SUPPLEMENTARY FIGURE LEGENDS

Fig. S1. Colony formation assay. CHO cells were seeded into 24-well dishes containing the indicated concentrations of colcemid (A) or vinblastine (B). The cells were grown for 7 d and stained with 0.05% methylene blue.

Fig. S2. Drug inhibition of cell migration. CHO cells were seeded into 6 well dishes and grown until they reached approximately 80% confluence. A scratch was made at the center of the plate. The distance between the leading edges of the cell monolayers was measured as a function of time in the presence and absence of 2 nM vinblastine. Brightfield images of the cells are shown. Bar = 100 μm.

Fig. S3. Microtubule fragment formation. The indicated cell lines were treated with vinblastine for 1 h, fixed, and stained for tubulin (green) and DNA (red). The drug concentration used was twice the IC_{50} for inhibition of cell division in each of the cell lines. Similar results were obtained with other microtubule active drugs. Note that the drug effects are rapid and that microtubule fragments are found predominantly near the nuclear area at these early time points. Bar = 10 μm. Inserts show 1.5 fold enlargements of a portion of the cell to more easily point out the microtubule fragments (arrowheads); bar = 5 μm.

Fig. S4. Microtubule dynamics in mitotic cells. CHO cells were transfected with EGFP-MAP4, grown overnight, and examined by time-lapse fluorescence microscopy at 37 ˚C 30 min after addition of vinblastine. Prophase cells were located and images were taken every 5 sec. The microtubule length, measured from an arbitrary fixed point, was plotted against time to generate life history plots. Each line represents a different microtubule. The drug concentrations were chosen to be below (5 nM) and above (35 nM) the concentrations that inhibit cell division. Note that mitotic microtubules undergo more frequent excursions of growth and shortening compared to interphase cells, but their dynamic behavior is still suppressed by drug concentrations below those needed to inhibit mitosis.

Fig. S5. Drug sensitivity of mitotic versus interphase cells. *A.* PtK2 cells were treated with their IC_{50} concentration of vinblastine (17 nM) for 1 h and stained for tubulin immunofluorescence. Note that a prophase cell (upper left) has highly fragmented microtubules while a nearby interphase cell (lower right) is only beginning to show drug effects. Bar = 10 μm. *B.* Cell proliferation (●), the percent of prophase cells with fragmented microtubules (□), and the percent of interphase cells with fragmented microtubules (O) were measured at a series of vinblastine concentrations. The results indicate that microtubule fragments are generated in prophase cells at a lower drug concentration than in interphase cells.

Video 1. Untreated CHO cells migrating into a wound. Cells at the edge of a scratch wound (located to the right) were photographed every 15 min using a 10 X objective. A movie was made using 21 successive frames (encompassing 5 h) shown for 0.2 s each.

Video 2. Vinblastine treated CHO cells migrating into a wound. The conditions were identical to video 1 except the wound is to the left and the cells were treated with 4 nM vinblastine for 4 h prior to the sequence that is shown.

