Supplementary Information (SI) to accompany

Millisecond curing time of a molecular adhesive causes velocity-dependent cargo-loading of molecular shuttles

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The Supplementary Information contains the following sections:

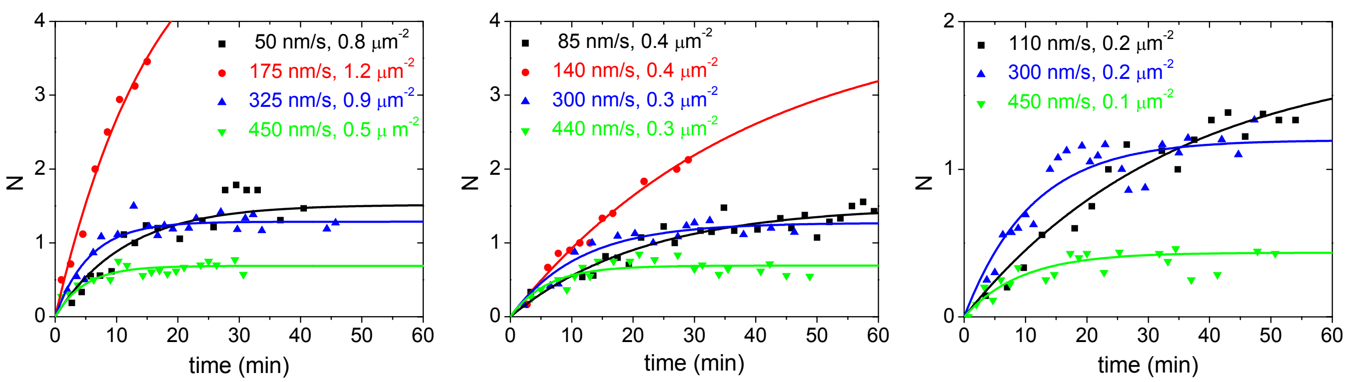
(1) **Supplementary Data** detailing the results obtained from microtubule loading experiments. **Supplementary Figure 1** illustrates the fits of the various datasets to the attachment and detachment rate equation. **Supplementary Table 1** lists the k_{on} and k_{off} values obtained from the fits and the associated $S_{MT\text{-}NS}$ values.

(2) **Supplementary Calculations:**

- Analytical model for the attachment and detachment process

- Calculations for the grasp of a nanosphere, g_{NS}

(1) Supplementary Data:



Supplementary Figure 1: Experimental Results. Observed nanosphere loading for various microtubule speeds and nanosphere densities as a function time elapsed since nanosphere injection. The data points are fitted to equation 1 (main text) to determine attachment and detachment rates.

v (nm/s)	σ (1600^{-1} μm^{-2})	k_{on} (min^{-1})	k_{off} (min^{-1})	$S_{MT\text{-}NS}$
50 ± 2	1364 ± 37	0.132 ± 0.019	0.087 ± 0.016	0.685 ± 0.064
175 ± 9	1900 ± 43	0.357 ± 0.036	0.061 ± 0.019	0.377 ± 0.067
325 ± 11	1375 ± 37	0.229 ± 0.032	0.178 ± 0.029	0.180 ± 0.028
452 ± 20	800 ± 28	0.143 ± 0.098	0.208 ± 0.166	0.139 ± 0.012
85 ± 3	675 ± 26	0.069 ± 0.007	0.046 ± 0.007	0.423 ± 0.133
140 ± 9	550 ± 23	0.105 ± 0.007	0.026 ± 0.006	0.479 ± 0.120
296 ± 10	510 ± 23	0.113 ± 0.014	0.089 ± 0.014	0.259 ± 0.096
442 ± 22	432 ± 21	0.114 ± 0.021	0.165 ± 0.035	0.209 ± 0.032
100 ± 4	272 ± 16	0.052 ± 0.005	0.029 ± 0.006	0.603 ± 0.086
297 ± 10	288 ± 17	0.109 ± 0.012	0.091 ± 0.014	0.446 ± 0.051
420 ± 23	238 ± 15	0.047 ± 0.011	0.108 ± 0.030	0.154 ± 0.023

Supplementary Table1: Results from microtubule loading experiments carried out at different microtubule velocities.

(2) Supplementary Calculations:

Analytical model for the attachment and detachment process.

