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A pneumatically operated, submersible, 3-dimensional water sampler for microscale studies

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Abstract

To determine how marine microorganisms locally interact, it is critical to assess their 3-dimensional distribution with microscale spatial sampling resolution and minimal disturbance. Here we describe a 3-dimensional syringe pump water sampling device that overcomes the constraints of previous 1- or 2-dimensional samplers. 3DMAPPER consists of a 1-m^3 cubic stainless-steel structure supporting three 10-by-10 arrays of 60-mL syringes with an intersample distance of 5 cm. The design ensures a high stability of syringe sampling position and an undisturbed sampling volume for mean flow velocities ranging from 5 to 180 cm s^{-1}. 3DMAPPER can easily be disassembled into subsections to simplify the handling of multiple samples, the syringes are easy to replace, and the sampling of multiple large volumes allows statistical treatment of multiple variables. The 300 syringes of 3DMAPPER are drained under water by increasing the ambient hydrostatic pressure using a SCUBA tank, and simultaneously and immediately filled upon relieving the inside pressure via a surface valve. 3DMAPPER is independent of a surface power supply and is deployed directly in the water to be sampled, making it versatile and appropriate for in situ deployment.

Introduction

Understanding the 3-dimensional scales of variability of plankton populations in the ocean is a necessary step in interpreting the resources available to herbivores and carnivores and understanding the structure of the environment through which they must navigate. Such information is even more critical at microscales (< 1 m), as individual key processes such as nutrient uptake, viral infection, cell lysis, light harvesting, competition, mating, and predation typically occur across distances of millimeters to centimeters. The consequences of these interactions influence processes such as climate and fisheries productivity up to the global scale (Rivkin and Legendre 2001). Sampling techniques that enable the study of local spatial distributions and predator-prey relationships in 3 dimensions are vital for an improved understanding of species interactions and hence biogeochemical processes.

The exploration of microscale structure of plankton in the oceans has mainly been a technology-limited enterprise, owing to the minute size of the target organisms. Recent major advances came with the development of 1-dimensional (Cowles et al. 1993; Wolk et al. 2002; Mitchell 2004) and 2-dimensional (Franks and Jaffe 2001) laser fluorometers, video plankton recorders (Davis et al. 2004, 2005), hydroacoustic techniques (Traykovski et al. 1998; Warren et al. 2002; Wiebe et al. 2002), and 3-dimensional digital holography (Katz et al. 1999; Hobson et al. 2000; Malkiel et al. 2003; Watson 2004). These techniques are particularly appealing, as they enable large areas of the ocean to be surveyed noninvasively in relatively short periods of time much more efficiently than conventional techniques. They are very expensive, however, require considerable postprocessing effort, are limited to the largest particulate fraction of plankton population (i.e., large phytoplankton and mesozooplankton), and most of them cannot be deployed from small platforms or in certain environments such as lakes, rivers, estuaries, and shallow coastal areas.

A wide range of automated water samplers have been described in the literature. The original samplers were based

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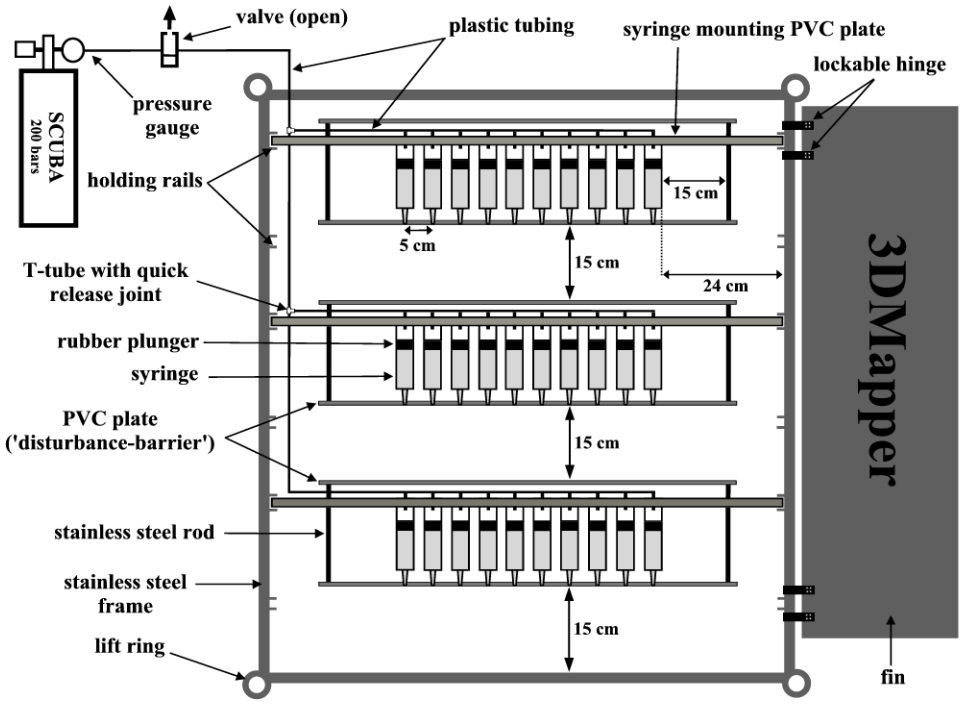