TABLE S1
Effects of Vinblastine on CHO microtubule dynamics

	0	0.05	0.1	0.5	1	2.5	15 nM
Growth							
Rate (μm/min)	16.2 ± 0.8	16.0 ± 1.2	14.9 ± 1.8	12.6 ± 0.8*	11.8 ± 0.6*	11.6 ± 0.7*	10.2 ± 0.7*
Distance (μm)	3.2 ± 0.4	2.0 ± 0.2*	2.0 ± 0.3	1.6 ± 0.3*	1.4 ± 0.1*	1.2 ± 0.2*	1.1 ± 0.2*
Duration (sec)	12.2 ± 1.2	7.2 ± 0.3*	7.8 ± 0.9*	7.4 ± 0.9*	7.0 ± 0.5*	6.1 ± 0.6*	6.3 ± 0.4*
Shortening							
Rate (μm/min)	28.2 ± 2.3	20.4 ± 1.3*	17.3 ± 1.5*	17.3 ± 1.2*	14.5 ± 1.0*	13.3 ± 1.2*	10.8 ± 0.8*
Distance (μm)	5.2 ± 0.6	3.2 ± 0.4*	2.5 ± 0.4*	2.4 ± 0.6*	2.0 ± 0.3*	1.6 ± 0.2*	1.3 ± 0.1*
Duration (sec)	11.2 ± 1.1	8.9 ± 0.8	8.0 ± 0.7	7.3 ± 0.7*	7.4 ± 0.7*	7.5 ± 0.6*	6.7 ± 0.5*
% of time in							
Growth	31.1 ± 2.2	23.7 ± 2.1	15.6 ± 2.6*	20.3 ± 2.1*	17.0 ± 1.9*	10.4 ± 1.4*	11.4 ± 1.9*
Shortening	21.1 ± 1.9	20.6 ± 2.0	23.1 ± 3.3	16.5 ± 2.0	16.0 ± 2.1	11.8 ± 1.9*	13.1 ± 1.7*
Pause	47.8 ± 3.1	55.7 ± 3.5	61.4 ± 5.1*	63.2 ± 3.2*	67.0 ± 3.2*	77.8 ± 2.5*	75.6 ± 3.3*
Frequency (min^{-1})							
Catastrophe [1]	1.5 ± 0.1	1.9 ± 0.2	2.3 ± 0.3*	1.8 ± 0.2	1.6 ± 0.2	1.2 ± 0.2	1.4 ± 0.2
Rescue [2]	6.3 ± 0.5	7.0 ± 0.5	7.2 ± 0.8	9.2 ± 0.6*	8.3 ± 0.8*	8.5 ± 0.7*	9.1 ± 0.7*
Dynamicity[3] (μm/min)	9.0 ± 0.9	8.6 ± 1.1	6.8 ± 1.3	5.7 ± 0.9*	4.5 ± 0.5*	2.6 ± 0.3*	2.8 ± 0.5*
Number microtubules	30	15	10	15	15	15	15

Values shown represent the mean ± s.e.m.
[1] Catastrophe: the transition from growth or pause to shortening.
[2] Rescue: the transition from shortening to growth or pause.
[3] Dynamicity: the total change of length per unit time of individual microtubules during their life histories.
* $p < 0.05$ compared to 0 concentration.

