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Running title: Melanosomes transport by myosin-V

Supplementary Materials and Methods.

Numerical simulations

Numerical codes were written and run in Matlab environment (The Mathworks, Natick, Massachussets).

Simulations of particles moving through the actin network were performed considering that the actin matrix is composed by linear filaments which length (L) follows a lognormal distribution with mean 1.3 μm and standard deviation equal to 1. Filaments are considered to be randomly organized and thus each filament may intersect other filaments at any position in every angle. The distribution of the number of intersections per filament (Nt) was assumed to be a Gaussian centered at Nt = 8 with σ =2. The distribution of L and Nt were obtained by fitting the mentioned functions to the experimental distributions of these parameters determined by Snider et al. (19).

The distribution of run lengths of myosin V was considered to follow an exponential-decay function with a characteristic length of 800 nm (21)

Hybrid-trajectories consisted of segments of simulated active transport and of segments obtained from experimental trajectories of melanosomes in myosin-V dominant negative cells. Periods of active motion were simulated using the event-driven method (22) consisting in sorting at each step the coordinates of the following decision point located at a distance λ_i defined as described in the text. The particle is then assumed to move to the decision point along a linear path at a constant velocity randomly selected from the distribution showed in this work (Fig. 4). The coordinates of the particle as a function of time are obtained afterwards using the solution of the evolution equation. The trajectories of particles during periods of diffusion were segments randomly sampled from experimental trajectories of melanosomes in myosin-V dominant negative cells. The initial position of each experimental segment was previously subtracted in order to match the actual position of the particle at the moment of switching from active transport to diffusion. Trajectories were re-sampled with 70 ms of temporal resolution by using a linear interpolation procedure.

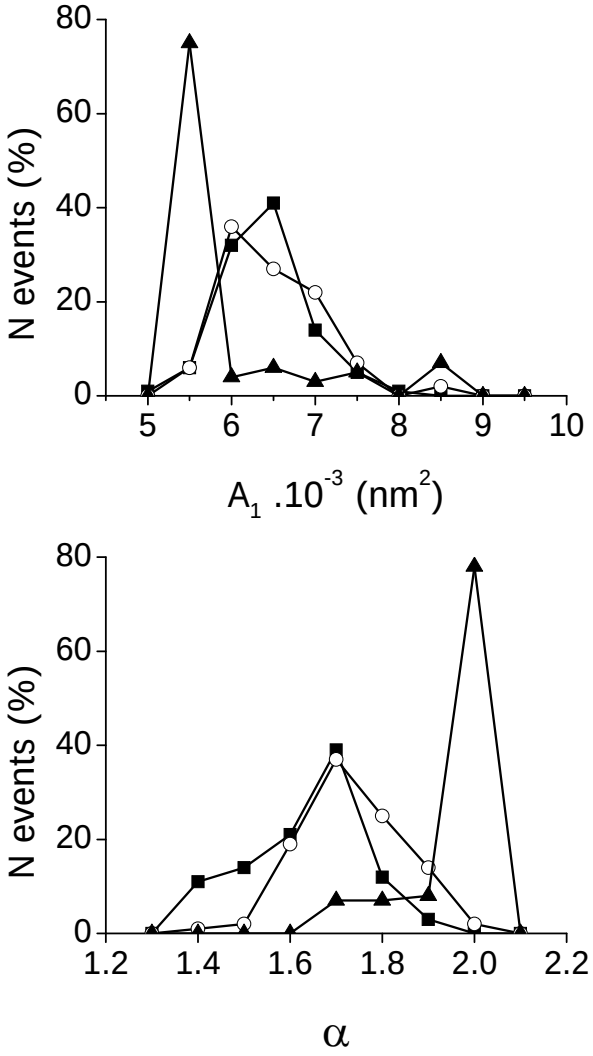

FIGURE S1: Distribution of A_1 (A) and α (B) predicted by modeling continuous melanosome transport along a randomly organized actin network. 100 trajectories of melanosomes moving on an actin network with properties similar to those described before (19) were simulated considering a switching probability of 0 % (▲) 50 % (○) or 80 % (■). This last probability was taken from those data obtained by Ali et al (33) without considering the probability of termination of the motor run. In every case, MSD_o was not significantly different from zero. The bin size of each histogram was set by following the method proposed previously (43)

