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CURRENT EVIDENCE

Can small zooplankton mix lakes?

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Scientific Significance Statement

Biomixing is the mixing of waters by living organisms in oceans and lakes. Research of the past several decades has provided important insights about the role of biomixing in oceans, showing that vertical migrators, such as crustacean zooplankton, may be able to enhance ocean mixing. However, there is little evidence for the role of biomixing in lakes, including the organisms that might contribute to it, and its potential effects on lake processes. If biomixing occurs in lakes, it has the potential to weaken vertical temperature stratification and enhance fluxes of nutrients and dissolved substances. We argue that there is a need for studies, particularly field studies, on the potential of vertical migrators to generate biomixing in lakes.

Abstract

The idea that living organisms may contribute to turbulence and mixing in lakes and oceans (biomixing) dates to the 1960s, but has attracted increasing attention in recent years. Recent modeling and experimental studies suggest that marine organisms can enhance turbulence as much as winds and tides in oceans, with an impact on mixing. However, other studies show opposite and contradictory results, precluding definitive conclusions regarding the potential importance of biomixing. For lakes, only models and lab studies are available. These generally indicate that small zooplankton or passive bodies generate turbulence but different levels of mixing depending on their abundance. Nevertheless, biogenic mixing is a complex problem, which needs to be explored in the field, to overcome limitations arising from numerical models and lab studies, and without altering the behavior of the animals under study.

Mixing is defined as the combined action of dispersion of dissolved or suspended substances (chemicals or sediment) and enhancement of diffusion of fluid properties, such as heat or salinity (Thorpe 2005). Mixing in lakes plays an important role because it can affect biological and chemical processes (Fischer et al. 1979). External forces acting on lakes can deliver energy into the water column and can drive different local mixing mechanisms depending on the part of the lake under investigation (*see* Fig. 1). The surface layer is the most dynamic and energetic environment; here wind events (A in Fig. 1) usually provide most of the kinetic energy, creating shear, and inducing mixing. During storms, intense mixing can also be generated close to the surface via formation and breaking of surface waves (B) or seiche activity (C). Other processes, such as nocturnal convection (D), when the lake surface cools at night, may alter the potential energy of the water column and affect the lake stratification (Jonas et al. 2003). In the littoral zone, mixing can be enhanced when physical processes (E), such as seiches or wind-generated internal waves, interact with lake physical boundaries and generate boundary mixing with a possible impact on nutrient fluxes (MacIntyre et al. 1999).

The lake interior, below the surface and away from the bottom and shores, responds differently to external forces because of the vertical temperature stratification. The lake interior is

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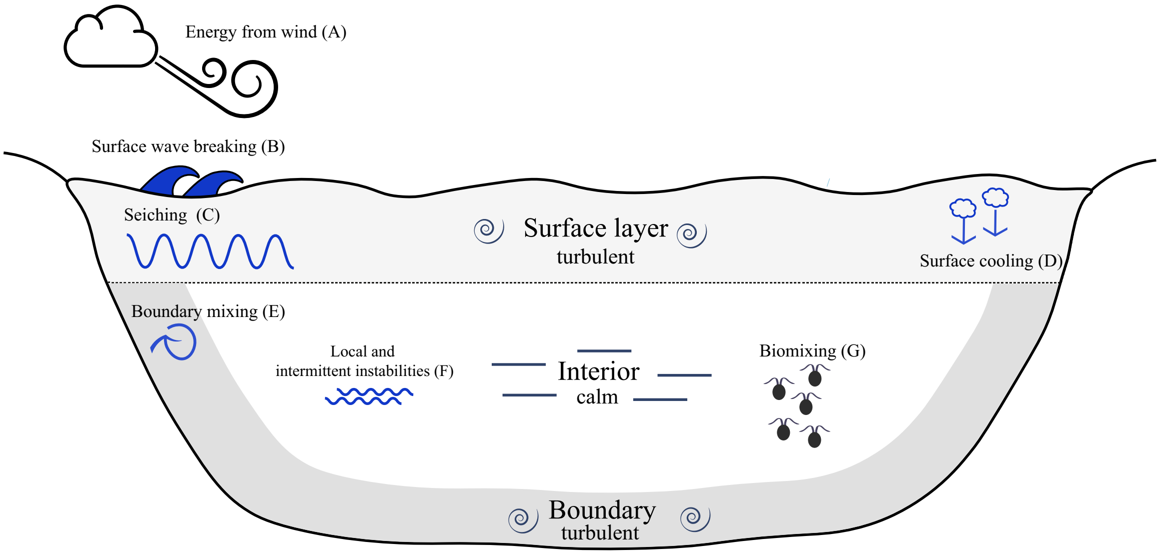

Fig. 1. Sketch illustrating main mixing processes operating in three different lake regions. In the surface layer, energy from wind (**A**) leads to mixing by breaking surface waves (**B**) or via seiching motions (**C**) or convective mixing can act at night when the surface is cooling (**D**). Boundaries are subjected to mixing events (**E**) for example via interactions of internal waves. The lake interior is the calmest region with local and intermittent mixing events (**F**). Vertical migrators (**G**) may provide energy for enhancing the mixing in this layer. Eddies indicate layers with mixing, while straight lines in the interior indicate that energy production is extremely weak and sporadic.

the most quiescent part of a lake where mixing events (F in Fig. 1) are intermittent and localized processes (Wüest and Lorke 2003; Bouffard and Boegman 2012). For this reason, understanding which mechanisms drive interior mixing is of crucial importance for lake ecosystem functioning. Recent research suggests that swimming organisms may operate as a previously neglected mixing mechanism in the interior (Fig. 1, G): by creating hydrodynamic disturbances, such as jets or turbulent eddies, organisms may deliver potential energy to the water column, with a significant contribution toward interior mixing. Recent investigations show that the contribution of horizontal migrators, such as fish, is usually negligible (Gregg and Horne 2009; Pujiana et al. 2015) and attention should instead be focused on vertically migrating zooplankton.