Fig. 1. Schematic view of 3DMAPPER during sampling.

on modified single or multilevel Kemmerer, Van Dorn, or Niskin samplers; manually, mechanically, or pneumatically activated; designed to operate in a horizontal and/or vertical position to investigate microstructure in open water, stratified waters, and at the water-sediment interface (Lund 1954; Walker 1955; Van Dorn 1957; Finucane and May 1961; Gleason and Goff 1963; Joeris 1964; Summerfelt and Lewis 1968; Culberton and Pytkowicz 1970). They have been progressively replaced by smaller and more versatile multilevel glass bottle (Baker 1970) and syringe (Broenkow 1969; Clasby et al. 1972; Heaney 1974; Blackar 1979; Baker et al. 1985; Bjørnsen and Nielsen 1991; Seymour et al. 2000; Waters and Mitchell 2002; Waters et al. 2003) samplers. In general, syringe samplers are small, inexpensive, and versatile, making them highly appropriate for in situ investigations in a range of habitats. Currently available samplers are either spring-loaded or pneumatically operated and incorporate linear series and 2-dimensional arrays of syringes capable of collecting discrete water samples with a spatial resolution and sampling volume ranging from ~ 0.5 to 5 cm and from 50 μL to 60 mL, respectively (e.g., Broenkow 1969; Heaney 1974; Blackar 1979; Cline et al. 1982; Baker et al. 1985; Seymour et al. 2000, 2004; Waters and Mitchell 2002; Waters et al. 2003). In most of these designs, the syringes are attached to a wire line or rod and are activated manually with a messenger, pneumatically, an electric timer (Friederich et al. 1986; Bell et al. 2002), or a microprocessor and a battery contained in a pressure case (Martin et al. 2004).

The previously described samplers have, however, several potential limitations. All require a mechanical link between the actuator and the sampling tube and thus a substantial mounting frame or a continuous electrical supply, which makes them unsuitable for most field deployments, especially underwater. The required electrical power supply of current syringe pumps is not readily accessible in open freshwater and marine environments, and the small sampling volumes of spring-actuated samplers (i.e., 50 to 100 μL; Seymour et al. 2000, 2004) limits the number of parameters that can be examined simultaneously. In addition, the issue of local hydrodynamic disturbance generated by the deployment of the samplers, critical in microscale studies aimed at sampling undisturbed environments, has barely been objectively and quantitatively addressed, and none of the currently available designs are compatible with a 3-dimensional sampling strategy. These constraints considerably limit the generality of using the currently available syringe samplers to assess microscale plankton patterns and processes.

Here we describe 3DMAPPER, an underwater syringe pump that simultaneously samples three 10-by-10 arrays of 60-mL syringes with an intersample distance of 5 cm. Its design ensures high stability of the syringe sampling position, an undisturbed sampling volume under conditions of low and high flow, ease of disassembly into subsections to simplify the handling of multiple samples, easy replacement of syringes, and the sampling of multiple relatively large volumes that allow statistical treatment of multiple variables. Powered by a

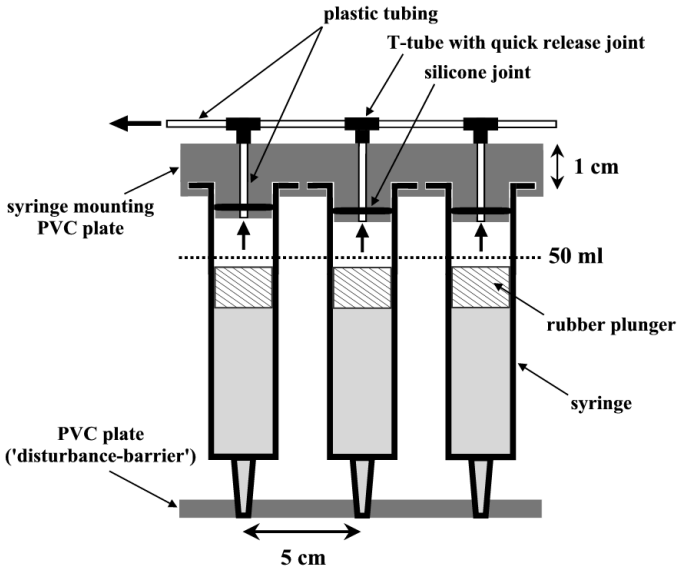

Fig. 2. Schematic close-up view of the syringe assembly.

