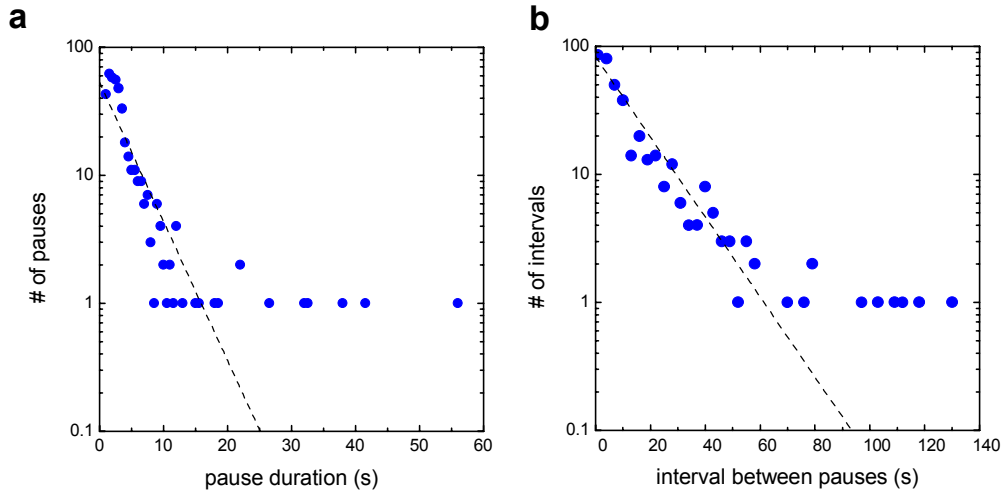
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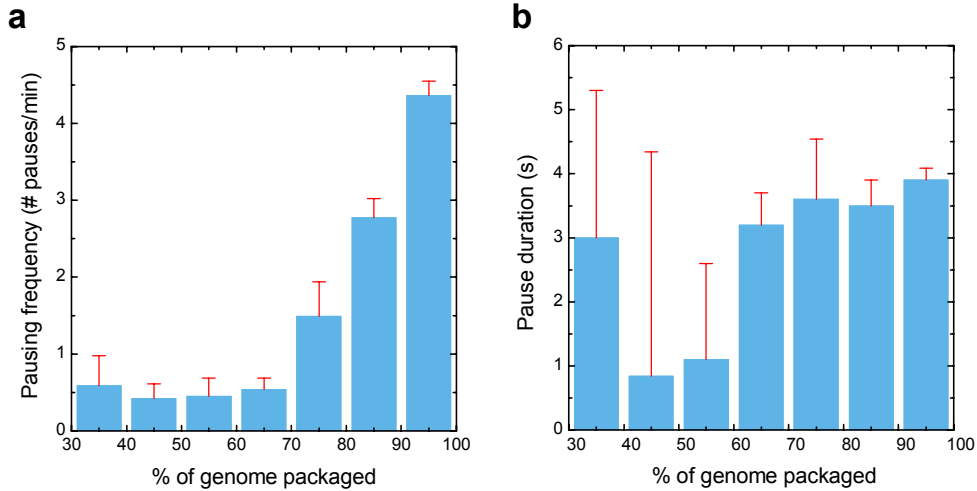
[¶] These authors contributed equally to this work.



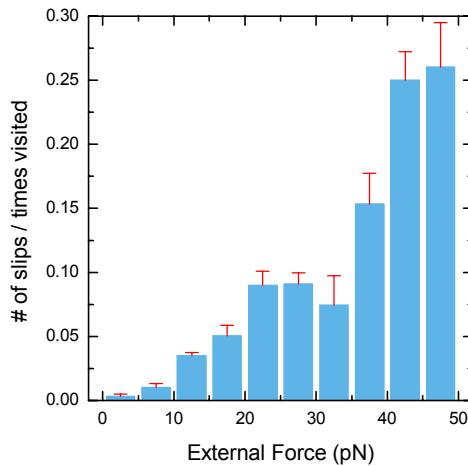
Supplemental figure 1a Velocity vs. time for a packaging complex held under a constant load of ~ 5 pN. Velocity was determined by linear fitting in a 1 s sliding window. These data correspond to the black line shown in Fig. 1b. **b**, To determine the experimental noise, velocity vs. time was also obtained for a 15 kb long DNA molecule that was directly attached between the two beads. The velocity fluctuations of the packaging complex are ~ 5 x larger than the measurement noise (~ 4 bp/s RMS in a 1 Hz bandwidth).



