

Supplemental Material for “Anomalous protein kinetics on low-fouling surfaces”

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SURFACE COVERAGE

As discussed in the main text, the probability that a site is occupied obeys the recurrence relation

$$\frac{dP_{\text{on}}(t)}{dt} = kP_{\text{off}}(t) - \int_0^t kP_{\text{off}}(t')\psi(t-t')dt', \quad (1)$$

where $P_{\text{on}}(t)$ is the probability of the site being occupied and $P_{\text{off}}(t)$ is the probability of the site being empty at time t ,

$$P_{\text{off}}(t) + P_{\text{on}}(t) = 1, \quad (2)$$

and for notation simplicity we have used $k = k_{\text{ads}}A_1$, where k_{ads} is the adsorption rate with units of $\mu\text{m}^{-2}\text{s}^{-1}$ and A_1 is the area of a single site. Because $\frac{dP_{\text{on}}(t)}{dt} = -\frac{dP_{\text{off}}(t)}{dt}$, we can rewrite Eq. (1) as

$$\frac{dP_{\text{off}}(t)}{dt} = -k \left[P_{\text{off}}(t) - \int_0^t P_{\text{off}}(t')\psi(t-t')dt' \right]. \quad (3)$$

The second term on the right has the form of a convolution and can thus be solved using a Laplace transform,

$$sP_{\text{off}}(s) - 1 = -k [P_{\text{off}}(s) - P_{\text{off}}(s)\psi(s)], \quad (4)$$

where we have assumed the site is empty at $t = 0$, i.e. $P_{\text{off}}(t = 0) = 1$. We can obtain the Laplace transform of $\psi(t)$ by using the Tauberian theorem on the survival probability [1]. Namely, given $S(t) \sim S_0/t^\alpha$ we obtain

$$S(s) = S_0\Gamma(1-\alpha)s^{\alpha-1} \quad (5)$$

in the small s limit. Then we can find $\psi(s)$ from the relation

$$S(t) = 1 - \int_t^\infty \psi(t')dt' \quad (6)$$

that yields $\psi(s) = 1 - sS(s)$. Thus

$$\psi(s) = 1 - S_0\Gamma(1-\alpha)s^\alpha. \quad (7)$$

Combining Eq. (7) and Eq. (4), we can solve for $P_{\text{off}}(s)$:

$$P_{\text{off}}(s) = \frac{1}{s + kS_0\Gamma(1-\alpha)s^\alpha} \quad (8)$$

and in the limit $s \rightarrow 0$, it is further simplified into

$$P_{\text{off}}(s) = \frac{1}{kS_0\Gamma(1-\alpha)s^\alpha} \quad (9)$$

when $\alpha < 1$. Again using the Tauberian theorem, we can invert $P_{\text{off}}(s)$:

$$P_{\text{off}}(t) \sim \frac{t^{\alpha-1}}{kS_0\Gamma(1-\alpha)\Gamma(\alpha)}. \quad (10)$$

By using Eq. (2), we can find the time dependence of P_{on} ,

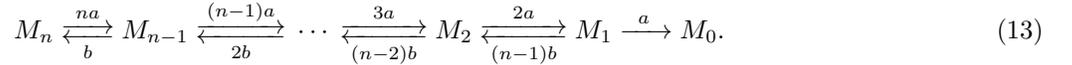
$$P_{\text{on}}(t) \sim 1 - \frac{t^{\alpha-1}}{kS_0\Gamma(1-\alpha)\Gamma(\alpha)}. \quad (11)$$

At last, the surface density is

$$\rho(t) = \frac{P_{\text{on}}(t)}{A_1}. \quad (12)$$

MULTIMERIC DESORPTION MODEL

The desorption reaction for a multimer of n monomers can be written as



To provide an example of how we solve the temporal evolution of this reaction we employ the case $n = 3$. In this case, the reaction simplifies to



Casting this reaction into a set of differential equations yields

$$\begin{aligned} \frac{dp_3}{dt} &= -3a p_3 + b p_2 \\ \frac{dp_2}{dt} &= 3a p_3 - (2a + b) p_2 + 2b p_1 \\ \frac{dp_1}{dt} &= 2a p_2 - (a + 2b) p_1 \\ \frac{dp_0}{dt} &= a p_1. \end{aligned}$$