There is currently insufficient understanding of the role of vertically migrating zooplankton as agents of biomixing: these organisms can swim against the stable density stratification, with potential effects on water column mixing and ecological processes. For example, biomixing from vertical migrators may be able to replenish nutrients in surface-depleted waters and stimulate primary production by phytoplankton. If nutrients are brought to the surface, they can also be redistributed via other surface mixing events (such as wind-driven transport or river inflows) to other regions. Oxygen distribution may be altered as well: biomixing enhancement of oxygen fluxes between the surface and metalimnion could reduce deep-water oxygen depletion, with impacts on habitat quality and biogeochemical cycling. Vertical migrators, once they reach the epilimnion, may still enhance turbulence and mixing in unstratified surface waters. Zooplankton-generated turbulent motions can alter ecological interactions by advecting passive bodies such as algae, and increasing encounter rates between zooplankton grazers and their phytoplankton resources (Harris et al. 2000). Given these under-studied possibilities, it is important to study the ecological significance of biomixing in lakes.

Quantifying biomixing is a complex problem because results depend on several factors such as the organisms under investigation, their swimming mode, their concentration and their interactions with the environment. Direct comparisons between current models in the literature and field measurements is not always possible, because probes are not able to sample what happens near the organism's body while swimming.

In the following, we provide a theoretical framework to understand the fundamental physics of biomixing along with some results from in situ ocean observations. We then discuss current studies in lakes and suggest that there is insufficient evidence about the role of biomixing in freshwater bodies. Field observations are needed to overcome some limitations of current studies, and to verify the potential role of biomixing in lakes.

Measuring biomixing

Mixing in lakes can be generally described through a turbulent kinetic energy (TKE) balance, which in the simplest case reads (Osborn 1980; Ivey and Imberger 1991):

$$m = b + \varepsilon \tag{1}$$

where m is the production of TKE, b is the buoyancy flux accounting for the vertical mixing and ε the TKE dissipation rate. External forces, such as wind at the lake surface or eddies generated by swimming organisms in the interior, can provide TKE and contribute to the production term (*m*) in Eq. 1. Part of the source energy is inevitably dissipated as heat (ε) by viscous processes acting at the molecular level. However, some energy may be converted into potential energy (*b*) and affect the position of fluid particles. Changes in the potential energy of the water column can partially destroy the stable vertical stratification and lead to mixing (Fig. 2). Dissipation rates ε can be measured in situ through specific devices, such as shear probes or temperature microstructure profilers, but ε does not provide direct information about mixing. When an increase of ε is observed, it means that energy (*m*) is transferred in the fluid but mixing may not occur, if no input energy is transferred into the component *b*.

Energy dissipation rates (ε) can however be linked to vertical mixing (*b*) via the vertical eddy diffusion coefficient K_V (Osborn 1980):

$$K_V = b / N^2 = \Gamma \cdot \varepsilon / N^2 \tag{2}$$

where $N = [-(g/\rho)\partial\rho/\partial z]^{1/2}$ is the buoyancy frequency describing the vertical stratification which depends on the gravitational acceleration g, water depth z, density ρ and its gradient $\partial\rho/\partial z$. The estimation of the eddy diffusion coefficient K_V is relevant for the quantification of mass vertical fluxes and mixing: when the coefficient is enhanced with respect to background conditions, oxygen or other nutrients can spread in the water column and to different lake layers. The flux F_S of a substance with concentration C_S in the lake can be described using the Fick's law, once K_V is known:

$$F_S = K_V \frac{dC_S}{dz} \tag{3}$$

For waters stratified by temperature, mixing can be enhanced if $K_V > D_T$, where $D_T = 10^{-7}$ m^2 s^{-1} is the molecular temperature diffusivity. However, when $K_V \approx D_T$ dissolved substances will spread very slowly at the molecular level only.

In Eq. 2, Γ is a parameter representing the efficiency of the mixing and provides an estimate of how much energy is converted to mixing (*b*) with respect to the dissipated energy ε. Laboratory and experimental observations suggest $\Gamma \leq 0.2$ (Ivey et al. 2008; Bouffard and Boegman 2013) for wind-generated turbulence. However, for biogenic mixing the value for Γ is still not known. Several conditions and parameters affect the biomixing process, and thus Γ, such as the species of organisms concerned, their size, concentration, swimming behavior, and the environmental conditions such as the stratification strength and the background turbulence dissipation level. If swimming organisms do not efficiently mix the water column, creating small water disturbances, Γ would be too small and K_V does not increase.

Kunze et al. (2006) measured for the first time ε generated by the vertical migration of a population of krill (organism's length $l_{OR} = 1$–2 cm) in Saanich Inlet (Canada). Observed dissipation rates of TKE from biogenic inputs peaked between 10^{-4} W Kg^{-1} and 10^{-5} W Kg^{-1}, compared to typical background level of 10^{-9} W Kg^{-1}. Dissipation spanned five orders of magnitude, suggesting an important krill biomixing contribution as much as mixing from wind and tides. High concentration, and associated multi-body hydrodynamic interactions, probably played an important role, despite weak wind forcing and the strong stratification gradient. The estimated eddy diffusivity from Eq. 2, assuming $\Gamma = 0.2$, ranged between 2×10^{-1} and 2×10^{-2} m^2 s^{-1}, an increase of five orders of magnitude when compared to the daily-averaged level. However, elevated TKE rates were observed by Kunze only for a few minutes during the migration, indicating that the source of turbulence is not constant in time, as was later observed by Rousseau et al. (2010). Rippeth et al. (2007) drew the same conclusions and did not observe such important increases in turbulence from their measurements of TKE dissipation rates in stratified coastal waters of the UK.

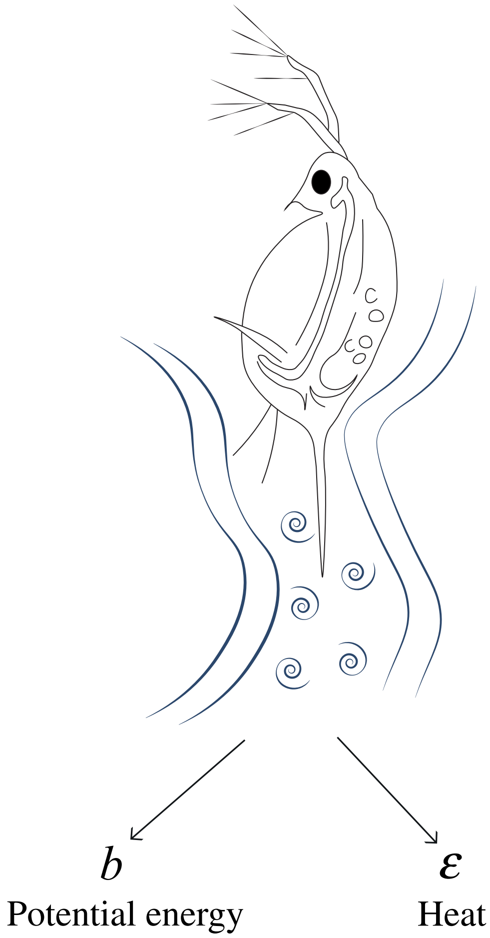

Fig. 2. Schematic of the partition of turbulent kinetic energy imparted by a swimmer (*Daphnia* spp.). The continuous line depicts the wake left by the swimmer, while the eddies are the turbulent instabilities created within the wake that can be a source of TKE. The source energy is converted into potential energy (*b*), increasing the mixing, and into heat as energy is dissipated (ε) due to water molecular viscosity.

