10. Estimation of the population size of *Prochlorococcus* that becomes well-mixed within ecologically relevant time scales

Estimating the distance between two 'just-divided' daughter cells over time

Prochlorococcus cells are non-motile, neutrally buoyant, and do not form aggregates. Therefore, the dispersal of single cells is dictated by Brownian motion at early times (order of seconds) and by ambient fluid motion at longer times. Here we focus on the latter, since the former only dominates for very short timescales. Importantly, only relative fluid motion (i.e., differences in fluid velocity) matters for dispersal (*66*), because a uniform flow transports cells without changing their relative distances. A dominant source of relative fluid motion in the ocean is turbulence, which entails velocity differences between different points in space. Since non-motile, neutrally buoyant cells cannot move relative to the fluid, those same velocity differences govern the separation between cells. As a consequence, any two cells tend, on average, to separate over time.

A fundamental length scale in turbulence is the Kolmogorov scale (*67*), η, the scale at which the kinetic energy transferred down from larger scales by inertia balances the dissipation of energy by viscous forces. Typical values of the Kolmogorov scale in the upper ocean are η = 1-5 mm. At scales smaller than η, turbulence reduces to laminar shear, where the velocity difference between two points, u_S, simply increases linearly with their separation distance, d, and with the magnitude of the fluid velocity gradient, γ, as (*68*):

(10.1) $u_S = 0.42\gamma d = 0.42(\varepsilon/\nu)^{1/2} d \qquad (\text{for } d \ll \eta),$

where $\nu = 10^{-6}\ \mathrm{m^2 s^{-1}}$ is the kinematic viscosity of water and $\gamma = (\varepsilon/\nu)^{1/2}$ is the Kolmogorov shear rate. At separation distances d larger than η, the velocity difference between two points, u_L, scales with the one third power of the energy dissipation rate, as (*68*):

(10.2) $u_L = 1.37(\varepsilon d)^{1/3} \qquad (\text{for } d \gg \eta)$.

We can use these expressions for the separation velocities to compute the separation distance of two *Prochlorococcus* cells over time. First we ask how much time it takes for two 'just-divided' cells to be at a distance greater than η (see also ref. (*69*)). This time is obtained by integrating the inverse of the velocity in Equation 10.1 over the separation distance, from the initial separation distance (taken to be the cell diameter of *Prochlorococcus*, D = 0.6 μm) to the Kolmogorov length scale:

(10.3) $T_S = \int_D^\eta \frac{\mathrm{d}x}{u_S(x)} = \int_D^\eta \frac{\mathrm{d}x}{0.42(\varepsilon/\nu)^{0.5} x} = \frac{1}{0.42(\varepsilon/\nu)^{0.5}} \ln(\frac{\eta}{D})$

Typical values of ε in the ocean range from $10^{-8}\ \mathrm{m^2 s^{-3}}$ below the mixed layer to $10^{-6}\ \mathrm{m^2 s^{-3}}$ within the mixed layer. For these values, one obtains $\eta = (\nu^3/\varepsilon)^{1/4}$ = 3.2 mm and 1 mm, respectively, resulting in T_S = 204 s below the mixed layer and T_S = 18 s within the mixed layer. Therefore, two 'just-divided' cells will be separated by a distance larger than the Kolmogorov scale within a few minutes at most, and from that point on, the distance between them is prescribed by Equation 10.2.

We can apply the same approach to compute the separation time once cells are in the second regime (separation distance larger than η), obtaining:

$$(10.4)\quad T_L = \int_{\eta}^{L} \frac{dx}{u_L(x)} = \int_{\eta}^{L} \frac{dx}{1.37(\varepsilon x)^{1/3}} = \frac{1}{2/3}\frac{1}{1.37\varepsilon^{1/3}}\left(L^{2/3} - \eta^{2/3}\right) = \frac{L^{2/3}-\eta^{2/3}}{0.91\varepsilon^{1/3}},$$

which can be solved to obtain the separation distance L after a given time T_L:

$$(10.5)\quad L = \left(0.91\varepsilon^{1/3}T_L + \eta^{2/3}\right)^{3/2} \sim 0.87\varepsilon^{1/2}{T_L}^{3/2} \qquad (\text{for } L >> \eta)$$

For $\varepsilon = 10^{-8}\ \mathrm{m^2s^{-1}}$, the separation distance estimated by this approach will be L ~ 19 m after T_L = 1 hour; L ~ 2.2 km after T_L = 1 day; and L ~ 41 km after T_L = 1 week.

At long times, these values of L might be an overestimate of the actual separation distance and the actual values of L may be an order of magnitude smaller. This can be seen by considering results from tracer dispersal experiments in the ocean. Because *Prochlorococcus* cells can be assumed to behave as passive tracers (they are non-motile and neutrally buoyant), we can empirically estimate their dispersal also by using dispersion coefficients obtained experimentally for patches of tracers injected in the ocean. This is done through estimation of the variance σ_{rc}^2 of the patch radius (a proxy for the area of the patch) after a time t following point-injection of the tracer (*70-73*). Observations resulted in the empirical relation $\sigma_{rc}^2 = 0.0108t^{2.34}$, where σ_{rc} is in cm and t is in seconds (see Fig. 1 in ref. (*70*)). This relation yields $\sigma_{rc} = 6$ km after a time t = 1 week.

Therefore, both estimates indicate that two 'just-divided' *Prochlorococcus* cells will be separated by at least a few kilometers over the course of a week, for typical turbulence conditions in the upper ocean. We conclude that a conservative estimate is that over the course of one week, cells in a horizontal area of 3 km by 3 km in the upper ocean are well mixed.

To summarize, in characteristics upper ocean water' just-divided' *Prochlorococcus* cells will not be within the same milliliter of water within minutes, will be tens of meters apart within one hour, and will be a few kilometers away within a week.