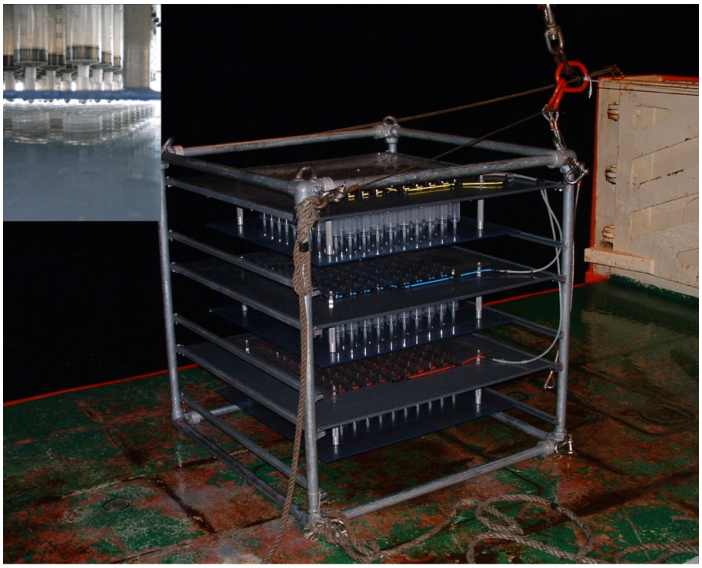

Fig. 3. Picture of 3DMAPPER and close-up of the syringe array upon sample recovery.

SCUBA tank, 3DMAPPER is independent of a surface power supply and is deployed directly in the water to be sampled, making it versatile and appropriate for in situ deployment.

Materials and procedure

Overview of 3DMAPPER—Schematic drawings for construction of the sampler are shown in Figures 1 and 2, and photographs of the instrument are shown in Figure 3. The design of 3DMAPPER consists of a stainless-steel frame (34 mm diameter), a syringe assembly, and a SCUBA tank. The dimensions of the stainless-steel structure of 3DMAPPER have been specifically chosen to fit the definition of microscale, i.e., scales smaller than 1 m that are of ecological relevance to microorganisms under most conditions (e.g., Mackas et al. 1985). The resulting 1-m^3 stainless-steel frame supports three 10-by-10 arrays of 60-mL transparent plastic syringes with an intersample distance of 5 cm (Figure 1). The distance between 2 arrays is 30.5 cm. The SCUBA tank provides the force required to collect and retrieve the samples. 3DMAPPER can be used with 1 to 3 syringe arrays. Eight stainless-steel lift rings welded to the 1-m^3 frame allow the sampler to be weighted and lowered in the water with the syringe arrays positioned vertically or horizontally, and 2 removable fins connected to the frame by lockable hinges ensure the alignment of the device with the main flow. The in situ component of the sampler consists of a stainless-steel frame and syringe assembly, independent of a surface power supply. With a weight of ~ 60 kg, the sampler is versatile enough to be deployed from most boats.

Syringe assembly—The plastic shafts of the syringe plungers have been cut behind the rubber plunger, which can then move up or down the barrel of the syringe under the influence of external or internal pressure. The 1-cm shafts are retained to act as rudders. On each array, the syringes are individually connected to a PVC plate by stainless steel rods. Once inserted on the rod, a 90-degree rotation holds each syringe in place by locking its tabs in a trench of the PVC plate. Torus silicone o-rings prevent waters from entering the airline. Syringes are individually connected to the airline by 3-mm-diameter plastic tubing by T-tube with quick release joints. Any leakage in the airline can then be easily fixed by locally replacing the faulty parts. Syringes with central needle lugs have been used to minimize orientation errors associated with eccentric lugs. The syringe lugs are inserted into a 5-mm-thick PVC plate to ensure the stability of their position and to minimize the hydrodynamic disturbance that may be related to water motion around the barrels and the lugs (Figure 2). Each syringe array is held in place on stainless-steel rails welded to the 1-m^3 frame, and secured by lockable hinges. The plastic tubing manifold of each array is isolated from the surrounding flow by a 5-mm-thick, transparent PVC plate, and connected to common 5-mm-diameter plastic tubing through T-tubes with quick release joints.

Power assembly—The power assembly consists of a SCUBA tank, a pressure gauge, a diving regulator, and a valve. The 5-mm-diameter plastic tubing is connected to a SCUBA tank through a valve and the primary stage of the diving regulator. The regulator is fitted to a pressure gauge, allowing tank pressure to be continuously monitored. The SCUBA tank thus provides the power to drain the syringes under water by increasing the ambient hydrostatic pressure. To prevent premature filling during lowering, the internal pressure is kept above ambient hydrostatic pressure. The pressure differential is controlled by the pressure gauge at the surface. At the required depth of sampling, the syringes are filled simultaneously and instantaneously upon opening of a surface valve that releases the internal pressure. When sampling is conducted near to the

surface, a further pressure decrease is needed to overcome frictional forces; this is achieved using any standard laboratory vacuum pump. On retrieval, 3 PVC racks holding 60-mL vials are inserted below the syringe arrays on stainless-steel rails, the valve is closed, the air line is re-pressurized, and the samples are ejected into the vials.

Assessment

Flow disturbances—The deployment of any sampler generates disturbances related to lowering the sampler in the water column and the surrounding waters flowing around the sampler structure once it has reached the required depth of sampling. In any case, the sampler needs to remain steady during lowering and during sampling, as a wake and a boundary layer are generated, respectively, around the stainless-steel frame and on the PVC plate covering the plastic tubing manifold and the syringe lugs of each array. These issues are critical to ensure the relevance of the results, especially when sampling in rivers, estuaries, and the coastal ocean, where the flows can reach velocities of up to 2 m s^{-1} (e.g., Guichard and Bourget 1998; Seuront 2005).