Table 1. Main biomixing studies in the literature classified by type of study. For the different kind of analyzed organisms and swimming behaviors, we reported the main results for generated turbulence and mixing. Gray-shaded rows show the few biomixing observations for freshwater zooplankton.

Reference	Type of study	Organism (size)	Swimming behavior	Average ε (W kg^{-1})	Mixing
Huntley and Zhou (2004)	Model	Euphausiids-Whales	Aggregated	10^{-5}	-
Kunze et al. (2006)	Field (ocean)	Krill (1–2 cm)	Aggregated	10^{-5}–10^{-4}	$K_V = 2 \times 10^{-1}$–2×10^{-2} m^2 s^{-1} (with $\Gamma = 0.2$)
Rippeth et al. (2007)	Field (ocean)	Krill	Aggregated	No enhancement	-
Gregg and Horne (2009)	Field (ocean)	Nekton	School	10^{-6}–10^{-5}	No enhancement
Rousseau et al. (2010)	Field (ocean)	Euphausiids	Aggregated	$<10^{-8}$	$K_V \sim 10^{-5}$ m^2 s^{-1} (with $\Gamma = 0.2$)
Thiffeault and Childress (2010)	Model	Krill	Aggregated	$\sim 10^{-6}$	-
Lorke and Probst (2010)	Field (lake)	Perch	Aggregated	3×10^{-9}–10^{-8}	-
Leshansky and Pismen (2010)	Model	Small zooplankton	Aggregated	2×10^{-7}	-
Kunze (2011)	Model	Small zooplankton	Aggregated	10^{-9} (assumption)	$K_V = 2 \times 10^{-7}$ m^2 s^{-1}
Noss and Lorke (2012)	Laboratory	*Daphnia magna* (4 mm)	Tethered on a filament	8×10^{-7} (max: 2×10^{-5})	-
			Freely swimming	2×10^{-6} (max: 3×10^{-4})	$K_V \sim 10^{-5}$ m^2 s^{-1}
Noss and Lorke (2014)	Laboratory	*Daphnia magna* (3 mm)	Aggregated	-	$K_V \sim 10^{-9}$ m^2 s^{-1}
Wagner et al. (2014)	Model	Small zooplankton	Single organism	-	$\Gamma \sim 0.03$
Dean et al. (2015)	Model	Krill	Aggregated	10^{-6}–10^{-7} (highest concentration)	-
Wang and Ardekani (2015)	Model	Small zooplankton	Aggregated	-	$K_V \sim 10^{-6}$ m^2 s^{-1}
Tanaka et al. (2017)	Laboratory	Sardine	Aggregated	2.3×10^{-4}	$K_V \sim 10^{-2}$–10^{-1} m^2 s^{-1} $\Gamma = 0.02$–0.08

Other ocean studies estimated dissipation rates ε and eddy diffusivity K_V through laboratory experiments and models. A summary is presented in Table 1. These studies show that mixing by krill is not feasible (Rousseau et al. 2010) and only possible with high concentrations (Kunze et al. 2006; Dean et al. 2015) but other vertical migrators, such as copepods or other small zooplankton, may still be able to enhance ocean mixing (Huntley and Zhou 2004; Katija 2012). Direct comparisons of dissipation ε, between current models in the literature and field measurements, is not always possible because microstructure profilers, such as the one used by Kunze et al. (2006), are not able to sample turbulence near the organism's body, providing smaller turbulence dissipations than those estimated from models. Finally, the quantification of biomixing, as done by Kunze et al. (2006), must not rely only on the estimation of dissipation rates (ε) and on the assumption that $\Gamma = 0.2$ (Visser 2007*a*,*b*; Subramanian 2010) but must also be based on direct assessment of Γ and K_V in Eq. 2.

Biomixing in lakes

Biomixing observations in lakes are very limited. So far, the only experimental biomixing study in a lake was conducted by Lorke and Probst (2010) for perch (*Perca fluviatilis*), while the first investigations of zooplankton-generated mixing were carried out under controlled laboratory conditions for *Daphnia* only. *Daphnia* is a very common zooplanktonic genus in lakes, with body lengths approximately between 1 mm and 3 mm. Organisms within this genus often undertake diel vertical migration (DVM), ascending at dusk toward the food-rich surface layer to forage on phytoplankton, and sinking back at dawn into deeper, aphotic waters (Ringelberg 1999). DVM is mainly adopted as a predator-avoidance mechanism but other migratory drivers, such as UV exposure or temperature, may play a role (Williamson et al. 2011). Migrations can last anywhere from minutes to a few hours, and their magnitude differs among lakes and between seasons (Ringelberg 2010).

Noss and Lorke (2012) conducted the first laboratory study of dissipation rates (ε) of TKE produced by *Daphnia*. By using a particle image velocimetry (PIV) technique combined with laser-induced fluorescence, they could estimate some energetic parameters of the planktonic organism swimming in different configurations with a density gradient typical of the thermocline ($N = 0.07$ s^{-1}). TKE dissipation rates (ε) and diffusion coefficient (K_V) were estimated considering the water volume influenced by the organism while swimming, which is usually larger than the organism size. Estimated average dissipation was 2×10^{-6} W kg^{-1} with a maximum of 3×10^{-4} W kg^{-1}, in accordance with results from Huntley and Zhou (2004)'s model. Eddy diffusivity was enhanced in the organism vicinity ($K_V \sim 10^{-5}$ m^2 s^{-1}) and was two orders of magnitude bigger than the molecular heat diffusivity ($D_T \sim 10^{-7}$ m^2 s^{-1}), indicating the potential for an impact on temperature gradients in lakes. However, during the experiment, K_V was not measured in the whole tank, therefore it is not certain whether the zooplankton could have affected mixing on scales larger than the organism size. Moreover, the impact of the re-stratification was not evaluated and no conclusion can be drawn about the mixing efficiency Γ.