TABLE S2
Effects of Colcemid on CHO microtubule dynamics

	0	0.01	0.3	1	3	7	50 nM
Growth							
Rate (μm/min)	16.2 ± 0.8	15.0 ± 1.2	14.1 ± 1.0	13.0 ± 1.1*	11.0 ± 0.7*	10.1 ± 0.5*	12.0 ± 0.8*
Distance (μm)	3.2 ± 0.4	2.0 ± 0.2*	1.6 ± 0.1*	1.5 ± 0.2*	1.2 ± 0.1*	1.1 ± 0.1*	1.3 ± 0.1*
Duration (sec)	12.2 ± 1.2	7.8 ± 0.5*	7.3 ± 0.6*	6.6 ± 0.5*	6.5 ± 0.4*	6.7 ± 0.4*	6.6 ± 0.5*
Shortening							
Rate (μm/min)	28.2 ± 2.3	21.9 ± 1.7	16.6 ± 1.5*	15.4 ± 1.9*	15.7 ± 1.0*	14.2 ± 1.5*	12.5 ± 0.8*
Distance (μm)	5.2 ± 0.6	3.2 ± 0.4*	2.3 ± 0.3*	1.9 ± 0.3*	1.9 ± 0.2*	1.5 ± 0.1*	1.6 ± 0.2*
Duration (sec)	11.2 ± 1.1	8.3 ± 0.6	8.4 ± 0.6	7.3 ± 0.5*	7.1 ± 0.5*	6.6 ± 0.4*	7.5 ± 0.6
% of time in							
Growth	31.1 ± 2.2	22.13 ± 2.2*	17.3 ± 2.9*	10.3 ± 1.4*	12.0 ± 1.4*	13.3 ± 2.0*	10.3 ± 1.1*
Shortening	21.1 ± 1.9	20.3 ± 2.0	23.1 ± 2.8	16.6 ± 1.7	15.7 ± 0.9*	12.7 ± 1.3*	14.8 ± 2.2
Pause	47.8 ± 3.1	57.4 ± 3.6	59.6 ± 5.1*	73.1 ± 2.7*	72.3 ± 1.6*	72.5 ± 2.9*	75.4 ± 2.4*
Frequency (min^{-1})							
Catastrophe [1]	1.5 ± 0.1	1.9 ± 0.2	2.3 ± 0.3*	1.7 ± 0.2*	1.7 ± 0.2	1.4 ± 0.2	1.5 ± 0.2
Rescue [2]	6.3 ± 0.5	7.2 ± 0.7	7.0 ± 0.7	8.4 ± 0.7*	8.2 ± 0.7	9.5 ± 0.5*	8.6 ± 0.5*
Dynamicity[3] (μm/min)	9.0 ± 0.9	8.1 ± 1.0	6.9 ± 0.6	4.0 ± 0.7*	3.8 ± 0.4*	2.9 ± 0.3*	3.1 ± 0.5*
Number microtubules	30	16	12	15	10	10	11

Values shown represent the mean ± s.e.m.
[1] Catastrophe: the transition from growth or pause to shortening.
[2] Rescue: the transition from shortening to growth or pause.
[3] Dynamicity: the total change of length per unit time of individual microtubules during their life histories.
* $p < 0.05$ compared to 0 concentration.