The occupancies of states '3' and '2' and '1' (shown in Fig. 5(a) in the main text) can be calculated using the following relations:

$$\frac{dP_i}{dt} = P_{i+1}k_{i+1i} + P_{i-1}k_{i-1i} - P_i k_{ii+1} - P_i k_{ii-1}$$

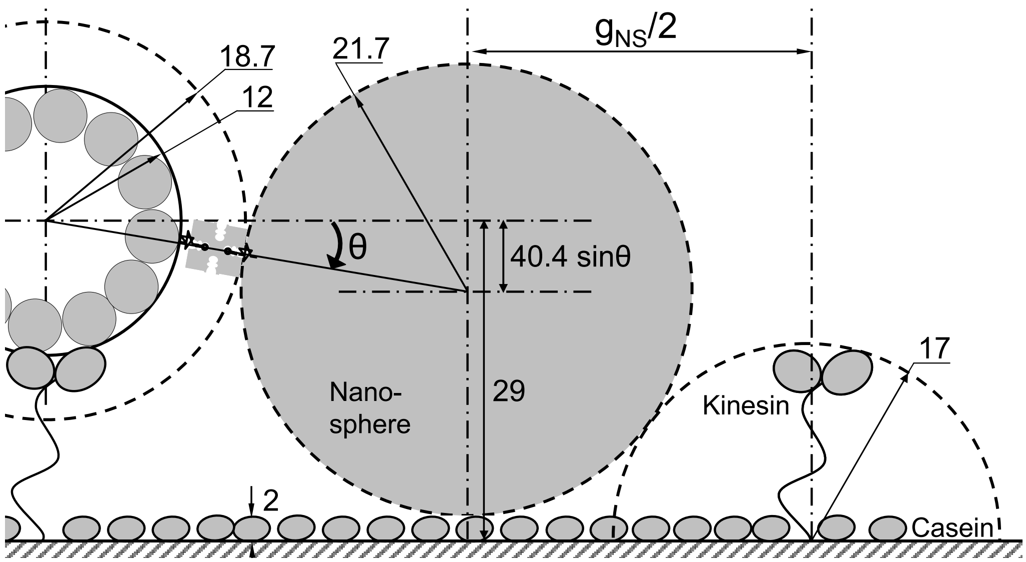
$$k_{ii+1} = \frac{\sqrt{\mathrm{K}_{mi}\mathrm{K}_{bi}}}{2\pi\zeta}\exp\{-(\frac{E_{bi}(F) - E_{mi}(F)}{k_B T})\}$$

$$k_{i+1i} = \frac{\sqrt{\mathrm{K}_{mi+1}\mathrm{K}_{bi}}}{2\pi\zeta}\exp\{-(\frac{E_{bi}(F) - E_{mi+1}(F)}{k_B T})\}$$

Where $k_{i\ i+1}$ is the transfer rate from state i to i+1, the subscripts *mi* and *bi* refer to the i^{th} metastable state and the i^{th} barrier respectively, K is the curvature of the local landscape, ζ is the damping coefficient and E, the depth/height of an energy minima/barrier. The values for K, ζ, and E were obtained from Pincet et. al.[19] State '3' for the attachment process and state '1' for the detachment process were assumed to initially contain 100% of the population.

The transfer rates depend on the shape of the potential energy surface which itself is altered in the presence of the force. Hence, P_{on}, the probability of filling up of state '1', was solved for as a function of force, F and time of contact, t_c. It was fitted to the experimental $S_{MT\text{-}NS}$ values after converting microtubule velocities to corresponding times of contact. The best fit was generated for the value of F as 53±5 pN. Similarly, P_{off} (the probability of emptying state '1') was calculated in the presence of an opposing force of 53 pN.

Grasp of a nanosphere, g_{NS}.



Supplementary Figure 2: Geometry of the microtubule-nanosphere-kinesin system during a detachment process.

A microtubule rotates about its longitudinal axis as it is propelled along its length by kinesin. Hence a nanosphere loaded onto a microtubule can be present anywhere along the circumference of a microtubule (θ can vary from 0 to 2π). Therefore, g_{NS} is calculated as a function of θ (Supplementary Fig. 2) and averaged over all angles:

$$\langle g_{NS} \rangle = \frac{\int g_{NS}(\theta) d\theta}{\int d\theta}$$

The calculation of $g_{NS}(\theta)$ is divided into three angular sections:

- When $13\pi/12 < \theta < 23\pi/12$, the nanosphere is raised by more than 17nm from the surface and the grasp is zero.

- For $23\pi/12 < \theta < \pi/24$ and $23\pi/24 < \theta < 13\pi/12$, the geometry of the system provides:

$$g_{NS}(\theta) = 2 * \sqrt{(38.7)^{\wedge 2} - (29 - 40.4\sin\theta)^{\wedge 2}}\, nm$$

- For $\pi/24 < \theta < 23\pi/24$, the nanosphere touches the impenetrable 2 nm thick casein layer, yielding a grasp of 61 nm. It is assumed that the microtubule has to deform while it is being rotated and propelled to allow the nanosphere cargo to remain attached to it.

Combined, the average value of the grasp of a nanosphere for a kinesin $\langle g_{NS} \rangle$ is determined to be 33 nm.