Later Noss and Lorke (2014) studied the same organism in different swimming configurations and quantified mixing via the diffusion of a fluorescent dye (Rhodamine 6G) injected into a stratified water tank ($N = 0.08$ s^{-1}). *Daphnia* (max. concentration ~ 4 org. L^{-1}) were forced to vertically migrate generating a global diffusivity in the tank as low as 10^{-9} m^2 s^{-1}. Even when swimming in aggregations, *Daphnia* had a small impact on dissolved substances or gases, whose molecular diffusivity D_G is 10^{-9} m^2 s^{-1}. This result differs however from the previous study, because it provides the diffusion coefficient affected at larger scales, while Noss and Lorke (2012) measured the diffusivity in the near vicinity of a single organism only. For *Daphnia*, at organism-scale dissipation ε and mixing can be enhanced, but when K_V is assessed over the effective and larger volume influenced by *Daphnia* migration, the impact on mixing is negligible if compared to wind-induced mixing. To affect temperature stratification in lakes, *Daphnia* aggregation must be able to increase K_V above $D_T = 10^{-7}$ m^2 s^{-1}.

Wilhelmus and Dabiri (2014) later performed another laboratory experiment in an unstratified tank to analyze the fluid instabilities and mixing induced by *Artemia salina*, a small zooplanktonic species ($l_{OR} = 5$mm) that lives in saline lakes. During the vertical migration, induced artificially with a laser, collective swimming dynamics from different organisms created a large downward jet. The length of the generated eddies near its boundary was considerably larger ($l \sim 1$ cm) than a single organism. Their measurements clearly show that swimmers, when present at high concentration, can deliver kinetic energy at scales bigger than the single organism's length with a possible impact on mixing. However, the lack of a stable stratification did not allow the estimation of the real migration effect on mixing after buoyancy restores the initial density gradient: displaced water parcels and properties can return to their initial position with no effect on mixing if swimmers are not sufficiently efficient.

Physics-based models can also be used to evaluate biogenic mixing for lakes. Kunze (2011) estimated the eddy diffusivity coefficient from simple physical considerations and by assuming that each organism can transport a water volume comparable to its size as it swims in a dense aggregation. Kunze (2011) found that the apparent diffusivity depends on the organism concentration C and for *Daphnia* with $C = 100$ org. L^{-1}, the resulting diffusivity is $K_V = 1.7 \times 10^{-7}$ m^2 s^{-1}, suggesting a negligible enhancement in mixing. More importantly, the model does not consider any re-stratification effect and is not suitable for small zooplankton, such as for *Daphnia*, because it assumes that the organism Reynolds number $\mathrm{Re}=U \cdot l_{OR}/\nu < 1$, where U is the organism's speed and ν the kinematic water viscosity.

Laboratory experiments show that $Re \sim 30$–80 for *Daphnia* (Noss and Lorke 2014; Wickramarathna et al. 2014). Furthermore, inertial forces neglected by the model, can further enhance mixing (Noss and Lorke 2014). Another simple and similar approach was previously proposed by Leshansky and Pismen (2010). In their model, swimmers can disperse the turbulent local flow as a function of the school concentration C, the turbulent dissipation ε, the size l of the produced hydrodynamic instabilities, and speed U. By assuming that for a *Daphnia* swarm, $C = 100$ org. L^{-1}, $\varepsilon = 10^{-9}$ W kg^{-1}, $U = 30$ mm/s and $l = l_{OR} = 1$ mm (Gries et al. 1999; Wickramarathna et al. 2014), the diffusion coefficient is 4×10^{-7} m^2 s^{-1}. Diffusivity increases to $K_V = 10^{-5}$ m^2 s^{-1} when $C = 10{,}000$ org. L^{-1}. Estimated coefficients from these models provide a lower bound of mixing and generally suggest that zooplankton may not be able to alter vertical temperature stratification, since $K_V \approx D_T$.

Wagner et al. (2014) provided instead an estimation of mixing in terms of its efficiency Γ (Eq. 2). In their model, each organism is considered very small and swimming in a stable stratified fluid. For a single vertically migrating zooplankton $\Gamma \sim 0.03$, but it may achieve unity depending on the organism's length, swimming mode, and stratification. The model suggests that biomixing seems a feasible mechanism but does not provide any information about the eddy diffusion coefficient K_V. Moreover, the model is more suitable for micro-organisms and does not consider any influence of the zooplankton packaging density C, which may be the main boosting factor for the mixing.

Finally, Wang and Ardekani (2015) numerically resolved the flow field influenced by an aggregation of interacting swimmers in a stratified medium in the intermediate Reynolds number regime. The model is particularly suitable to model small zooplankton and provide a complete description of biomixing. Simulations were performed with a small number of swimmers and aggregations corresponding to very high densities of $C = 10{,}000$ org. L^{-1} to provide an upper-bound for mixing. In particular, organism swimming behavior was modeled as a "squirmer" (Lighthill 1952; Blake 1971) and controlled by a parameter β which scales with the

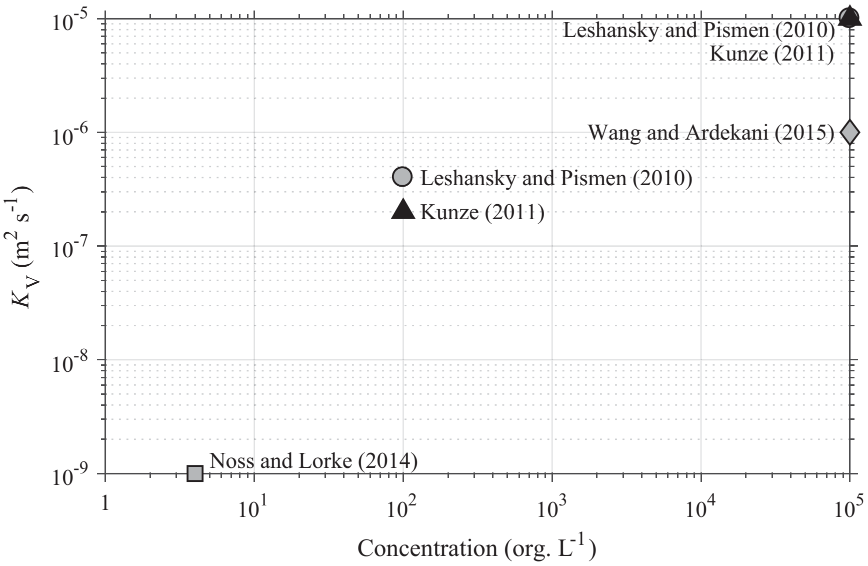

Fig. 3. Eddy diffusivity K_V as a function of zooplankton concentration C from numerical simulations and laboratory experiments.