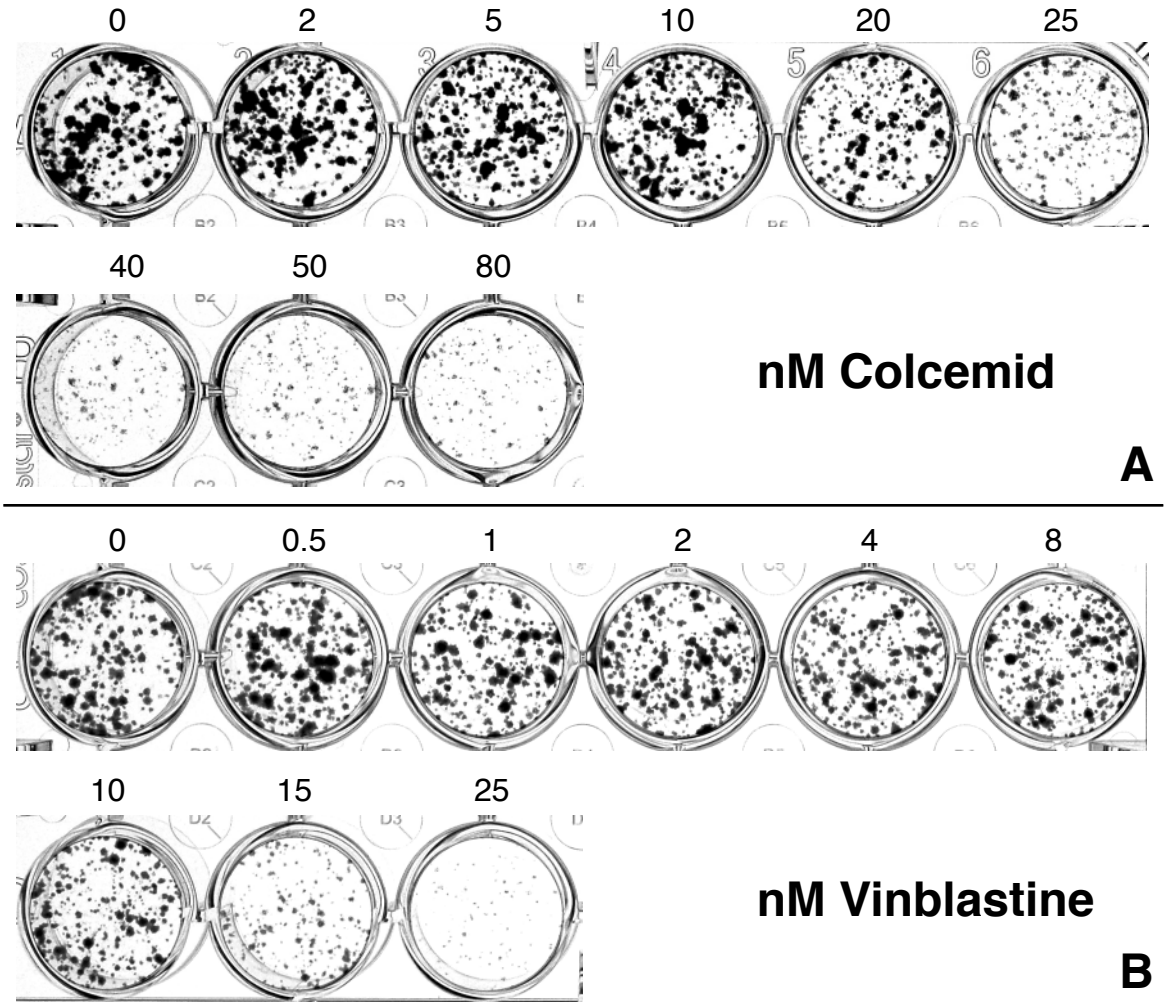

0
2
5
10
20
25
40
50
80
nM Colcemid
A
0
0.5
1
2
4
8
10
15
25
