SUPPORTING INFORMATION

Measurement of streptavidin density on the surface of droplets. To estimate the density of streptavidin on the surface, we proceed as follows. First, we use the adsorption of fluorescently labelled streptavidin onto solid polystyrene particles with known surface area to make a calibration plot that relates fluorescence intensity measured at the surface of a particle to the amount of adsorbed streptavidin molecules per m^2. We then use this calibration plot to estimate the density of streptavidin on the oil/water interface from the fluorescence intensity.

Samples of 20 μL with different concentrations of Alexa Fluor 633-labelled streptavidin (0.2nM, 1nM, 1.6nM, 5nM, 10nM and 100nM) were prepared. To each streptavidin solution 1 μL of washed 6 μm polystyrene particles was added (total area 13 mm^2 in each). After incubation for 2 hours at room temperature, fluorescence intensity on particles' surface was measured in the confocal microscope (at identical detector settings). For the lowest streptavidin concentrations, it was necessary to increase the laser transmission and the detector integration time. It was checked that the measured intensity increases linearly with laser transmission and integration time, so that we could normalize the intensity to correct for this.

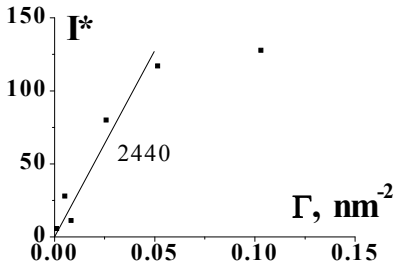

Figure S1. Normalized fluorescence intensity as a function of the amount of streptavidin per unit area.

Figure S1 shows the normalized fluorescence intensity as a function of the number of streptavidin molecules available per nm^2 of bead area. Initially, the intensity increases approximately linearly with the available streptavidin. At higher streptavidin dosages, the intensity levels off towards a plateau. We assume that the plateau corresponds to a dense monolayer of streptavidin. It is reasonable to assume that in the initial part of the curve nearly all streptavidin is bound to the particle surface. This means that the initial slope of the curve can be used to relate the fluorescence intensity to the number of streptavidin molecules on the surface per nm^2. This calibration is then used to estimate the streptavidin density on the oil droplets from the measured fluorescence intensities. Figure S2 shows the resulting streptavidin surface density as a function of the total concentration in the sample. The density increases linearly with increasing amount of streptavidin in the sample, which suggests that all streptavidin is bound to the interface.

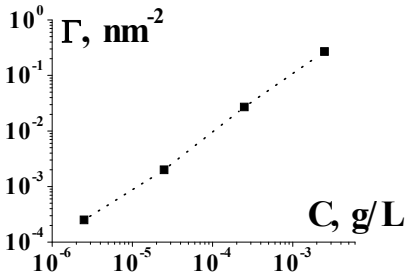

Figure S2. Streptavidin density on the oil/water interface (obtained from fluorescence intensity) as a function of the total streptavidin concentration in the sample.

Clustering of lipids and streptavidin. Biotinylated lipids showed a tendency to cluster on the oil/water interface. This causes inhomogeneous distribution of streptavidin and streptavidin-modified particles on the interface as shown in Fig. S3. Adding unsaturated lipids to the mixture could prevent the clustering. As shown in Fig. 3D-E, no clustering occurred and the layers were completely homogeneous, even for relatively high lipid concentrations.