Considering that when a molecule adsorbs to the surface it should bind via one of its monomers, the initial condition is setting the system in state M_1 . However, the long-time evolution of the system is not sensitive to its initial condition (as long as it is not in state M_0). Thus we set $p_1(0) = 1$ and $p_0(0) = p_2(0) = p_3(0) = 0$. In order to solve the set of differential equations we employ a Laplace transform,

$$\begin{aligned} s p_3(s) &= -3a p_3(s) + b p_2(s) \\ s p_2(s) &= 3a p_3(s) - (2a + b) p_2(s) + 2b p_1(s) \\ s p_1(s) - 1 &= 2a p_2(s) - (a + 2b) p_1(s) \\ s p_0(s) &= a p_1(s). \end{aligned}$$

which is solved to yield

$$p_0(s) = \frac{a [6a^2 + (5a + b)s + s^2]}{s [6a^3 + (11a^2 + 7ab + 2b^2)s + (6a + 3b)s^2 + s^3]}. \quad (15)$$

To solve for the long time asymptote, we consider the small s behavior,

$$p_0(s) = \frac{6a^3}{s [6a^3 + (11a^2 + 7ab + 2b^2)s]}. \quad (16)$$

The inverse Laplace transform of $p_0(s)$ is

$$p_0(t) = 1 - \exp(-k_3 t), \quad (17)$$

where

$$k_3 = \frac{6a^3}{11a^2 + 7ab + 2b^2}. \quad (18)$$

Further, when the adsorption is faster than desorption as should be the case for a particle on the surface, we have $a \ll b$ and we can rewrite Eq. (18) as

$$k_3 = 3\frac{a^3}{b^2}. \quad (19)$$

Following the same methodology, it is shown for any chosen n , in the limit $a \ll b$, that

$$k_n = n\frac{a^n}{b^{n-1}}. \quad (20)$$

DISTRIBUTION OF MULTIMER SIZES

Individual proteins in solution can spontaneously overcome a free energy barrier to dimerize. Subsequently, this soluble dimer can dissociate into single monomers or spontaneously associate to another monomer to form a soluble trimer, and so on. The overall reaction can be described as



Assuming that each additional monomer requires a free energy ΔF , the overall energy of a multimer of size n is $n\Delta F$. In thermodynamic equilibrium the system obeys a Boltzmann distribution,

$$p(n) = c \exp(-n\Delta F/k_B T) \quad (22)$$

where $k_B T$ is thermal energy and c is a normalization constant. The characteristic number of monomers is $n_0 = k_B T \Delta F$ and the normalization constant is $c = \exp(1/n_0) - 1$.

MULTIMERIC DESORPTION WHEN $a/b \ll 1$

When $a/b \ll 1$, we cannot neglect higher order terms in a/b that lead to Eq. (19) and Eq. (20). Without these approximation, we were unable to obtain a full expression for any number of monomers n . Here, we solve for the first six terms in $S(\tau) = \sum_n S_n(\tau)p(n)$, where $S_n(\tau) = \exp(-k_n \tau)$, and $p(n)$ follows a Boltzmann distribution (Eq. (22)).

Solving the multimer desorption reaction (Eq. (13)) for $1 \leq n \leq 5$ yields

$$\begin{aligned} k_1 &= a \\ k_2 &= \frac{2a^2}{b + 3a} \\ k_3 &= \frac{3! a^3}{2b^2 + 7ab + 11a^2} \\ k_4 &= \frac{4! a^4}{3! b^3 + 26ab^2 + 46a^2b + 50a^3} \\ k_5 &= \frac{5! a^5}{4! b^4 + 126ab^3 + 274a^2b^2 + 326a^3b + 274a^4} \end{aligned}$$

Given these five terms, the survival probability $S(\tau)$ using $a/b = 0.57$ as found for the high density PEG is shown in Fig. S7.