The alignment of 3DMAPPER with the main flow field was tested with and without the two removable fins. This was investigated over a tidal cycle onboard the NO "Côte de la Manche" from an anchor station located in the coastal waters of the eastern English Channel (50°47′300″N, 1°33′500″E). The orientation of 3DMAPPER and the direction of the current were measured independently. Current speed and direction were measured with an Acoustic Doppler Current Profiler, and the orientation of 3DMAPPER was inferred from the angle between its frame and the axis of the ship bow. 3DMAPPER was lowered into the water with the syringe arrays oriented vertically and horizontally. In both cases, no significant differences were observed between the orientation of the flow and 3DMAPPER (Wilcoxon-Mann-Whitney U test, $P > 0.05$) for tidal flow velocity ranging from 5 to 180 cm s^{-1} when the 2 removable fins were connected to the frame. In contrast, without fins 3DMAPPER was not stable and continually rotated around the axis of the holding cable.

We investigated the potential effect of the wake generated around the stainless-steel frame on the sampling area. Each sampling area is a square of 45 by 45 cm located 24 cm inside the stainless-steel frame, except the lower array of syringes, which is located 15 cm above the frame (see Figure 1). The device has been designed to sample with the syringe arrays parallel to the flow field. The wake generated around the frame is then likely to influence the sampling area for downstream distances ranging from 24 to 69 cm if it becomes wider than 15 cm. A piece of the stainless-steel frame was positioned in the middle of a circular flume, where a steady flow was generated by surface friction of rotating circular PVC plates. The wake generated by the frame was investigated for flow velocities and downstream distances ranging from 5 to 150 cm s^{-1} and from 10 to 100 cm, respectively. The width of the wake was measured through the upstream release of a solution of fluorescein as the width of the nonfluorescent water mass downstream of the frame. The width of the wake estimated at downstream distances ranging from 10 to 100 cm was always significantly smaller than 15 cm ($P < 0.01$, Figure 4a) and converged toward constant values of ~ 5.4 and 9.2 cm at downstream distances bounded between 24 and 70 cm for flow velocities higher than 1 m s^{-1} (Figure 4b).

Finally, the thickness of the boundary layer, δ, created by the water flowing over the surface of the PVC plates covering the plastic tubing manifolds opposite to the arrays of syringes (see Figure 1) was estimated as $\delta = (x\nu/u)^{0.5}$ (Mann and Lazier 1991) where x(m) is the distance from the leading edge, ν the kinematic viscosity ($\nu = 10^{-6}$ m^2 s^{-1}), and u (m s^{-1}) the free-

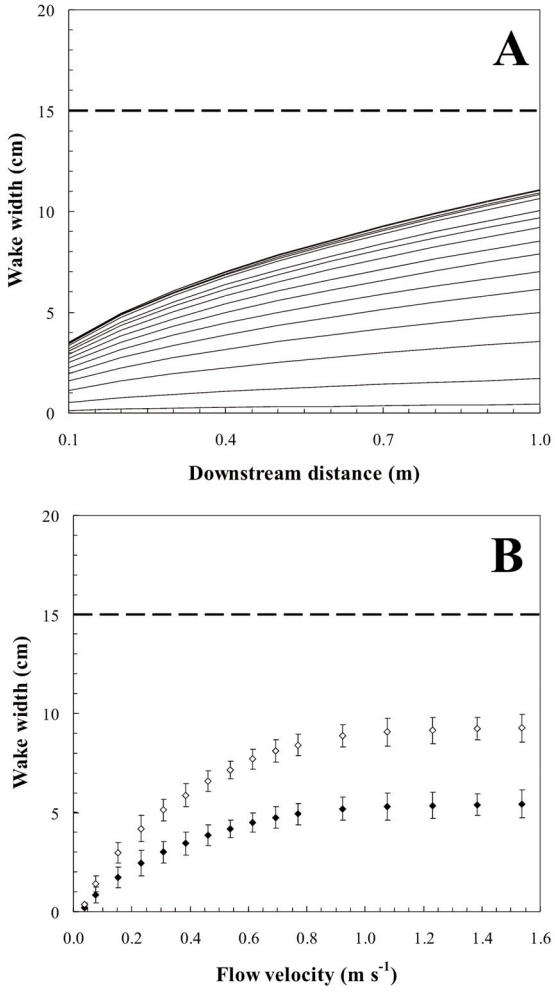

Fig. 4. Mean width of the wake generated around the stainless-steel frame of 3DMAPPER estimated in a circular flume as a function of (A) downstream distance for flow velocity ranging from 0.04 to 1.54 m s^{-1} (from bottom to top; no error bars are shown to increase the readability of the graph and because all standard deviations were lower than 1% of the mean) and (B) flow velocity at downstream distances of 24 cm (open diamonds) and 70 cm (black diamonds) from the frame. The dashed lines represent the minimum distance between the sampling areas and the stainless-steel frame. The error bars are standard deviations of 10 replicates.