organism's size l_{OR}, velocity U and fluid generated vorticity; for *Daphnia* $\beta = 1$ (Wickramarathna et al. 2014; Wickramarathna 2016). From this model, the estimated mixing efficiency Γ for *Daphnia* was 0.01, and eddy diffusivity K_V was as low as 2×10^{-7} m^2 s^{-1} but for a very strong density stratification with $N = 1.9$ s^{-1}. However, for a weaker but more realistic stratification, the numerical model by Wang and Ardekani (2015) showed that swimmers were less efficient ($\Gamma = 3 \cdot 10^{-4}$) but generate a higher diffusivity, with $K_V = 10^{-6}$ m^2 s^{-1}. Change of swimming trajectories, vertical orientation as well as organism buoyancy can further enhance these values (Wang and Ardekani 2015).

The need for field studies

Biomixing studies for oceans cannot be used to draw conclusions for lakes because oceans are physico-chemically different to freshwater bodies, and because marine planktonic organisms are more diverse and potentially larger than their freshwater counterparts (Hessen and Kaartvedt 2014), and biomixing is an organism-dependent mechanism. The few studies in the literature for freshwater zooplankton collectively yield differing conclusions about the role of biomixing (Fig. 3). Numerical simulations by Wang and Ardekani (2015) show that biomixing by *Daphnia* is a feasible process when the zooplankton concentration is as high as $C = 10{,}000$ org. L^{-1}. On the other hand, the experimental study by Noss and Lorke (2014) suggests that mixing is negligible with a smaller concentration of organisms from the same genus (4 org. L^{-1}). In the two studies K_V varies by three orders of magnitude, while the concentration C covers four orders of magnitude. Zooplankton abundance depends on both biotic and abiotic environmental conditions; their density in lakes can vary greatly and can be substantially higher than that used in the experiment by Noss and Lorke (2014), especially during the DVM (George and Hewitt 1999; Straile and Adrian 2000; Hembre and Megard 2003; Talling 2003).

Zooplankton aggregation density is important and may have emergent effects on biomixing: higher concentrations can enable interactions of wakes originating from single organisms and enhance shear and mixing in the same fashion as observed by Wilhelmus and Dabiri (2014). The form of the relationship between zooplankton density and biomixing is currently not known e.g., there may be a concentration threshold over which biomixing is enhanced. In addition, numerical simulations currently simplify taxonomic variability in biomixing potential e.g., Kunze (2011) and Wang and Ardekani (2015) describe all the zooplanktonic species with general models, while in reality zooplankton species swim in different ways, and species-specific models may be more suitable to model *Daphnia* and to describe their particular swimming behavior (Jiang and Kiørboe 2011). These interactions, taking place in a real environment, between individuals from multiple species may be stochastic and challenging to describe mathematically. However, community-level effects may be observable in the field.

Field observations are needed to understand the feasibility of biomixing by freshwater zooplankton communities generally, and *Daphnia* specifically, for several reasons. With field studies, it is possible to overcome limitations arising from laboratory experiments under controlled conditions. In the laboratory, diel vertical migration cycles are artificially simulated by alternating light and dark periods with LED panels or using laser beams with a constant intensity. These methods trigger the zooplankton primary phototaxis, which is the movement toward or away from a light beam. *Daphnia* DVM in the field is instead triggered by the secondary phototactic behavior, which is the reaction due to the rate of change in light intensity, usually peaking at dusk and sunrise only (Ringelberg 1999, 2010). These two different behavioral responses also explain why the DVM does not occur during the day or at night and therefore zooplankton responses in lab tanks may be very different from those in the field. Without field observation, it is not known whether the difference in the DVM trigger can affect *Daphnia* swimming responses and, thus, biomixing. Moreover, it is not certain how laser beams, used to fluoresce the fluid in the tank, impact upon zooplankton migration behavior. The use of artificial light, generated by LEDs in Noss and Lorke (2014) to trigger the migration, may explain why only 16% of the organisms into the tank moved and why some of them remained at the tank top or bottom. Field sampling allows the study of organisms in their natural environment without altering the behavior, potentially increasing the realism of biomixing estimates. Field studies also allow understanding the zooplankton concentration during the DVM, compared to the daily zooplankton densities in the lake. Finally, lakes are populated by variable abundances of zooplanktonic species (species of *Daphnia*, *Bosmina*, *Cyclops*, etc.) and other migrators that can interact with *Daphnia*. Other species can affect *Daphnia* density and force them to frequently change their swimming direction in the migrating layer, which could affect the vertical mixing. Such species interactions cannot be easily reproduced in lab experiments, and they may be difficult to address numerically with models. However, field studies would allow us to construct empirical relationships between abundance and biomixing for communities of different compositions, against which to test developing theoretical expectations.

Migration frequently acts as an avoidance mechanism from visual predators such as larval or juvenile fish (Ringelberg 1999; De Robertis 2002; Waya 2004). The presence of chemical substances released by predators, such as kairomones, and sensed by zooplankton, affect DVM leading to increased migration amplitude or faster swimming reactions (Loose and Dawidowicz 1994; Dodson et al. 1997; Ringelberg 1999, 2010). These behavioral responses can increase the size of the generated instabilities and may increase the vertical diffusion K_V. Moreover, food in lab experiments is usually absent and its availability in real lakes, such as a surface or deep chlorophyll maxima, may be another key factor affecting migration amplitude (Dodson et al. 1997; Ringelberg 1999; Rinke et al. 2007). Tank size, light distribution, temperature, and other features of the environment can also change the swimming behavior and limit the swimming reaction (Buchanan et al. 1982; Dodson et al. 1997). Field studies are needed to confirm whether results from experiments under simplified conditions and numerical models are applicable to biomixing mechanisms in complex natural environments. Only field measurements can tell us which lakes are, and are not, prone to such effects so that we can make generalizations about the importance of biomixing.