nM Vinblastine
B

Figure S1

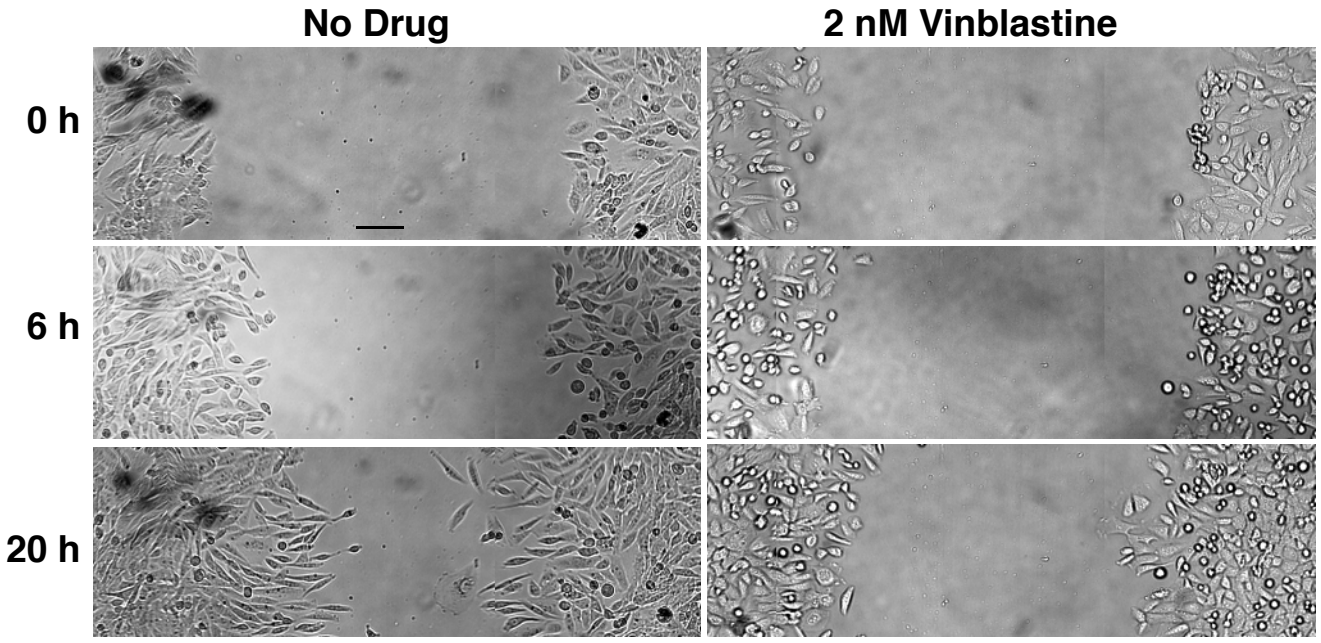


Figure S2

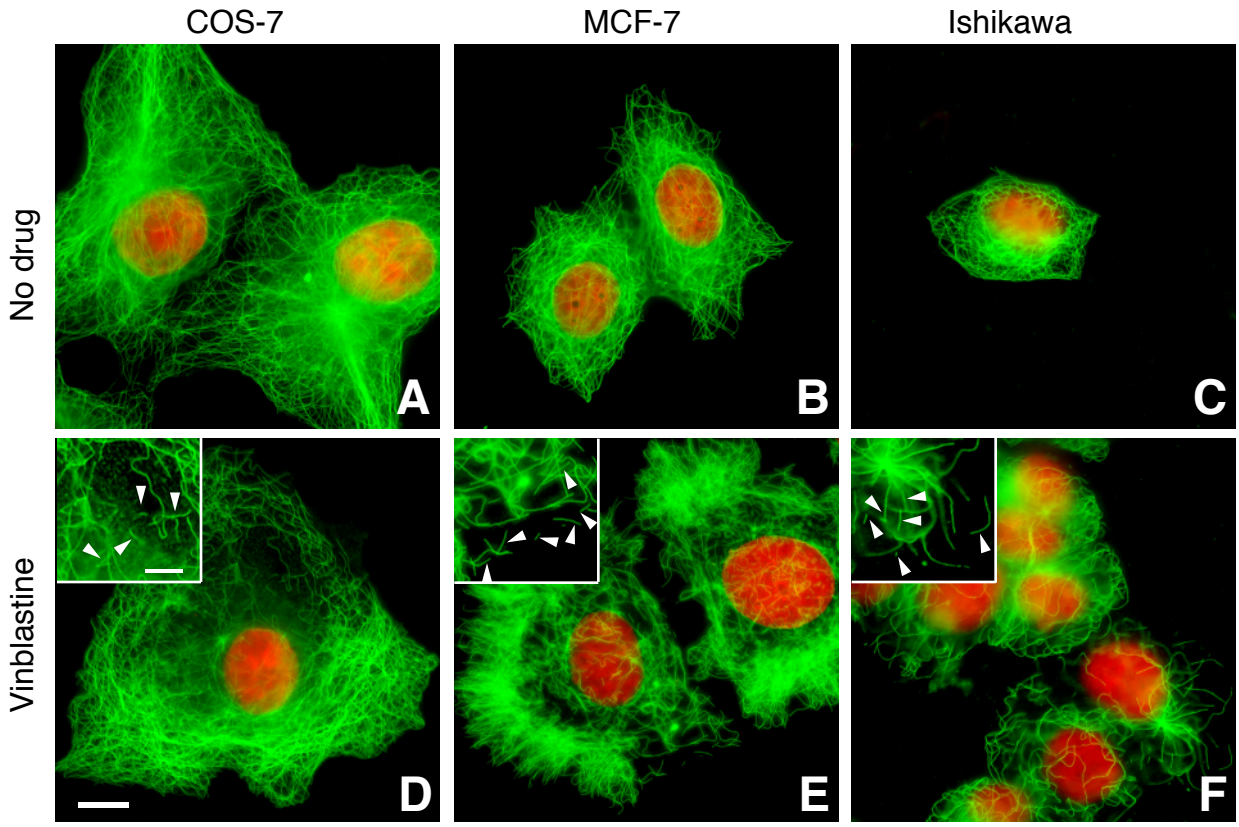