Table 1. Results of the 2-layer stratification experiments when the halocline was created with 3DMAPPER in the tank (still experiment) and before lowering 3DMAPPER in the tank (mixed experiment).

	Still experiment			Mixed experiment		
Top layer						
Salinity (psu)	30	15	6	30	15	6
Chl. *a* (µg l-1)	5.2 (0.1)	5.0 (0.1)	5.2 (0.1)	5.1 (0.2)	5.1 (0.3)	5.3 (0.2)
Chl. *a* (CV)	1.9	2.0	2.3	3.5	6.7	4.8
Bottom layer						
Salinity (psu)	33	33	33	33	33	33
Chl. *a* (µg l-1)	25.1 (0.1)	25.1 (0.1)	25.1 (0.1)	24.9 (0.2)	25.0 (0.3)	24.8 (0.3)
Chl. *a* (CV)	0.4	0.6	0.4	0.9	1.4	1.4

The values in parenthesis are the standard deviations, and CV is the coefficient of variation.

stream velocity. For free-stream velocities u and distances x ranging respectively from 0.05 to 2 m s^{-1} and from 15 to 50 cm, the maximum value taken by δ is 0.4 cm. This is negligible compared to the distance separating the arrays of syringes from the PVC plates (i.e., 15.5 cm, see Figure 1) and thus will not affect 3DMAPPER sampling.

Effective undisturbed sampling under stratified conditions—Undisturbed sampling is a highly desirable feature in aquatic sciences. This issue is even more critical for sampling strategies devoted to investigate stratified waters related to thermocline, pycnocline, and halocline, and more specifically to thin layers that are typically microscale structures (Alldredge et al. 2002; McManus et al. 2003). 3DMAPPER has been designed to minimize the flow disturbance during lowering and during sampling, and to ensure that it does not affect the sampling volume. The 1-m^3 structure is, however, likely to bias directly or indirectly the results of any sampling strategy investigating any types of stratified patterns via, for example, destruction of the stratification patterns and the subsequent need for the sampler to be left vertically to ensure complete restratification before sampling. As the latter would be very difficult to achieve in the field, we tested the ability of 3DMAPPER to successfully sample a stratified water column, with its syringe arrays oriented vertically and horizontally.

Three stratification patterns were created in the laboratory to mimic haloclines of different strength. Two-layer stratification was created in a 60-cm-deep tank filled with seawater from the coastal waters of the eastern English Channel (salinity: 33 psu, chlorophyll concentration: 25.1 ± 0.1 µgChl*a* L^{-1}, mean ± 1.96 × SD, n = 30) and treatment waters made from natural seawater progressively diluted with a combination of seawater filtered through 0.45-µm pore size filter and deionized water to obtain surface layers of different salinity but similar chlorophyll concentration. The bottom half of the tank was filled with 30, 15, and 6 psu water containing 5.2 ± 1 µgChl*a* L^{-1} (n = 27), 5.0 ± 0.1 µgChl*a* L^{-1} (n = 26), and 5.2 ± 0.1 µgChl*a* L^{-1} (n = 30), respectively. Natural seawater was then gently siphoned through a tube positioned at the bottom of the tank.

To account for the slightly different designs of the vertically and horizontally oriented 3DMAPPER, 2 experiments were conducted with the syringe arrays oriented horizontally and vertically. In the former and the latter, the natural seawater layers were 35 and 45 cm thick, respectively. For each set of experiments, the potential effect of lowering 3DMAPPER through a stratified water column on the resulting chlorophyll *a* spatial distributions was assessed through the comparison of the spatial distribution obtained (1) creating the halocline with 3DMAPPER in the tank and (2) creating the halocline in a tank where 3DMAPPER was subsequently lowered. 3DMAPPER was always lowered slowly (i.e., 0.2 m s^{-1}) to minimize the disturbance; this speed is fully compatible with most of the available lowering devices routinely used at sea. These 2 experiments will be referred to as "still" and "mixed" experiments hereafter.

Table 1 lists the chlorophyll *a* concentrations estimated from the 2 horizontally oriented 10 by 10 arrays of syringes after lowering 3DMAPPER into the stratified tank for the three 2-layer stratifications considered here. These estimates were not significantly different (Kruskal-Wallis test, $P > 0.05$) from those obtained for the still experiment in the bottom and surface layers. In contrast, the coefficients of variation estimated for chlorophyll *a* distributions above and below the haloclines increased after lowering 3DMAPPER through the halocline (Table 1). These results suggest that the mixing induced by moving 3DMAPPER through the stratified water column could affect the spatial distribution of chlorophyll *a* concentration. This was further investigated using Moran's I and Geary's c spatial autocorrelation statistics (Moran 1950; Geary 1954) to infer the presence of significant spatial structure in the 2D chlorophyll *a* distributions obtained in the still and mixed experiments. Moran's I and Geary's c spatial autocorrelation statistics were tested against the hypothesis of a random distribution. None of the observed distributions were significantly autocorrelated ($P > 0.05$). As the 2 arrays of syringes are separated by 30.5 cm and were 10 to 20 cm away from the interface, these results suggest that the mixing associated with lowering 3DMAPPER with its syringe arrays oriented horizontally does not affect any stratification over distances larger than 10 to 20 cm.