Challenges for future field investigations

Field investigation should be performed on vertical migrators during the DVM of zooplankton. *Daphnia* are a good candidate to develop our understanding of freshwater biomixing because (1) they are a very common and abundant migrating species in lakes. (2) Despite their smaller size, dissipation rates of kinetic energy are higher for *Daphnia* compared to theoretical estimates for other zooplanktonic species due to their unique swimming mode (Wickramarathna et al. 2014). (3) Finally, they have been studied in the lab, therefore field studies can be used to validate numerical models and compare experimental results under very controlled conditions.

In particular, the DVM can be directly studied both through zooplankton collection and analysis and indirectly via acoustic devices such as ADCPs or echo sounders, allowing a higher spatial and temporal resolution (Lorke et al. 2004; Rinke et al. 2007; Huber et al. 2011). These instruments are usually employed to measure current velocities in three dimensions and to infer turbulence levels as well. The backscatter strength (BS) or amplitude of the scattered wave provided by ADCPs can be used as a proxy for the zooplanktonic concentration and to estimate zooplankton velocities. Higher values of BS indicate higher zooplankton abundance while lower values usually indicate a lack of scatterers in the water. Recent studies by Huber et al. (2011) and Lorke et al. (2004) suggest that ADCPs can be calibrated against the zooplankton concentrations estimated by more traditional means, allowing continuous estimation of their abundance in the water column. However, these devices do not directly provide any information about the zooplankton abundance, size or taxonomy, but they can be used to track their displacement, to understand the timing of the migration and the part of the water column they inhabit during the day.

A first step in assessing biomixing in the field is to measure TKE dissipation rates ε. Generated turbulence during the DVM in lakes can be measured with microstructure profilers which are nowadays normally employed in sampling TKE dissipation rates. In particular, turbulence should be sampled before and after the DVM, to characterize the background turbulence condition without migrators, and during the zooplankton ascent. The duration of observations depends on the time scale of biomixing and measurements should continue for the whole migration duration to understand whether turbulence is patchy and short-lived or energy production by zooplankton is a regular process. Vertical migrators usually swim unsteadily (Noss and Lorke 2012) but asynchronous motions of organisms in the migrating layer may lead to quasi-stationary conditions of turbulence production. If turbulence is enhanced during the migration, this is an indication that energy is generated by zooplankton but, alone, this is not a sufficient proof of biologically-generated mixing. Available energy (m in Eq. 1) can be dissipated as heat with no changes in the potential energy b. However, if no turbulence is observed, zooplankton DVM is not a feasible mechanism for mixing water. Eddy diffusivity K_V can also be inferred from turbulence measurements by using parametrization of Eq. 2, but attention must be paid to the models used because the underlying hypotheses of the mixing parametrizations may not be applicable to biomixing.

If turbulence is generated during the DVM, the next natural step would be to directly measure mixing efficiency Γ or eddy diffusivity K_V via tracer injections (Wüest et al. 1996; Goudsmit et al. 1997; Wain et al. 2013) to measure the effect of the DVM on the eddy diffusivity K_V. This assessment should rely on measurements and comparison of diffusion before and during the DVM. The duration of tracer sampling should continue until after the migration is completed, and longer than the dissipation measurements. This allows understanding of how tracer diffusion is affected over longer time scales, when stratification restores the initial water column density structure affected by the zooplankton migration.

Attempts to study biomixing in the field can however pose important challenges. For example, zooplankton may avoid plankton nets (Brinton 1967; Harris et al. 2000) but disturbance can be limited by using nets with mouth-reducing cones or by reducing the towing speed (UNESCO 1968). The same avoidance mechanisms might be adopted toward free-falling probes (Benoit-Bird et al. 2010; Ross 2014) however, probes are usually designed to avoid any forward disturbances while sampling turbulence. Moreover, turbulence probes may not be able to resolve turbulence produced by a single organism: generated fluid structures from a single individual are generally smaller than the instrument spatial resolution or the turbulence signal may be contaminated by noise.

Zooplankton spatial heterogeneity is another important issue relevant to the role of biomixing in the field. If biologically generated mixing is sampled in the field, results of the measurements may depend on the chosen location within the lake interior because of horizontal zooplankton patchiness (Thackeray at al. 2004; Blukacz et al. 2009). Turbulence profile collection should therefore be coupled with ADCP measurements to continuously measure zooplankton concentration. ADCPs with multiple beams, bottom-mounted in different lake locations, or surveys with a boat-mounted ADCP, allow understanding of vertical and horizontal variations in abundance in the migrating layer and during the DVM. Vertical distribution before DVM and also horizontal patchiness and temporal variation in zooplankton concentration in the migrating layer are relevant to the spatio-temporal dynamics of biomixing. These dynamics can only be observed in the natural environment.

Conclusions

In this paper, we presented an overview of existing studies of turbulence and mixing generated by small zooplankton in lakes. Lake research currently yields mixed conclusions about the feasibility of biomixing, generally showing that small zooplankton can generate turbulence but different levels of mixing depending on the type of study and on the zooplankton abundance. Field studies are needed to overcome limitations arising from lab studies and to confirm the importance of biomixing in complex natural environments such as lakes, and without altering the behavior of the animals generating the biomixing under study.

References

Benoit-Bird, K. J., M. A. Moline, O. M. Schofield, I. C. Robbins, and C. M. Waluk. 2010. Zooplankton avoidance of a profiled open-path fluorometer. J. Plankton Res. **32**: 1413–1419. doi:10.1093/plankt/fbq053

Blake, J. R. 1971. A spherical envelope approach to ciliary propulsion. J. Fluid Mech. **46**: 199. doi:10.1017/S002211207100048X

Blukacz, E. A., B. J. Shuter, and W. G. Sprules. 2009. Towards understanding the relationship between wind conditions and plankton patchiness. Limnol. Oceanogr. **54**: 1530–1540. doi:10.4319/lo.2009.54.5.1530

Bouffard, D., and L. Boegman. 2012. Basin-scale internal waves, p. 102–107. *In* L. Bengtsson, R. W. Herschy, and R. W. Fairbridge [eds.], Encyclopedia of lakes and reservoirs. Springer Netherlands.

