

Reply:

The authors of the letter have identified an error in our derivation of Eq. AII11, which was based on the persistent random walk model assumed in the Appendix. However, the correct general formula for $\Phi(\tau_1, \tau_2, \tau_3, \tau_4)$ is

$$\begin{aligned} \Phi(\tau_1, \tau_2, \tau_3, \tau_4; P, S) &= S^4 e^{-(\tau_4 - \tau_1)/P} \\ &\times \left(e^{(\tau_3 - \tau_2)/P} \frac{1}{2} (1 + e^{-4(\tau_3 - \tau_2)/P}) \right. \\ &\left. - e^{-(\tau_3 - \tau_2)/P} \right) \end{aligned} \quad (1)$$

This equation should replace the similar but incorrect equation AII11 in the article, which was

$$\begin{aligned} \Phi(\tau_1, \tau_2, \tau_3, \tau_4; P, S) &= S^4 e^{-(\tau_4 - \tau_1)/P} \\ &\times \left(e^{(\tau_3 - \tau_2)/P} - e^{-(\tau_3 - \tau_2)/P} \right) \end{aligned} \quad (2)$$

Integration of AII12 with the corrected integrand Φ yields the following correction to Eqs. AII13 and Eq. 24

$$\begin{aligned} \phi(T_1, T_2, T_3; P, S) &= 4S^4 \left(2 - e^{-T_1/P} - e^{-T_3/P} \right) \left(\left(\frac{T_2}{P} \right) \left(1 + \frac{8}{3} e^{-T_2/P} \right) - \frac{16}{9} \left(1 - e^{-T_2/P} \right) - \left(1 - e^{-2T_2/P} \right) + \frac{1}{36} \left(1 - e^{-4T_2/P} \right) \right) \\ &2S^4 \left(1 - e^{-T_1/P} \right) \left(1 - e^{-T_3/P} \right) \left(2 \left(1 - e^{-2T_2/P} \right) - \frac{8}{3} \left(\frac{T_2}{P} \right) e^{-T_2/P} - \frac{1}{9} \left(1 - e^{-4T_2/P} \right) - \frac{8}{9} \left(1 - e^{-T_2/P} \right) \right) \\ &+ 8S^4 \left(\frac{1}{2} \left(\frac{T_2}{P} \right)^2 + \frac{40}{9} \left(1 - e^{-T_2/P} \right) - \left(\frac{T_2}{P} \right) \left(\frac{11}{4} + \frac{8}{3} e^{-T_2/P} \right) + \frac{1}{2} \left(1 - e^{-2T_2/P} \right) - \frac{1}{144} \left(1 - e^{-4T_2/P} \right) \right) \end{aligned} \quad (3)$$

The corrected form of Eq. AII14, with μ replacing S , follows by substituting $S^2 = 2\mu/P$.

Equation AII13 impacts the calculation of the variance-covariance matrix \mathbf{V} . To quantify the impact of the error, Figure 1 recreates Figure 5a from the original article showing simulated mean-squared displacement curves and the standard errors obtained from the diagonals of the correct and incorrect forms of \mathbf{V} . As shown, both versions reflect the spread in the simulated mean square displacement curves, but the corrected version more accurately estimates the standard deviation from the mean.

Since the method implementing the incorrect version of AII12 has been used by us and others since publication, we have assessed how this error affects the

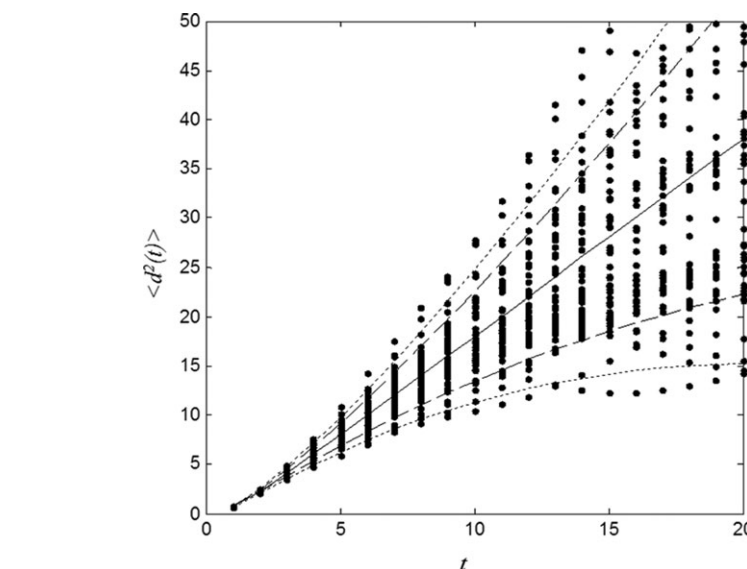


Figure 1. Calculated mean-squared displacements from 50 simulated trajectories (80 positions at time interval 1.0; $S = 1.0$; $P = 1.0$).