The vertical patterns of chlorophyll *a* concentrations obtained in the still and mixed experiments were slightly dif-

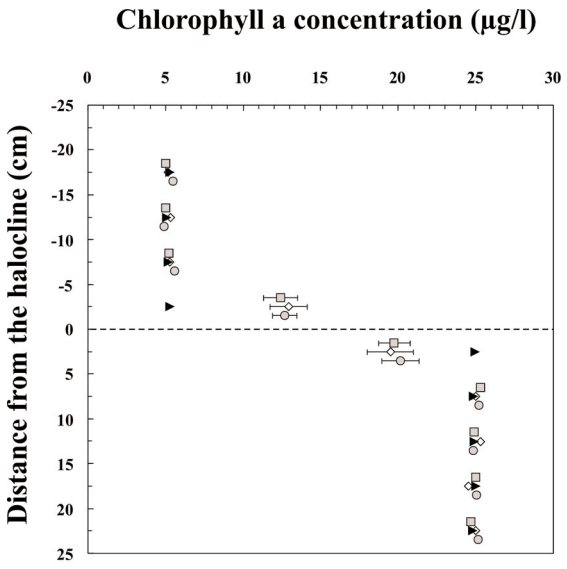

Fig. 5. Mean chlorophyll *a* concentrations obtained from vertically oriented arrays of syringes as a function of the distance from the halocline (30 replicates were done for each distance, and positive and negative values refer to samples taken above and below the halocline, respectively) during the still experiment (black triangles) and the mixed experiment when the salinity of the top layer was 30 psu (gray squares), 15 psu (gray dots), and 6 psu (open diamonds). The error bars are the standard deviations of 30 replicates, and the dashed line indicates the position of the halocline.

ferent (Figure 5). No significant differences were found between the mean chlorophyll concentrations estimated for the 3 shallowest and the 4 deepest depths of the surface and bottom layers (Kruskal-Wallis test, $P > 0.05$). In contrast, the 2 rows of syringes located immediately (i.e., 2.5 cm) above and below the halocline exhibited significantly different chlorophyll concentrations (Wilcoxon-Mann-Whitney *U*-test, $P < 0.01$) and an increase in variability (Figure 5). As the chlorophyll *a* concentrations estimated from the rows of syringes located immediately above and below the halocline were not significantly different between the 2 experiments ($P > 0.05$), the mixing associated with lowering 3DMAPPER through a vertically stratified water column is restricted to the very few centimeters (between 2.5 and 7.5, see Figure 5) located above and below the original stratification.

These results demonstrate that lowering 3DMAPPER with its syringe arrays oriented horizontally or vertically through a stratified water mass does not significantly affect the qualitative and quantitative nature of the resulting 2D distributions. It only weakly affects a stratified water mass through the smoothing of the vertical gradients within a few centimeters above and below the stratification.

Discussion

We have demonstrated that despite its relatively large size (1 m^3), 3DMAPPER will allow sampling with minimal disturbances in stratified and mixed waters, in the water column or on the seafloor, and can be deployed in water masses dominated by strong flows such as tidally driven coastal waters or rivers. The design of 3DMAPPER, as well as its independence from a surface power supply, ensures potential wide-ranging applications within natural water systems, even in remote areas, large water bodies, or in environments dominated by strong flows such as shallow coastal waters, estuaries, and rivers. However, the steadiness and alignment of the device, and thus reliable sampling, ensured in a tidal flow ranging from 5 to 180 cm s^{-1}, can only be expected in conditions of relatively calm weather as those experienced during our field investigations (i.e., wind speed < 5 m s^{-1} and swell < 0.5 m). Rough seas would considerably increase the disturbance related to the deployment of 3DMAPPER in unpredictable ways, and thus critically question the significance of the resulting spatial patterns.

The size and the design of 3DMAPPER have been originally chosen to fit with the original definition of microscale, i.e., scales smaller than 1 m (e.g., Mackas et al. 1985). However, the sampler can be modified to accommodate investigators' needs, the organisms of interest, and site conditions. For example, a study devoted to investigate the spatial distributions and the interactions between flow cytometrically defined microbial populations may require the sampler to be made smaller with a millimeter-scale sampling resolution and the sample volume to be decreased to a few microliters. As the sampler can basically be fabricated from off-the-shelf parts, this can be quite easily achieved without increasing the cost, as previously demonstrated through the building of a 2-dimensional pneumatic sampler with sampling intervals and volumes of 4.5 mm and 50 µL, respectively (Seymour et al. 2000). It is even likely that this would decrease the overall cost of the device as less frame, tubing, and mechanical power would be necessary to achieve a reliable sampling. We nevertheless strongly recommend that any modification of the size and shape of the sampler should be carefully assessed as described in the present work to ensure that it will still allow undisturbed sampling.

