Actin-tactoid parameters

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1 Volume of a tactoid

Here we find the volume of a tactoid given a length L, a diameter d and an aspect ratio $\Lambda = L/d$. We assume that a tactoid can be approximated by an ellipsoid, then the volume of a tactoid is

$$V_T \approx \frac{4}{3}\pi.\frac{L}{2}.\frac{d}{2}.\frac{d}{2} = \frac{4\pi}{24}.L.\frac{L^2}{\Lambda^2}$$

$$V_T \approx \frac{\pi L^3}{6\Lambda^2} \tag{1}$$

For $L = 1\mu m$, $\Lambda = 2.4$, we find that $V_T = 0.091\mu m^3$.
For $L = 2\mu m$, $\Lambda = 2.4$, we find that $V_T = 0.727\mu m^3$.

2 Packing fraction of filaments

The volume of a single filament of length L_f, a diameter d_f is given by:

$$V_{filament} = \frac{1}{4}\pi d_f^2 L_f$$

Then, the volume of N_f filaments is given by:

$$V_{filaments} = \frac{1}{4}\pi d_f^2 L_f N_f \tag{2}$$

The packing fraction ϕ is defined by:

$$\phi = \frac{V_{filaments}}{V_T} = \frac{1}{4}\pi d_f^2 L_f N_f \frac{6\Lambda^2}{\pi L^3} = \frac{3\Lambda^2 d_f^2 L_f N_f}{2L^3} \tag{3}$$

3 Number of actin filaments

In experiments, we usually measure a molar concentration c in units of $M(mol/litre)$. This can be represented in terms of the number of molecules by:

$$c = \frac{\text{number of moles}}{\text{volume}} = \frac{N_{molecules}}{N_A V}$$

where $N_A = 6.022$ x $10^{23} mol^{-1}$ is the Avagadro number. In experiments, the molar concentration of actin monomers was approximated to be $c = 250\mu M = 0.25 mol m^{-3}$ [3]. The number of molecules in a tactoid of volume V (1) is given by:

$$N_{molecules} = c.N_A.V = (1.51 \text{ x } 10^{23} m^{-3})\frac{\pi L^3}{6\Lambda^2} \tag{4}$$

We can also find the number of filaments N_f since we know the length of a filament L_f, and the size of a molecule $L_{molecule}$ (in this case monomeric actin $\approx 2.7nm$ [3]).

$$N_f = \frac{N_{molecules}}{\text{Number of molecules in a filament}} = \frac{N_{molecules}}{L_f/L_{molecule}} = \frac{N_{molecules} L_{molecule}}{L_f}$$

$$N_f = (2.13 \text{ x } 10^{14} m^{-2}) \frac{L^3}{\Lambda^2 L_f} \tag{5}$$

For $L_f = 180nm$, $L = 1\mu m$, $\Lambda = 2.4$, we find that $N_f = 205$. This corresponds to a packing fraction $\phi = 0.0156(1.56\%)$.
For $L_f = 180nm$, $L = 2\mu m$, $\Lambda = 2.4$, we find that $N_f = 1,640$. This corresponds to a packing fraction $\phi = 0.0156(1.56\%)$.

4 Number of filamin molecules

Experiments have observed actin tactoids for filamin concentration $c_{filamin} = 2\% - 16\% c_{actin}$ [3]. Choosing the filamin concentration as 10%

$$c_{filamin} = 0.1 c_{monomeric-actin}$$
$$N_{filamin} = 0.1 N_{monomeric-actin}$$

Plugging in the number of actin monomer molecules (4), we find that:

$$N_{filamin} = (7.91 \text{ x } 10^{21} m^{-3}) \frac{L^3}{\Lambda^2} \tag{6}$$

For $L = 1\mu m$, $\Lambda = 2.4$, we find that $N_{filamin} = 1,375$.
For $L = 2\mu m$, $\Lambda = 2.4$, we find that $N_{filamin} = 11,000$.

5 Filamin parameters

Quantity	Symbol	Value	Range	Notes
Free length	l_0	125nm	100-150nm	[3], comm. with Kim Weirich
Spring constant	κ	$0.05pN/nm$	0.02-0.1pN/nm	comm. with Kim Weirich
Diffusion constant (singly-bound)	D_{sb}	$0\mu m^2 s^{-1}$	-	comm. with Kim Weirich
Diffusion constant (doubly-bound)	D_{db}	$0\mu m^2 s^{-1}$	-	comm. with Kim Weirich
Diffusion constant (free)	D_{free}	$1.0\mu m^2 s^{-1}$	-	
Unbinding load sensitivity	λ	0.5	-	chosen to tune energy dependence on binding and unbinding
Parallel to antiparallel ratio	P_{aff}	1.0	-	
Capture radius	r_c	$0.078\mu m$	-	$(D + l_0)/2$
Association constant	K_a	$3.22\mu M^{-1}$	-	[2]
Association constant	K_e/V_B	0.662	-	notes (6)
Turnover rate (doubly to singly)	$k_{o,d}$	$0.305s^{-1}$	-	notes (6)
Turnover rate (singly to unabound)	$k_{o,s}$	$0.305s^{-1}$	-	notes (6)

6 Derivation of rate kinetics

Most experiments measure off rates and association constants for single-stage binding of crosslinks to filaments, i.e. the crosslinks go from unbound to doubly-bound in a single step. To derive rate kinetics for a two-stage binding model, we need to consider both models and match them to get the desired rate constants.