Bouffard, D., and L. Boegman. 2013. A diapycnal diffusivity model for stratified environmental flows. Dyn. Atmos. Oceans **61–62**: 14–34. doi:10.1016/j.dynatmoce.2013.02.002

Brinton, E. 1967. Vertical migration and avoidance capability of euphausiids in the California Current. Limnol. Oceanogr. **12**: 451–483. doi:10.4319/lo.1967.12.3.0451

Buchanan, C., B. Goldberg and R. Mccartney. 1982. A laboratory method for studying zooplankton swimming behaviors. Hydrobiologia **89**: 77–89. doi:10.1007/BF00008635

De Robertis, A. 2002. Size-dependent visual predation risk and the timing of vertical migration: An optimization model. Limnol. Oceanogr. **47**: 925–933. doi:10.4319/lo.2002.47.4.0925

Dean, C., A. Soloviev, A. Hirons, T. Frank, and J. Wood. 2015. Biomixing due to diel vertical migrations of zooplankton: Comparison of computational fluid dynamics model with observations. Ocean Model. **98**: 51–64. doi:10.1016/j.ocemod.2015.12.002

Dodson, S. I., S. Ryan, R. Tollrian, and W. Lampert. 1997. Individual swimming behavior of Daphnia: Effects of food, light and container size in four clones. J. Plankton Res. **19**: 1537–1552. doi:10.1093/plankt/19.10.1537

Fischer, H. B., E. J. List, R. C. Y. Koh, J. Imberger, and N. H. Brooks. 1979. Mixing in inland and coastal waters, v. 114. Academic Press.

George, D. G., and D. P. Hewitt. 1999. The influence of year-to-year variations in winter weather on the dynamics of Daphnia and Eudiaptomus in Esthwaite Water, Cumbria. Funct. Ecol. **13**: 45–54. doi:10.1046/j.1365-2435.1999.00007.x

Goudsmit, G., F. Peeters, M. Gloor, and A. Wüest. 1997. Boundary versus internal diapycnal mixing in stratified natural waters. J. Geophys. Res. **102**: 27903–27914. doi:10.1029/97JC01861

Gregg, M. C., and J. K. Horne. 2009. Turbulence, acoustic backscatter, and pelagic nekton in Monterey Bay. J. Phys. Oceanogr. **39**: 1097–1114. doi:10.1175/2008JPO4033.1

Gries, T., K. Jöhnk, D. Fields, and J. R. Strickler. 1999. Size and structure of "footprints" produced by Daphnia: Impact of animal size and density gradients. J. Plankton Res. **21**: 509–523. doi:10.1093/plankt/21.3.509

Harris, R., P. Wiebe, J. Lenz, H. R. Skjoldal, and M. Huntley. 2000. Zooplankton methodology manual. Academic Press.

Hembre, L. K., and R. O. Megard. 2003. Seasonal and diel patchiness of a Daphnia population: An acoustic analysis. Limnol. Oceanogr. **48**: 2221–2233. doi:10.4319/lo.2003.48.6.2221

Hessen, D. O., and S. Kaartvedt. 2014. Top-down cascades in lakes and oceans: Different perspectives but same story? J. Plankton Res. **36**: 914–924. doi:10.1093/plankt/fbu040

Huber, A. M. R., F. Peeters, and A. Lorke. 2011. Active and passive vertical motion of zooplankton in a lake. Limnol. Oceanogr. **56**: 695–706. doi:10.4319/lo.2011.56.2.0695

Huntley, M. E., and M. Zhou. 2004. Influence of animals on turbulence in the sea. Mar. Ecol. Prog. Ser. **273**: 65–79. doi:10.3354/meps273065

Ivey, G. N., and J. Imberger. 1991. On the nature of turbulence in a stratified fluid. Part I: The energetics of mixing. J. Phys. Oceanogr. **21**: 650–658. doi:10.1175/1520-0485(1991)021 < 0650:OTNOTI>2.0.CO;2

Ivey, G. N., K. B. Winters, and J. R. Koseff. 2008. Density stratification, turbulence, but how much mixing? Annu. Rev. Fluid Mech. **40**: 169–184. doi:10.1146/annurev.fluid.39.050905.110314

Jiang, H., and T. Kiørboe. 2011. The fluid dynamics of swimming by jumping in copepods. J. R. Soc. Interface **8**: 1090–1103. doi:10.1098/rsif.2010.0481

Jonas, T., A. Stips, W. Eugster, and A. Wuest. 2003. Observations of a quasi shear-free lacustrine convective boundary layer: Stratification and its implications on turbulence. J. Geophys. Res. **108**: 1–15. doi:10.1029/2002JC001440

Katija, K. 2012. Biogenic inputs to ocean mixing. J. Exp. Biol. **215**(Pt 6): 1040–1049. doi:10.1242/jeb.059279

Kunze, E. 2011. Fluid mixing by swimming organisms in the low-Reynolds-number limit. J. Mar. Res. **69**: 591–601. doi:10.1357/002224011799849435

Kunze, E., J. F. Dower, I. Beveridge, R. Dewey, and K. P. Bartlett. 2006. Observations of biologically generated turbulence in a coastal inlet. Science **313**: 1768–1770. doi:10.1126/science.1129378

Leshansky, A. M., and L. M. Pismen. 2010. Do small swimmers mix the ocean? Phys. Rev. E **82**: 25301. doi:10.1103/PhysRevE.82.025301

Lighthill, M. J. 1952. On the squirming motion of nearly spherical defomable bodies through liquids at very small reynolds numbers. Commun. Pure Appl. Math. **5**: 109–118. doi:10.1002/cpa.3160050201

Loose, C. J., and P. Dawidowicz. 1994. Trade-offs in diel vertical migration by zooplankton: The costs of predator avoidance. Ecology **75**: 2255–2263. doi:10.2307/1940881

Lorke, A., D. F. Mcginnis, P. Spaak, and A. Wüest. 2004. Acoustic observations of zooplankton in lakes using a Doppler current profiler. Freshw. Biol. **49**: 1280–1292. doi:10.1111/j.1365-2427.2004.01267.x

Lorke, A., and W. N. Probst. 2010. In situ measurements of turbulence in fish shoals. Limnol. Oceanogr. **55**: 354–364. doi:10.4319/lo.2010.55.1.0354

MacIntyre, S., K. M. Flynn, R. Jellison, and J. R. Romero. 1999. Boundary mixing and nutrient fluxes in Mono Lake, California. Limnol. Oceanogr. **3**: 512–529. doi:10.4319/lo.1999.44.3.0512

Noss, C., and A. Lorke. 2012. Zooplankton induced currents and fluxes in stratified waters. Water Qual. Res. J. Can. **47**: 276. doi:10.2166/wqrjc.2012.135