3DMAPPER has been designed to improve previous 2-dimensional samplers and provide additional information through multiple (i.e., three) 2-dimensional layers. However, the resolution in the 3rd dimension (30.5 vs. 5 cm) and the lower number of samples (3 vs. 10) do not make 3DMAPPER a truly 3-dimensional sampler. Instead it should be referred to as a "quasi-3D" sampler, as any information in the 3rd dimension will be statistically different from the other 2 dimensions. This should be considered with caution upon examination and interpretation of the resulting spatial patterns. The 3-dimensional microscale nature of 3DMAPPER could nevertheless be conveniently used to gain further insight into the seldom-investigated internal structure of persistent thin layers (e.g., MacManus et al. 2003). Whereas the existence of thin layers has the potential to drive the trophic dynamics of the whole water column, the distribution of plankton organisms within these layers has potentially significant ecological con-

sequences, including increased probability of predator-prey encounter (Seuront et al. 2001), enhanced water-column productivity (Brentnall et al. 2003), and more understanding of the puzzling "paradox of the plankton" (Hutchinson 1961), in which high species diversity occurs in small, seemingly homogeneous bodies of water.

References

Alldredge, A. L., and others. 2002. Occurrence and mechanisms of formation of a dramatic thin layer of marine snow in a shallow Pacific fjord. Mar. Ecol. Prog. Ser. 233:1:12.

Baker, A. L. 1970. An inexpensive microsampler. Limnol. Oceanogr. 15:158-160.

———, K. Krommer Baker, and P. A. Tyler. 1985. A family of pneumatically-operated thin layer samplers for replicate sampling of heterogeneous water columns. Hydrobiol. 122: 207-211.

Bell, J., J. Betts, and E. Boyle. 2002. MITESS: a moored in situ trace element serial sampler for deep-sea mooring. Deep-Sea Res. I 49:2103-2118.

Bjørnsen, P.K., and T. G. Nielsen. 1991. Decimeter scale heterogeneity in the plankton during a pycnocline bloom of *Gyrodinium aureolum*. Mar. Ecol. Prog. Ser. 73:263-267.

Blackar, I. A. 1979. A close-interval water sampler with minimal disturbance properties. Limnol. Oceanogr. 24:983-988.

Brentnall, S. J., K. J. Richards, J. Brindley, and E. Murphy. 2003. Plankton patchiness and its effect on larger-scale productivity. J. Plankton Res. 25:121-140.

Broenkow, W. W. 1969. An interface sampler using spring-actuated syringes. Limnol. Oceanogr. 14:288-291.

Clasby, R. C., W. S. Reeburgh, and V. Alexander. 1972. A close-interval syringe sampler. Limnol. Oceanogr. 17:632-633.

Cline, J. D., H. B. Milburn, and D. P. Wisegarver. 1982. A simple rosette-mounted syringe sampler for the collection of dissolved gases. Deep-Sea Res. 29:1245-1250.

Cowles, T. J., R. A. Desiderio, and W. S. Neuer. 1993. In situ characterisation of phytoplankton from vertical profiles of fluorescence spectra. Mar. Biol. 115:217-222.

Culberton, C., and R. M. Pytkowicz. 1970. A near-bottom water sampler. Limnol. Oceanogr. 15:160-162.

Davis, C. S., Q. Hu, S. M. Gallager, X. Tang, and C. J. Ashjian. 2004. Real-time observation of taxa-specific plankton distributions: an optical sampling method. Mar. Ecol. Prog. Ser. 284:77-96.

———, F. T. Thwaites, S. M. Gallager, and Q. Hu. 2005. A three-axis fast-tow digital Video Plankter Recorder for rapid surveys of plankton taxa and hydrography. Limnol. Oceanogr. Methods 3:59-74.

Finucane, J. H., and B. Z. May. 1961. Modified van Dorn water sampler. Limnol. Oceanogr. 6:85-87.

Franks, P. J. S., and J. S. Jaffe. 2001. Microscale distributions of phytoplankton: initial results from a two-dimensional imaging fluorometer, OSST. Mar. Ecol. Prog. Ser. 220:59-72.

Friederich, G. E., P. J. Kelly, and L. A. Codispoti. 1986. An inexpensive moored water sampler for investigating chemical variability. *In*: J. Bowman, M. Yentsch, and W. T. Peterson, Eds. Tidal Mixing and Plankton Dynamics. Springer-Verlag. p. 463-482.

Geary, R. C. 1954. The contiguity ratio and statistical mapping. Incop. Statist. 5:115-145.

Gleason, G. R., and G. F. Goff. 1963. A multi-level water sampler. Prog. Fish. Cult. 25:104-105.

Guichard, F., and E. Bourget. 1998. Topographic heterogeneity, hydrodynamics, and benthic community structure: a scale-dependent cascade. Mar. Ecol. Prog. Ser. 171:59-70.

Heaney, S. I. 1974. A pneumatically-operated water sampler for close intervals of depth. Freshwater Biol. 4:103-106.