6.1 One-stage model

We start with the one-stage binding model:

$$\frac{d\psi}{dt} = \epsilon^2 c_0 k_{on} - k_{off}\psi \tag{7}$$

where ψ is the doubly-bound crosslinker density, ϵ is the site density per filament, c_0 is the unbound crosslinker concentration, and k_{on} and k_{off} are on and off rates between unbound and doubly-bound states (in units of $\mu M^{-1}s^{-1}$ and s^{-1}). From experiement, we know that $k_{on} = 1.3\mu M^{-1}s^{-1}$ and $k_{off} = 0.71s^{-1}$ [1]. We note that at steady-state:

$$\begin{aligned} \frac{d\psi}{dt} &= 0 \\ \epsilon^2 c_0 k_{on} - k_{off}\psi &= 0 \\ \psi = \epsilon^2 c_0 \frac{k_{on}}{k_{off}} &= \epsilon^2 c_0 K \end{aligned}$$

where K is an association constant.

6.2 Two-stage model

In the two-stage model, crosslinkers first go from unbound to singly-bound:

$$\frac{d\chi_i}{dt} = \epsilon c_0 k_{on,s} - k_{off,s}\chi_i \tag{8}$$

where χ_i is the singly-bound crosslinker density on filament i, and $k_{on,s}$ and $k_{off,s}$ are on and off rates between unbound and singly-bound states. At steady-state, this reduces to:

$$\begin{aligned} \frac{d\chi_i}{dt} &= 0 \\ \epsilon c_0 k_{on,s} - k_{off,s}\chi_i &= 0 \end{aligned}$$

$$\chi_i = \epsilon c_0 \frac{k_{on,s}}{k_{off,s}} = \epsilon c_0 K_a \tag{9}$$

where K_a is an assocation constant. It was measured to be $K_a = 3.22\mu M^{-1}$ [2].

Once singly-bound, crosslinkers can bind to a second filament to reach the doubly-bound state.

$$\frac{d\psi}{dt} = \frac{\epsilon k_{on,d}}{V_B}(\chi_i + \chi_j) - 2k_{off,d}\psi \tag{10}$$

where V_B is a binding volume that a crosslinker can bind to a second filament when it is singly-bound, and $k_{on,d}$ and $k_{off,d}$ are on and off rates between singly and doubly-bound states. The associated equilibrium constant is $K_e = k_{on,d}/k_{off,d}$. Plugging in the steady-state solution from unbound to singly-bound (9), we get:

$$\frac{d\psi}{dt} = \epsilon^2 c_0 \frac{2K_a k_{on,d}}{V_B} - 2k_{off,d}\psi \tag{11}$$

6.3 Matching the models

Matching the coefficients of (7) and (11):

$$k_{on} = \frac{2K_a k_{on,d}}{V_B} \qquad k_{off,d} = \frac{k_{off}}{2}$$

$$k_{on,d} = \frac{k_{on}V_B}{2K_a} \qquad k_{off,d} = 0.305s^{-1}$$

$$\frac{k_{on,d}}{k_{off,d}} = \frac{k_{on}V_B}{2K_a k_{off,d}}$$

$$K_e = 0.662V_B$$

$$\frac{K_e}{V_B} = 0.662$$

In our software, we can set the value of K_e/V_B, so we don't need to derive V_B. The only constant left is the off rate from singly-bound to unbound. Assuming the structure of the first head of filamin doesnt change upon unbinding the second head, a good guess would be $k_{off,s} = k_{off,d} = k_{off}/2 = 0.305s^{-1}$. All the constants for the two-stage model are then given by:

$$K_a = 3.22\mu M^{-1}$$

$$K_e/V_B = 0.662$$

$$k_{off,s} = 0.305s^{-1}$$

$$k_{off,d} = 0.305s^{-1}$$

References

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[2] Fumihiko Nakamura, Teresia M Osborn, Christopher A Hartemink, John H Hartwig, and Thomas P Stossel. Structural basis of filamin a functions. *The Journal of cell biology*, 179(5):1011–1025, 2007.

[3] Kimberly L Weirich, Shiladitya Banerjee, Kinjal Dasbiswas, Thomas A Witten, Suriyanarayanan Vaikuntanathan, and Margaret L Gardel. Liquid behavior of cross-linked actin bundles. *Proceedings of the National Academy of Sciences*, 114(9):2131–2136, 2017.