Photobleaching decay

The time over which a fluorophore emits fluorescence is an exponential random variable, with PDF $p(t) = t_{pb}^{-1} \exp(-t/t_{pb})$, where t_{pb} is the characteristic photobleaching time. Nevertheless, a single protein is labeled with multiple fluorophores (3 to 6 in our case) and, as a consequence, the bleaching time for a single protein with n fluorophores is the time of the last surviving one. Let us consider n independent, identically distributed (iid) times t_i with cumulative distribution function (CDF) $F_1(t)$, and the maximum of these times $t_{Max} = \max\{t_i\}$. The CDF of t_{Max} is the n^{th} power of the CDF of t [2],

$$F_n(t_{Max}) = [F_1(t_M)]^n. \quad (23)$$

For n exponential iid random variables, we obtain $F_n(t_{Max}) = [1 - \exp(-t_{Max}/t_{pb})]^n$ and thus,

$$f_n(t_{Max}) = \frac{n}{t_{pb}} \exp(-t_{Max}/t_{pb}) [1 - \exp(-t_{Max}/t_{pb})]^{n-1}. \quad (24)$$

In the case of BSA photobleaching the distribution of times is actually more complex because the number of fluorophores n is not fixed and its distribution is unknown. A first order approximation is only useful as an order of magnitude estimate and yields $t_{pb} = 2000$ s for the survival probability with time-lapse imaging (Fig. 1(a) in the main text), and $t_{pb} = 200$ s under continuous illumination (Fig. S2).

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- [1] Joseph Klafter and Igor M Sokolov, *First steps in random walks: from tools to applications* (Oxford University Press, 2011).
 [2] Oliver C. Ibe, *Fundamentals of applied probability and random processes* (Academic Press, 2014).

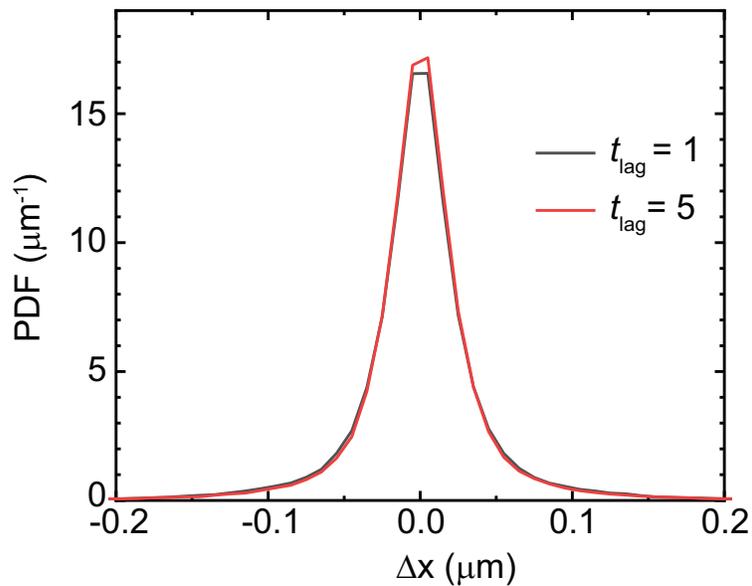


FIG. S1. Distribution of displacements along the projection on the x axis, Δx , for times equal to one frame (2 s) and five frames (10 s) for BSA molecules on a single PEG surface. Ignoring minor statistical errors, the two distributions are identical. The width of both distributions is dictated by the localization error of molecule detection. $n = 238,415$ displacements for a lag time $t_{lag} = 1$ frame and $n = 169,642$ displacements for $t_{lag} = 5$ frames.