Hobson, P. R., R. S. Lampit, A. Rogerson, J. Watson, X. Fang, and E. P. Krantz. 2000. Three-dimensional spatial coordinates of individual plankton determined using underwater hologrammetry. Limnol. Oceanogr. 45:1167-1174.

Hutchinson, G. E. 1961. The paradox of the plankton. Am. Nat. 95:137-145.

Katz, J., P. L. Donaghay, J. Zhang, S. King, and K. Russell. 1999. Submersible holocamera for detection of particle characteristics and motions in the ocean. Deep-Sea Res. I 46:1455-1481.

Joeris, L. S. 1964. A horizontal sampler for collection of water samples near the bottom. Limnol. Oceanogr. 9:595-598.

Lund, J. W. G. 1954. The seasonal cycle of the plankton diatom *Melosira italica* (Her.) Kütz subsp. *subarctica* O. Müll. J. Ecol. 42:151-179.

Mackas, D. L., K. L. Denman, and M. R. Abbott. 1985. Plankton patchiness: biology in the physical vernacular. Bull. Mar. Sci. 37:652-674.

Malkiel, E., J. Sheng, J. Katz, and J.R. Strickler. 2003. The three-dimensional flow field generated by a feeding calanoid copepod measured using digital holography. J. Exp. Biol. 206: 3657-3666.

Mann, K. H., and J. R. N. Lazier. 1991. Dynamics of Marine Ecosystems: Biological-Physical Interactions in the Oceans. Boston: Blackwell.

Martin, J. B., R. G. Thomas, and K. M. Hartl. 2004. An inexpensive, automatic, submersible water sampler. Limnol. Oceanogr. Methods 2:398-405.

McManus, M. A., and others. 2003. Characteristics, distribution and persistence of thin layers over a 48 hour period. Mar. Ecol. Prog. Ser. 261:1-19.

Mitchell, J. G. 2004. Rank-size analysis and vertical phytoplankton distribution patterns. In: L. Seuront, and P. G. Strutton, eds. Handbook of Scaling Methods in Aquatic Ecology: Measurement, Analysis, Simulation. CRC Press. p. 257-278.

Moran, P. A. P. 1950. Notes on continuous stochastic phenomena. Biometrika 37:17-23.

Rivkin, R. B., and L. Legendre. 2001. Biogenic carbon cycling in the upper ocean: effects of microbial respiration. Science 291:2398-2400.

Seuront, L. 2005. Hydrodynamic and tidal controls of small-scale phytoplankton patchiness. Mar. Ecol. Prog. Ser. 302:93-101.

———, F. Schmitt, and Y. Lagadeuc. 2001. Turbulence intermittency, small-scale phytoplankton patchiness and encounter rates in plankton: where do we go from here? Deep-Sea Res. I 48:1199-1215.

Seymour, J. R., J. G. Mitchell, L. Pearson, and R. L. Waters. 2000. Heterogeneity in bacterioplankton abundance from 4.5 millimeter resolution sampling. Aquat. Microb. Ecol. 22:143-153.

———, J. G. Mitchell, and L. Seuront. 2004. Microscale heterogeneity in the activity of coastal bacterioplankton communities. Aquat. Microb. Ecol. 35:1-16.

Summerfelt, R. C., and W. M. Lewis. 1968. A water sampler employing a solenoid tripping mechanism. Trans. Ma. Fisheries Soc. 97:287-289.

Traykovski, L.V., T. K. Stanton, P. H. Wiebe, and J. F. Lynch. 1998. Model-based covariance mean variance classification techniques: algorithm development and application to the acoustic classification of zooplankton. IEEE J. Ocean. Engin. 23:344-364.

Van Dorn, W. G. 1957. Large-volume water sampler. Trans. Am. Geophys. Union 37:682-684.

Walker, C. R. 1955. A modification of the Kemmerer water bottle for sampling shallow waters. Prog. Fish Cult. 17:41.

Warren, J. D., T. K. Stanton, D. E. McGehee, and D. Chu. 2002. Effect of animal orientation on acoustic estimates of zooplankton properties. IEEE J. Ocean. Engin. 27:130-138.

Waters, R. L., and J. G. Mitchell. 2002. Centimeter-scale spatial structure of estuarine *in vivo* fluorescence profiles. Mar. Ecol. Prog. Ser. 237:51-63.

———, J. G. Mitchell, and J. R. Seymour. 2003. Geostatistical characterization of centimetre-scale spatial structure of in vivo fluorescence. Mar. Ecol. Prog. Ser. 251:49-58.

Watson, J. 2004. HoloMar: a holographic camera for subsea imaging of plankton. Sea Tech. 45:53-55.

Wiebe, P. H., and others. BIOMAPER-II: an integrated instrument platform for coupled biological and physical measurements in coastal and oceanic regimes. IEEE J. Ocean. Engin. 27:700-716.

Wolk, F., H. Yamazaki, L. Seuront, and R. G. Lueck. 2002. A new free-fall profiler for measuring biophysical microstructure. J. Atmos. Ocean. Tech. 19:780-793.

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