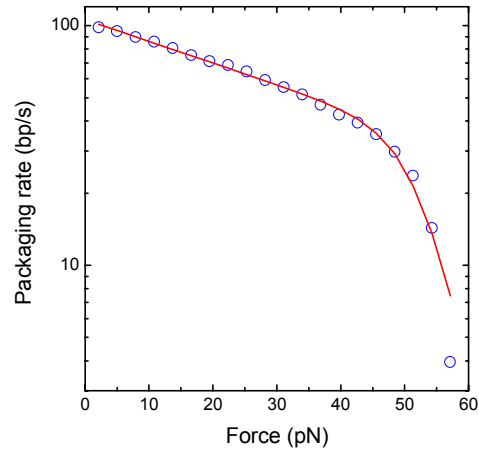
Supplemental figure 11a Histogram of pause duration. The dashed line corresponds to an interval distribution based on the mean pause duration ($Nw\lambda\exp(-\lambda t)$, where $N=425$ is the number of events, $w=0.5$ s is the bin size, and $\lambda=0.25$ is the reciprocal of the mean pause duration). This interval distribution is expected if pausing would be completely random. In this analysis, pauses were identified as plateaus in the length vs. time records, such as those shown in Fig 1b. During such plateaus the mean packaging rate was $1.8 \text{ bp/s} \pm 3.4 \text{ bp/s}$ and always $<10 \text{ bp/s}$. The average duration was 4 ± 5.1 s and always >0.5 s. In contrast, the mean packaging rate measured immediately before and after the pauses was significantly higher ($24 \pm 12 \text{ bp/s}$). **b**, Histogram of time intervals between pauses. The dashed line corresponds to the interval distribution ($Nw\lambda\exp(-\lambda t)$, where $N=384$ is the number of events, $w=3$ s is the bin size, and $\lambda=0.072$ is the reciprocal of the mean time interval). A simulation of a single time constant random process was also performed (data not shown) to investigate typical deviations from an ideal interval distribution (dashed lines) due to sample size. Comparison of the simulation to our data shows that the experimental points in these two plots are consistent with such a distribution, with the exception of a few of the events at the longest pause durations and intervals between pauses. For example, the probability of having three durations >35 s, as is the case in panel a (or three intervals >110 s in panel b), is $\sim 10^{-4}$.



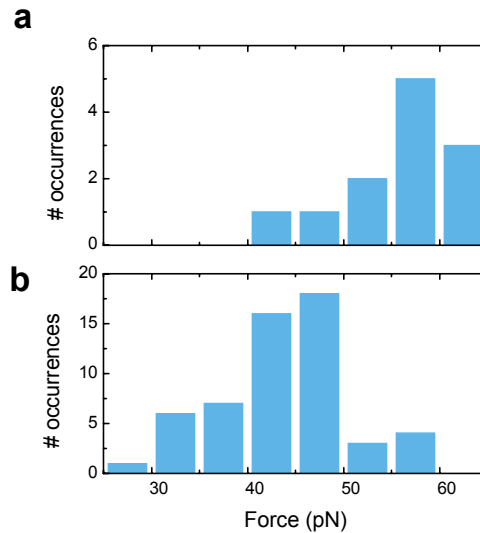
Supplemental figure IIIa Frequency of pausing vs. the amount of DNA packaged. Pauses appear to occur more often at higher filling levels. **b**, Pause duration vs. the amount of DNA packaged. Pause duration does not significantly increase at higher filling levels. These plots were composed from 425 pause events.



Supplemental figure IV Histogram of the forces at which slips occurred. These statistics were obtained from measurements in the 'no-feedback' mode (see Fig. 2, inset), where the external force increases. To account for the fact that many hook-ups broke before attaining the higher forces, the number of slips scored in each 5pN force window was divided by the number of times that force window was visited. The slip probability clearly increases with rising applied external force. This plot was composed from 141 slipping events.



Supplemental figure V Measured relation between applied force and packaging velocity (blue circles), and a fit to a simple model based on the Kramers relation (red line). In Kramers model, the rate (k) of a transition over a single reaction barrier induced by a force F is expressed as: $k = k_0 \exp[-(F\Delta x)/kT]$, where k_0 is the rate under zero load, and Δx the associated displacement. Here we consider a model with two force dependent steps: $1/k_t = 1/k_1 + 1/k_2$. For the step that is rate-limiting in the lower-force regime (k_1), the fit yields $k_0 = 106 \pm 0.3$ bp/s and $\Delta x = (0.1 \pm 0.002)$ nm. As seen in the graph, fitting is much less robust for the step that is rate-limiting at high forces (k_2), and is therefore not used in the interpretation in the main text. The fit values obtained here are: $k_0 = (1.5 \pm 3.1) \cdot 10^8$ bp/s and $\Delta x = (1.2 \pm 0.2)$ nm. The reduced chi-square value for the fit is 2.7 (15 degrees of freedom).



Supplemental figure VI Histograms of external forces measured during stalling for cases where 1/3 (a) and 2/3 (b) of the genome is packaged. Consistent with figure 3b in the main text, these figures show that upon going from 1/3 to 2/3 of the genome packaged the external force during stalling is reduced by ~ 14 pN. Fig. 3c in the main text shows the combined histogram after shifting the histogram for the 2/3 case by ~ 14 pN (internal force).