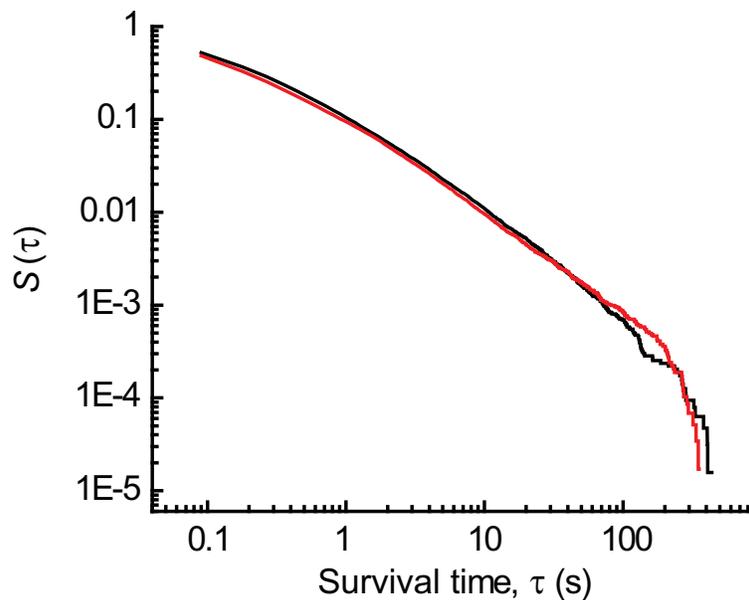


FIG. S2. The survival probability of BSA molecules on PEG surface. The data is similar to that of Fig. 1a in the main text, but the fluorophores are continually excited and the frame rate is 20 fold higher. As a consequence, the decay due to photobleaching takes place in a shorter timescale.

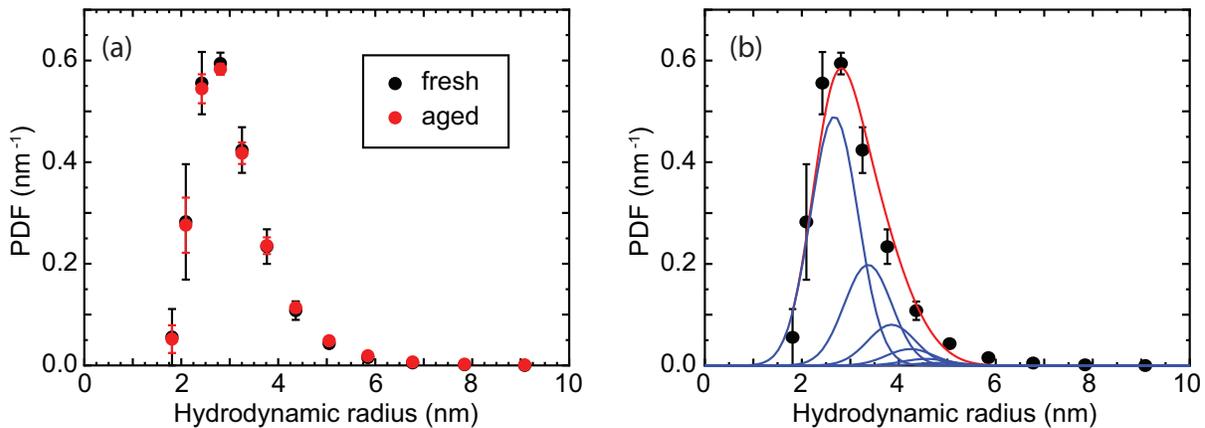


FIG. S3. Measurements of the distribution of Stokes hydrodynamic radii using dynamic light scattering (DLS). (a) Distributions of radii in a freshly prepared BSA solution and in a solution that was allowed to settle for a period of 90 minutes. The two distributions do not appear to be significantly different. (b) The distribution of radii is fit to a model where molecules are allowed to become multimers and they are in equilibrium, so that the multimer size follows a Boltzmann distribution. A least square fit to Eq. 12 in the main text (shown as a red line) yields $A_1 = 0.49$, $s = 0.4$, and $w = 0.49$ nm. The first six Gaussian terms of the sum in Eq. 12 are shown as blue lines.