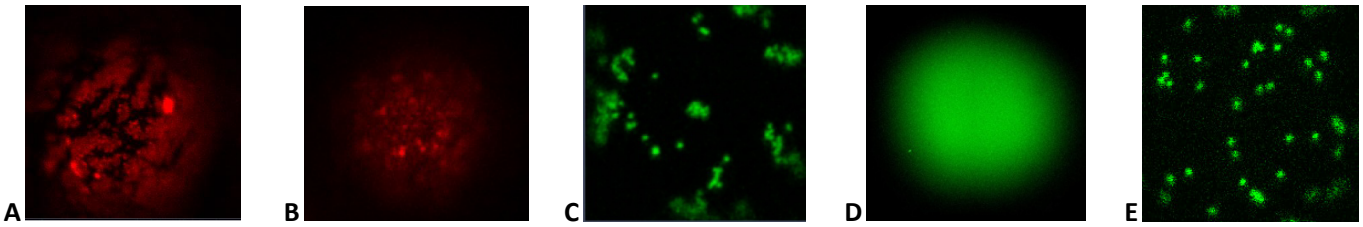

Figure S3. Examples of clustered (due to lipid clustering) and well-separated components. Oil with biotinylated lipids only (0.042 g/L): streptavidin clustering (A), streptavidin clustering on droplets made of sonicated oil (B), and clustered particles (C). Oil with lipid mixture (1 g/L 22:6 Cis PC and 0.25 g/L biotinylated lipids): homogeneous streptavidin layer (D) and well-separated particles suitable for tracking (E). Average droplet diameter is 20 μm).

The effect of streptavidin on binding of actin. In order to assess the effect of different concentrations of streptavidin on the amount of actin bound to the droplets' interface, intensity of Alexa-Fluor 597-labelled actin at the interface was studied for three concentrations of streptavidin. As shown in Fig. S4A, fluorescence intensity of labelled actin increases with the amount of streptavidin, by approximately 50% as the streptavidin density increases 100-fold, indicating that a higher streptavidin density on the surface results in an increase of the number of bound actin filaments due to increased amount of binding sites on a droplet's interface. It is most likely that the increase in streptavidin signal at low concentrations in Fig. S4B is due to cross-talk between the channels (actin's signal bleeds through and interferes with the streptavidin signal thus increasing it. This effect becomes negligible at higher streptavidin concentrations).

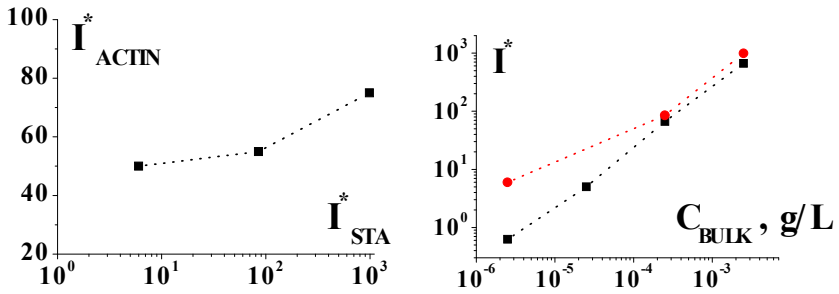

Figure S4: (A): Effect of the amount of surface-bound streptavidin on the fluorescence intensity of labelled actin on the interface. (B): Streptavidin binding isotherm on droplets containing biotinylated lipids, in the absence (■) and in the presence (●) of actin.

The effect of gelsolin on the length of actin filaments.

Actin was polymerized as described in the Materials and methods section, with varying amounts of gelsolin. The length distribution was measured with AFM as described. Figure S5 shows a typical AFM image showing actin filaments. From these images, contour lengths were measured to obtain a length distribution. Figure S6 shows the length distribution obtained in this way for different gelsolin/actin ratios R.

The table below shows the weight and number-averaged length, L_W and L_N respectively for the different R, (also plotted in the inset of figure 6A).

R	L_W	L_N	PDI	Number of lengths
0	4.73	3.33	1.42	480
0.001	2.95	1.46	2.03	840
0.01	2.29	1.12	2.04	1270
0.1	0.50	0.37	1.35	430