Noss, C., and A. Lorke. 2014. Direct observation of biomixing by vertically migrating zooplankton. Limnol. Oceanogr. **59**: 724–732. doi:10.4319/lo.2014.59.3.0724

Osborn, T. R. 1980. Estimates of the local rate of vertical diffusion from dissipation measurements. J. Phys. Oceanogr. **10**: 83–89. doi:10.1175/1520-0485(1980)010<0083:EOTLRO>2.0.CO;2

Pujiana, K., J. N. Moum, W. D. Smyth, and S. J. Warner. 2015. Distinguishing ichthyogenic turbulence from geophysical turbulence. J. Geophys. Res. Oceans **120**: 3792–3804. doi:10.1002/2014JC010659

Ringelberg, J. 1999. The photobehaviour of Daphnia spp. as a model to explain diel vertical migration in zooplankton. Biol. Rev. Camb. Philos. Soc. **74**: 397–423. doi:10.1017/S0006323199005381

Ringelberg, J. 2010. Diel vertical migration of Zooplankton in Lakes and Oceans (Springer). Springer Netherlands.

Rinke, K., I. Hübner, T. Petzoldt, S. Rolinski, M. König-Rinke, J. Post, A. Lorke, and J. Benndorf. 2007. How internal waves influence the vertical distribution of zooplankton. Freshw. Biol. **52**: 137–144. doi:10.1111/j.1365-2427.2006.01687.x

Rippeth, T. P., J. C. Gascoigne, J. A. M. Green, M. E. Inall, M. R. Palmer, J. H. Simpson, and P. J. Wiles. 2007. Turbulent Dissipation of Coastal Seas, a response to "Observations of Biologically Generated Turbulence in a Coastal Inlet." Science Electronic Letters.

Ross, T. 2014. A video-plankton and microstructure profiler for the exploration of in situ connections between zooplankton and turbulence. Deep-Sea Res. Part I **89**: 1–10. doi:10.1016/j.dsr.2014.04.003

Rousseau, S., E. Kunze, R. Dewey, K. Bartlett, and J. Dower. 2010. On turbulence production by swimming marine organisms in the open ocean and coastal waters. J. Phys. Oceanogr. **40**: 2107–2121. doi:10.1175/2010JPO4415.1

Straile, D., and R. Adrian. 2000. The North Atlantic Oscillation and plankton dynamics in two European lakes-two variations on a general theme. Glob. Chang. Biol. **6**: 663–670. doi:10.1046/j.1365-2486.2000.00350.x

Subramanian, G. 2010. Viscosity-enhanced bio-mixing of the oceans. Curr. Sci. **98**: 1103–1108.

Talling, J. F. 2003. Phytoplankton-zooplankton seasonal timing and the "clear-water phase" in some English lakes. Freshw. Biol. **48**: 39–52. doi:10.1046/j.1365-2427.2003.00968.x

Tanaka, M., T. Nagai, T. Okada, and H. Yamazaki. 2017. Measurement of sardine-generated turbulence in a large tank. Mar. Ecol. Prog. Ser. **571**: 207–220. doi.org/10.3354/meps12098 doi:10.3354/meps12098

Thackeray, S. J., D. G. George, R. I., Jones, and I. J. Winfield. 2004. Quantitative analysis of the importance of wind-induced circulation for the spatial structuring of planktonic populations. Freshw. Biol. **49**: 1091–1102. doi: 10.1111/j.1365-2427.2004.01252.x

Thiffeault, J. L., and S. Childress. 2010. Stirring by swimming bodies. Phys. Lett. A **374**: 3487–3490. doi:10.1016/j.physleta.2010.06.043

Thorpe, S. A. 2005. The Turbulent Ocean. Cambridge University Press.

UNESCO. 1968. Zooplankton sampling. UNESCO.

Visser, A. W. 2007*a*. Biomixing of the Oceans? Science (New York, N.Y.) **316**: 838–839. doi:10.1126/science.1141272

Visser, A. W. 2007*b*. Visser's response to Kunze (2006). Science **318**: 1239–1239. doi:10.1126/science.318.5854.1239b

Wagner, G. L., W. R. Young, and E. Lauga. 2014. Mixing by microorganisms in stratified fluids. J. Mar. Res. **72**: 47–72. doi:10.1357/002224014813758940

Wain, D., M. Kohn, J. Scanlon, and C. Rehmann. 2013. Internal wave driven transport of fluid away from the boundary of a lake. Limnol. Oceanogr. **58**: 429–442. doi: 10.4319/lo.2013.58.2.0429

Wang, S., and A. M. Ardekani. 2015. Biogenic mixing induced by intermediate Reynolds number swimming in stratified fluids. Sci. Rep. **5**: 17448. doi:10.1038/srep17448

Waya, R. 2004. Diel vertical migration of zooplankton in the Tanzanian waters of Lake Victoria. Tanzan. J. Sci. **30**: 123–124. doi:10.4314/tjs.v30i1.18394

Wickramarathna, L. N. 2016. Kinematics and energetics of swimming zooplankton. Universität Koblenz-Landau.

Wickramarathna, L. N., C. Noss, and A. Lorke. 2014. Hydrodynamic trails produced by Daphnia: Size and energetics. PloS One **9**. doi:10.1371/journal.pone.0092383

Wilhelmus, M. M., and J. O. Dabiri. 2014. Observations of large-scale fluid transport by laser-guided plankton aggregations. Phys. Fluids **26**: 101302. doi:10.1063/1.4895655

Williamson, C. E., J. M. Fischer, S. M. Bollens, E. P. Overholt, and J. K. Breckenridge, 2011. Towards a more comprehensive theory of zooplankton diel vertical migration: Integrating ultraviolet radiation and water transparency into the biotic paradigm. Limnol. Oceanogr. **56**: 1603–1623. doi:10.4319/lo.2011.56.5.1603

Wüest, A., D. C. Van Senden, J. Imberger, G. Piepke, M. Gloor, A. Wijest, G. Piepke, and M. Gloor. 1996. Comparison of diapycnal diffusivity measured by tracer and microstructure techniques. Dyn. Atmos. Oceans **24**: 27–39. doi: 10.1016/0377-0265(95)00408-4

Wüest, A., and A. Lorke. 2003. Small scale hydrodynamics in lakes. Annu. Rev. Fluid Mech. **35**: 373–412. doi:10.1146/annurev.fluid.35.101101.161220

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