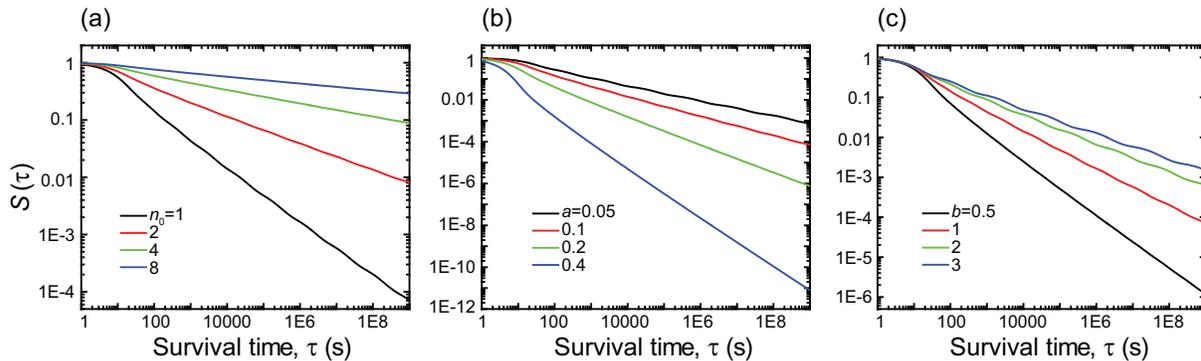


FIG. S4. Survival probability $S(\tau)$ according to the multimer model presented in the main text exhibits a power law tail. The dwell time of particles on the surface obeys an exponential distribution, $S_n(\tau) = \exp(-k_n\tau)$, with mean time, $1/k_n$ that depends on the number of monomers within the adsorbed particle. The *a priori* probability is given by $S(\tau) = \sum_{n=1}^{\infty} S_n(\tau)p(n)$ where $p(n)$ is the probability of the particle having n monomers and it obeys a Boltzmann distribution with characteristic number of monomers n_0 . (a) Survival probability for different n_0 values. Note that larger n_0 means higher probability of the proteins to self-associate. Binding coefficient is $b = 1$ and unbinding rate is $a = 0.1$. (b) Survival probability for different surface dissociation coefficients a . Binding coefficient is $b = 1$ and characteristic number of monomers is $n_0 = 1$. (c) Survival probability for different surface binding affinities b . Dissociation coefficient is $a = 0.1$ and characteristic number of monomers is $n_0 = 1$. Note that when the ratio a/b is very low, the survival probability also exhibits oscillations because the different modes separate in time.

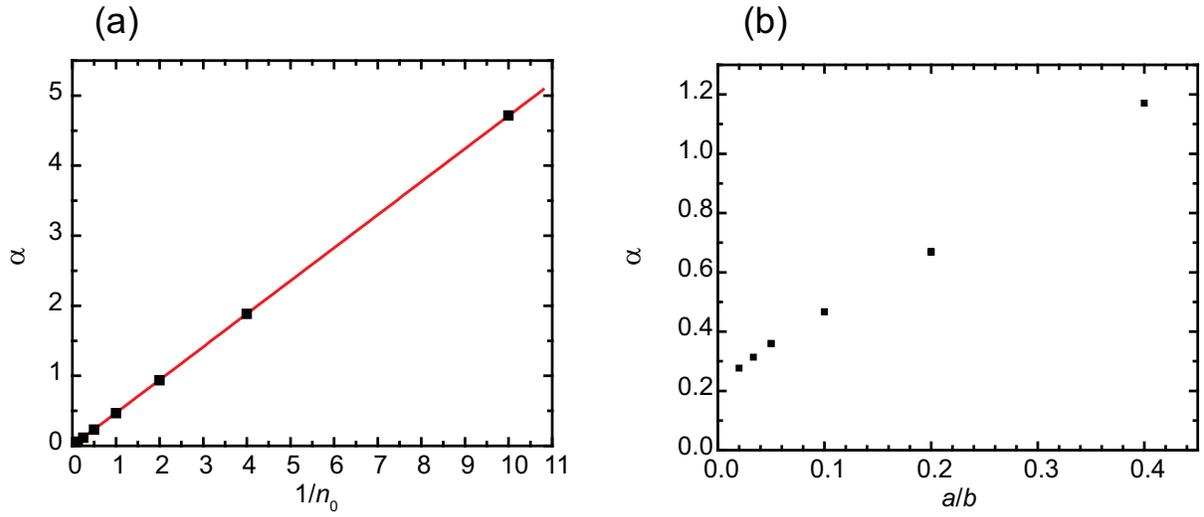


FIG. S5. Anomalous exponent α in the multimer model, where $S(\tau) = \sum_{n=1}^{\infty} S_n(\tau)p(n)$ and $S(\tau) \sim \tau^{-\alpha}$. The exponent α is shown for different choices of parameters n_0 (characteristic number of monomers in adsorbed particle) and a and b (dissociation and association coefficients, respectively). (a) α is shown as a function of $1/n_0$ to highlight the behavior $S \sim 1/n_0$. (b) α is shown as a function of a/b . Different sets of a and b values are used but they all fall within a master curve that only depends on a and b via their ratio a/b .