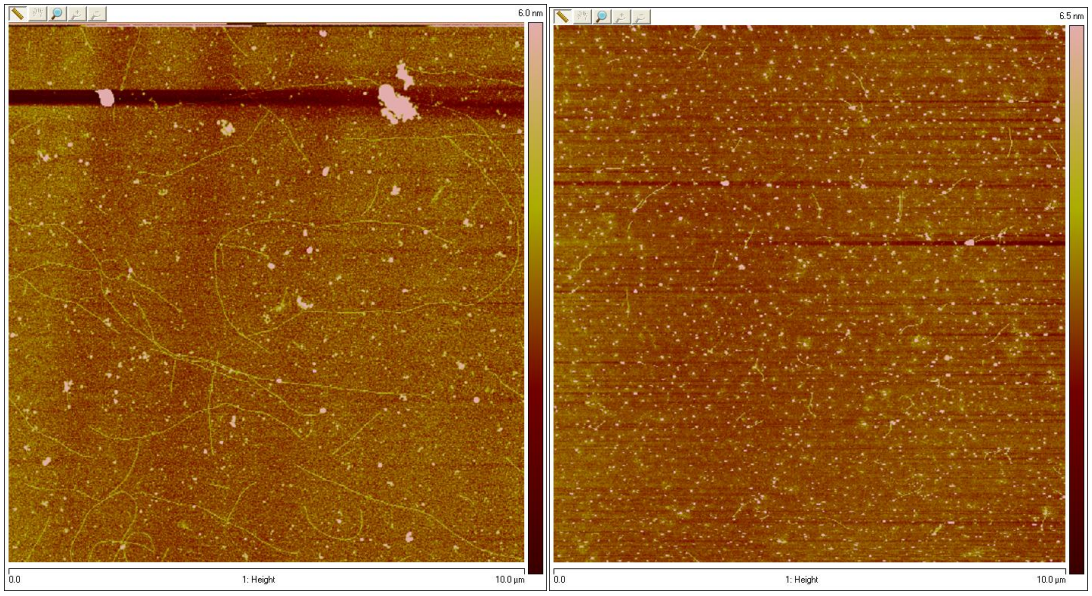

Figure S5: AFM pictures of untreated actin filaments (left, R=0) and treated with gelsolin (right, R=0.1, the highest ratio studied)

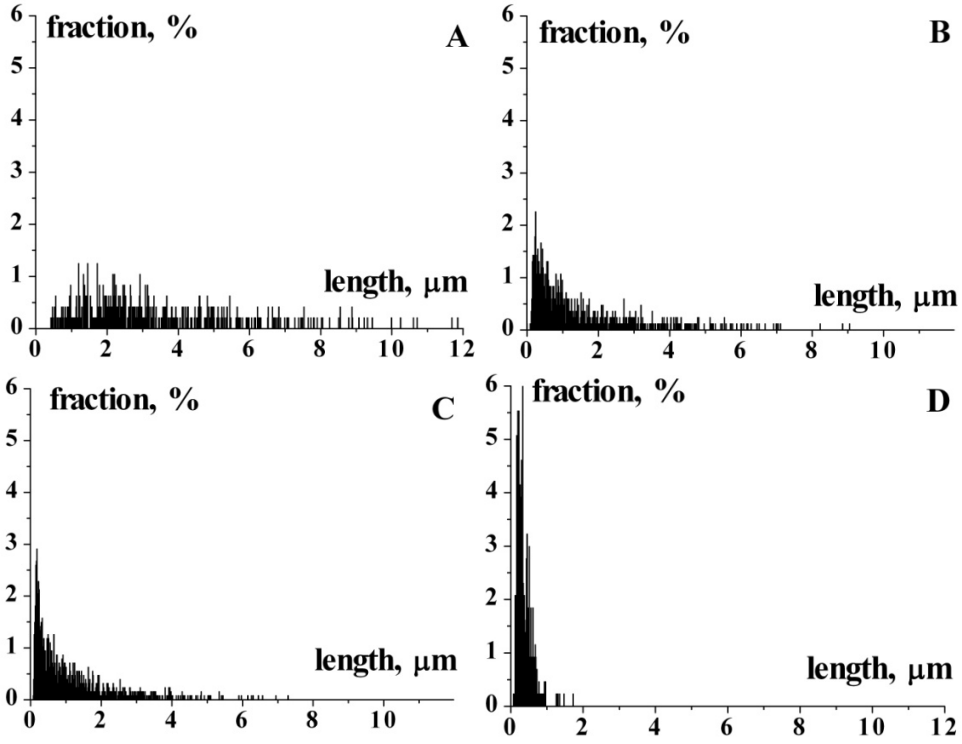

Figure S6: Effect of gelsolin on the filament length distribution, A: R=0, B: R=0.001, C: R=0.01, D: R=0.1; Horizontal axis: length; vertical axis: fraction of filaments with corresponding lengths.