## Supplementary Information for

## **Depletion Force Induced Collective Motion of**

## **Microtubules Driven by Kinesin**

Daisuke Inoue<sup>a</sup>, Bulbul Mahmot<sup>b</sup>, Arif Md. Rashedul Kabir<sup>a</sup>, Tamanna Ishrat Farhana<sup>c</sup>,

Kiyotaka Tokuraku<sup>d</sup>, Kazuki Sada<sup>a,c</sup>, Akihiko Konagaya<sup>b</sup> and Akira Kakugo<sup>a,c</sup>\*

<sup>a</sup>Faculty of Science, Hokkaido University, Sapporo, 060-0810, Japan

<sup>b</sup>Department of Computational Intelligence and Systems Science, Tokyo Institute of Technology, Yokohama, 226-8501, Japan

<sup>c</sup>Graduate School of Chemical Sciences and Engineering, Hokkaido University,

Sapporo, 060-0810, Japan

<sup>d</sup>Department of Applied Sciences, Muroran Institute of Technology, Muroran, 050-8585,

Japan

\* Corresponding author. E-mail: kakugo@sci.hokudai.ac.jp Telephone/FAX: +81-11-706-3474



**Fig. S1** Analysis method of the angles and degree of orientation of microtubules. (A) Fluorescent microscopic image of microtubules (left), and color map representing the distribution of microtubule orientation angles (right). (B) Circular histogram of orientation angles of microtubules obtained from the fluorescent microscopic image of microtubules in (A). (C) (D) (E) Directional analysis method. Here each bin in the circular histogram is considered as a vector (length:  $R_i$ , angle:  $\Theta_i$ ). (D) Nematic order parameter, S is calculated as the size of average vector (length: S, mean orientation angle  $\langle \theta_2 \rangle$ ) was used for evaluation of orientation direction and degree of orientation. (E) Relationship between the S and degree of orientation. If distribution of microtubules angles is dispersed, S becomes smaller ( $S_{dispersed}$ ) than that of highly oriented microtubules ( $S_{oriented}$ ).



**Fig. S2** Comparison of fluorescence images of microtubule streams observed under (left) confocal and (right) stimulated emission depletion (STED) microscope.



Fig. S3 Correlation between the number of microtubules in streams determined by a STED image and average fluorescence intensity obtained from a confocal image. The correlation curve is estimated by:  $f(x) = ax^2 + bx + c$ , with a = -0.045, b = 6.2, c = -4.5



Fig. S4 Distribution of estimated microtubule numbers at the time 90 min in microtubule gliding assay with MAP4 fragment (A) and without MAP4 fragment (B). Yellow: higher density, Blue: lower density. Density fluctuation,  $D_f$  is calculated by the distribution of microtubule density or the numbers of microtubules per tiles covering a movie frame.



Fig. S5 Disappearance of the stream of collectively moving microtubules in the presence of 0.3 wt% MC without MAP4 fragment. Scale bar: 50  $\mu$ m



Fig. S6 Effect of concentration of MAP4 fragment in the presence of 0.3 wt% MC on morphology of collectively moving microtubules. Scale bar:  $50 \ \mu m$ 



**Fig. S7** Effect of MAP4 fragment and MC on the behaviour of gliding microtubules. Probability of snuggling was evaluated in the presence or absence of MAP4 fragment without and with 0.3 wt% MC. Kinesin concentration was fixed at 1000 nM. '+' and '-' indicate 'presence' and 'absence', respectively.



Movie. 1 Emergence of collective motion of microtubules in the presence of 0.3 wt% MC without MAP4 fragment. Density of microtubules is  $65 \times 10^4$  filaments mm<sup>-2</sup>. Scale bar: 50  $\mu$ m