Figure S3

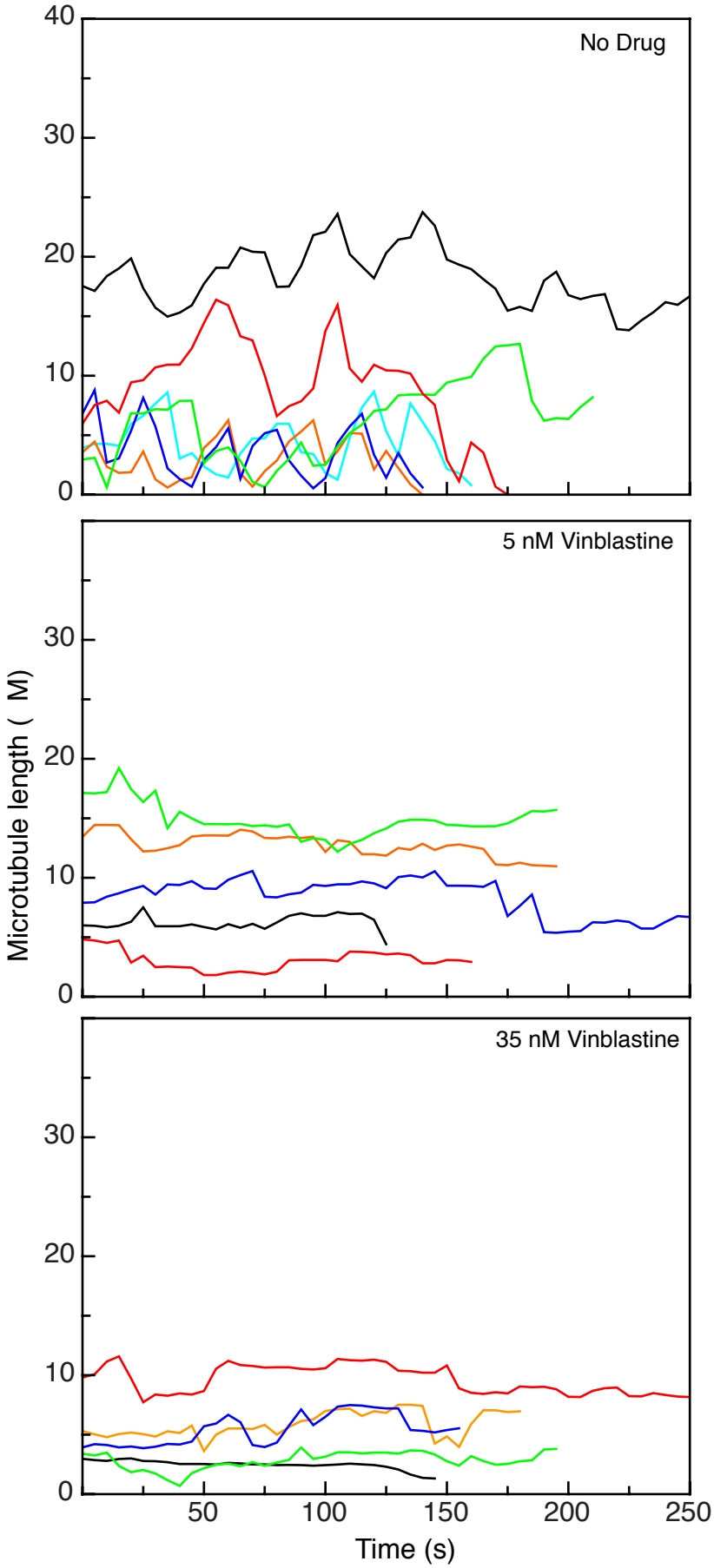
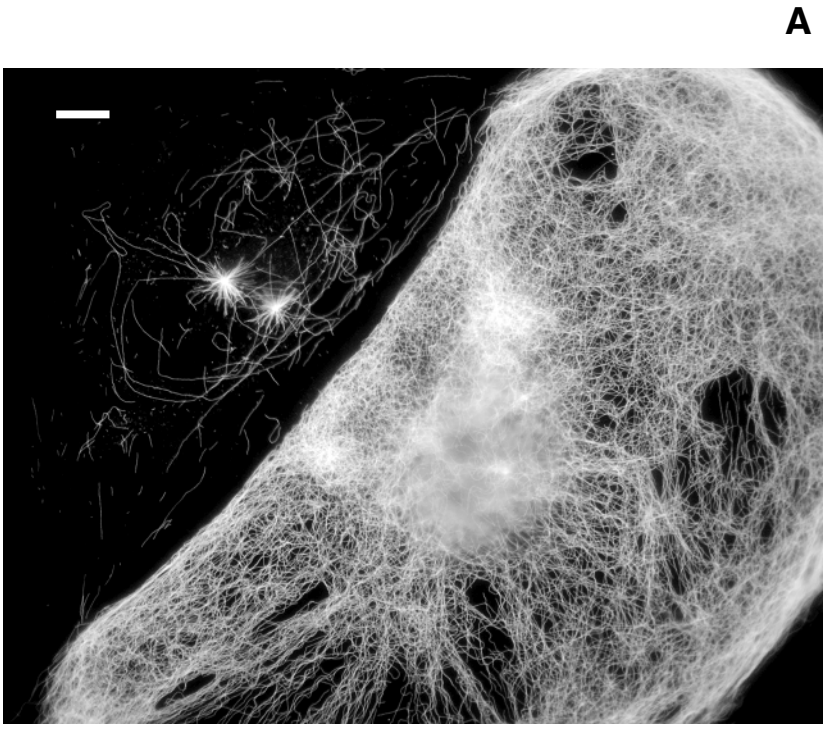