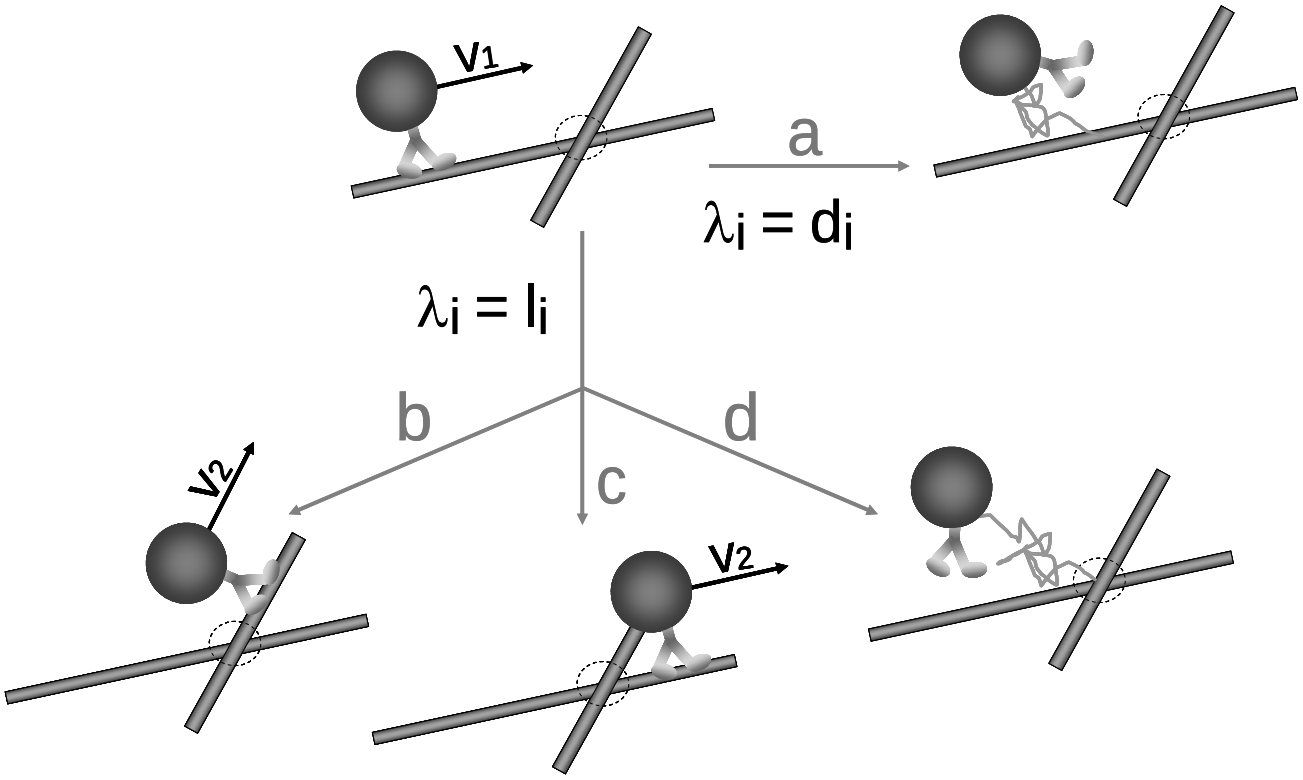

FIGURE S2: Schematic of the transport-diffusion model. A melanosome initially transported by myosin-V along a filament at a constant speed v_1 may spontaneously detach from the track and diffuse for a given time $t_{diffusion}$ (a) or continue moving along the filament until reaching to an intersection with a second filament. In this last case, the melanosome may switch filaments and move with a different velocity v_2 (b); cross the intersection (c) or; detach from the track and diffuse for a given time $t_{diffusion}$ (d).

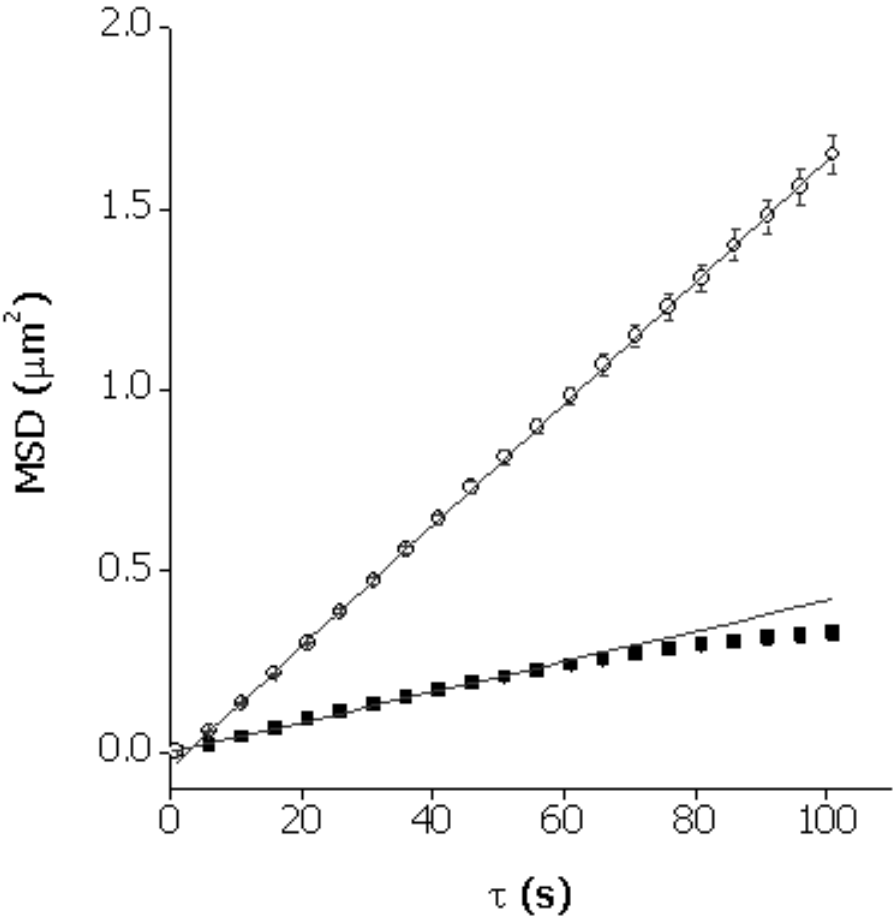

FIGURE S3: Predictions of the transport-diffusion model in a long time-window. The mean MSD value obtained by analyzing 100 trajectories simulated for $t_{diffusion}$ of 3 s (○) and 100 s (■) is represented as a function of the lag time. The error bars correspond to the standard error obtained in each case. Continuous lines are fits of linear equations in the range 1-100 s ($t_{diffusion}$ = 3 s) and 1-50 s ($t_{diffusion}$ = 50 s). For clarity, one every 4 data points are represented in the plot.