The solid line is the exact curve for a persistent random walk with these parameters, and dashed lines represents the calculated standard error in the mean-squared displacements resulting from the correct (long dashes) and incorrect (short dashes) forms of Eq. AII13. (Compare to Figure 5a in the original article).

estimation of the motility parameters. The error creates very little difference in the parameter estimates, but the incorrect form does overestimate the standard error in the parameter estimates calculated from the variance-covariance matrix \mathbf{V} (using Eq. 36 in the original article). To illustrate this effect, Figure 2 shows multiple simulated mean squared displacement curves that were fitted using ordinary least-squares regression (OLSR) or generalized least-squares regression (GLSR) using the original and corrected forms of \mathbf{V} (these plots correspond to Figure 5a of the original article). As shown, the parameter estimates for the correct and incorrect GLSR are consistently very close (and both yield significant improvement over OLSR), but the standard errors in the parameters calculated from Eq. 36 are too large by a factor of ~ 2 when using the incorrect form of Eq. AII13. Importantly, the error results in only more-conservative estimates of the parameter uncertainties than necessary. We surmise that the impact of

the error on conclusions from previous studies using the method should be negligible.

We take this opportunity to note three other typos in the article. In the middle-two integrals of the first term of Eq. AII12, the lower bounds τ_2 and T_1 should be interchanged. In the definition of T_3 in Eq. 17, indices i and j should be interchanged, consistent with Figure 4 where they are correct. In Eq. 19, the direction of the inequality signs should be reversed.

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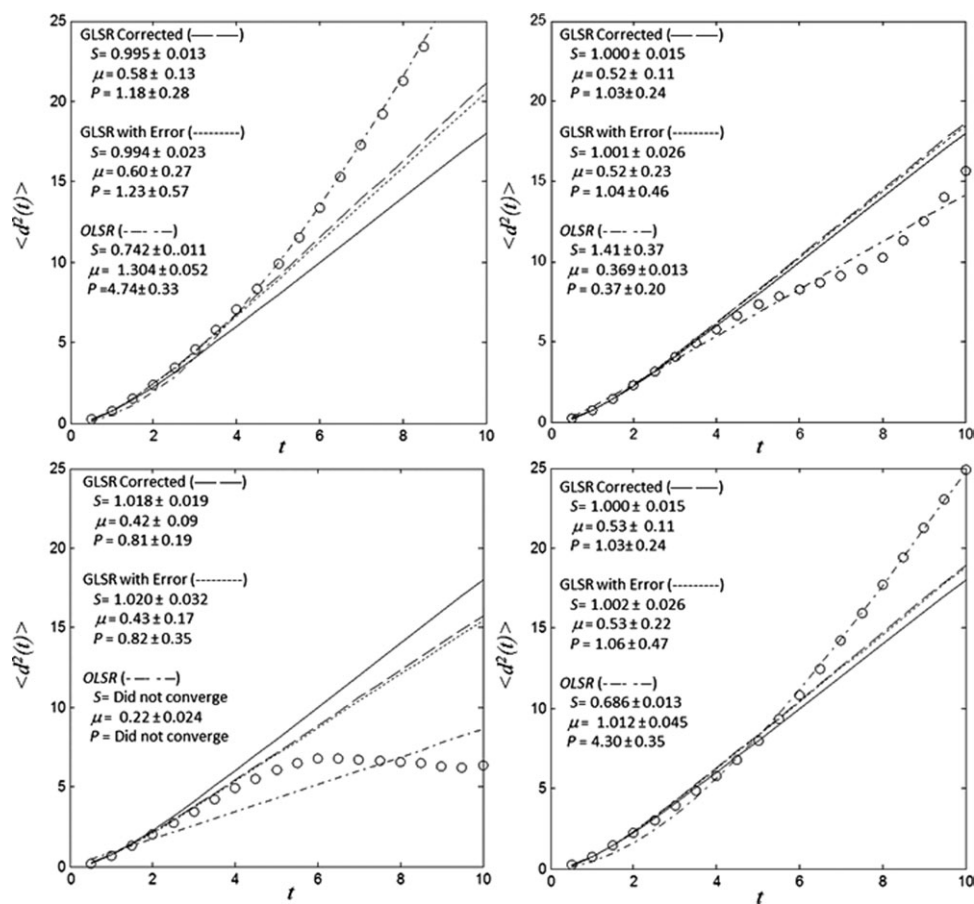


Figure 2. Four examples of fitted curves and resulting parameter estimates using GLSR with the correct (long dashes) and incorrect (short dashes) forms of Eq. A113 using simulated trajectories (40 positions at time interval of 0.5; fits based on first 20 time points). Also shown are the fits using OLSR (dot-dashed line).

The solid line is the exact curve for the simulation parameters ($P = 1.0$; $S = 1.0$; $\mu = 0.5$). As shown, the error causes an overestimation in the calculated uncertainty in the parameter estimates without largely affecting the estimated parameter values. (Compare to Figure 6 in the original article).