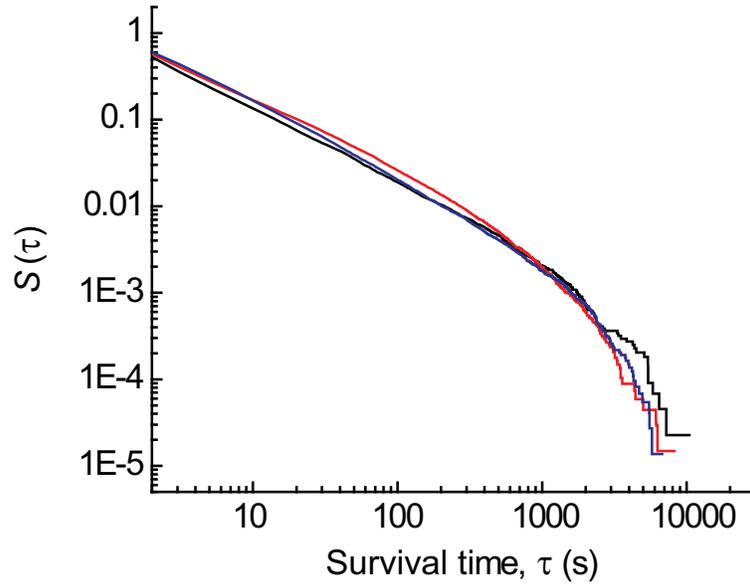


FIG. S6. Survival probability $S(\tau)$ for the surface dwell times of BSA molecules on high density PEG surface.

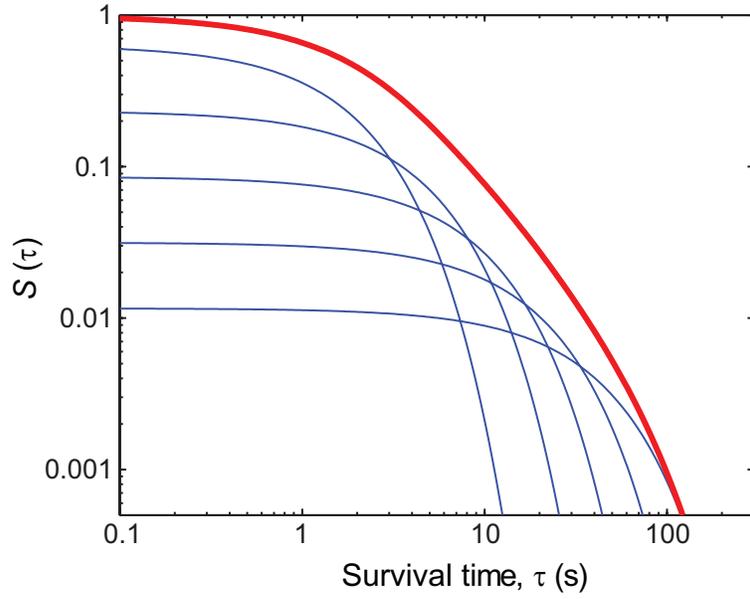


FIG. S7. Survival probability $S(\tau)$ according to the multimer model when the approximation $a/b \ll 1$ does not hold. Here, $a/b = 0.57$. Namely, $n_0 = 1$, $a = 0.57$, and $b = 1$. Only the first five modes of k_n are used. The thin blue lines show the contribution of each of these modes to the survival probability: $p(n) \exp(-k_n \tau)$. The thick red line shows the survival probability: $S(\tau) = \sum_{n=1}^5 S_n(\tau) p(n)$. This survival probability also shows a power law tail.