No Drug
5 nM Vinblastine
35 nM Vinblastine
Microtubule length (M)
Time (s)
0
10
20
30
40
50
100
150
200
250

Figure S4


A

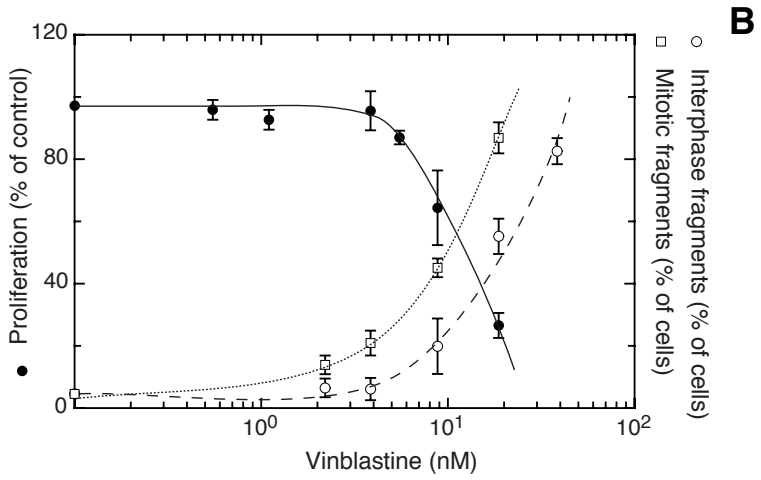

B
120
80
40
0
Proliferation (% of control)
Mitotic fragments (% of cells)
Interphase fragments (% of cells)
10^0
10^1
10^2
Vinblastine (nM)

Figure S5