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ScienceDirect
Harmful Algae 6 (2007) 837–848
HARMFUL ALGAE
www.elsevier.com/locate/hal

Towards an optimal sampling strategy for *Alexandrium catenella* (Dinophyceae) benthic resting cysts

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Received 8 November 2006; received in revised form 13 March 2007; accepted 30 April 2007

Abstract

The study proposes methodological developments to optimize sampling strategy of resting cysts of *Alexandrium catenella* to estimate their abundance with a predefined error. This work also aims to provide information on spatial distribution of resting cysts in sediments. The distribution mode of *A. catenella* resting cysts related to the abundance variability was studied through sediment cores sampling on four different spatial scales and using Ludox CLX gradient density method. The quantification method underestimates by a factor of 2 the resting cysts abundance in one gram of sediment. Application of Taylor's power law allowed us to define a compromise between sampling effort and abundance estimation error. In the case of *A. catenella* resting cysts from Thau lagoon, the optimal sampling strategy consists of sampling 10 stations on a surface of 2 km^2 for a given coefficient of variability (C) of 15%, sampling 3 sediment cores at each station ($C = 30\%$) and counting only one replicate by core ($C = 18\%$). Results related to the application of Taylor's power law are closely dependent on resting cyst density and aggregation in a given sediment. In our area, *A. catenella* resting cysts are mainly observed in the upper 3 cm of sediment. Horizontally, their heterogeneity is lower on 10 cm^2 surface and tends to stabilize itself beyond a surface of 10 m^2. Each author has to carry out this pre-sampling effort for his own resting cysts-forming species, in his own area, in order to increase accuracy of resting cyst mapping.

Keywords: *Alexandrium catenella*; Toxic dinoflagellate; Resting cyst; Optimum sample size; Spatial distribution; Vertical profiles

1. Introduction

Shellfish farming activities in Thau lagoon (South France) have been threatened since 1998 by recurrent toxic blooms of *Alexandrium catenella* (Lilly et al., 2002). The blooms seasonal pattern is related to resting forms. Like 10% of 2000 dinoflagellate species, *A. catenella* produces benthic resting cysts (Dale, 1983) whose excystment constitutes the starting point of their planktonic life cycle. "Cyst banks" of sediments play a key role in the bloom initiation phase (Anderson et al., 1982; Garcés et al., 1999; McGillicuddy et al., 2003). Understanding the initiation mechanism implies the acquisition of robust data on resting cysts distribution and density.

Resting cysts are mainly distributed at the sediment surface (Irwin et al., 2003; Garcés et al., 2004; Mizushima and Matsuoka, 2004). Their concentration increases in muddy sediment with water and organic matter contents (Erard-Le-Denn et al., 1993; Yamaguchi et al., 1996). However, the observed variations in resting cysts distribution and density could be linked to the encystment capability of planktonic species in the water column, sedimentation, transport

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1568-9883/$ – see front matter
doi:10.1016/j.hal.2007.04.007

Table 1
Sampling methods used to study the distribution of *Alexandrium* sp. benthic resting cysts (RC) in sediment

Alexandrium sp.	Sites	Sampling	Layer (cm)	Sediment preparation method	RC density	Unit**	References
A. tamarense	USA	Hand coring	*	Sonication and sieving (80–20 μm or 64–20 μm)	700 to 800	a	Anderson et al. (1982)
A. fundyense	USA	Hydraulically damped corer	0–3	Sonication and sieving (75–20 μm), primuline staining direct count	Up to 635	a	Anderson et al. (2005)
A. tamarense/catenella	China	Gravity core sampler	0–3	HCl and HF treatments, sonication and sieving (125–20 μm), primuline staining direct count	Up to 3778	b	Cho and Matsuoka (2001)
A. minutum	France	Spatula or hand-core	*	Sieving (100–60–20 μm), most probable number (MPN) technique	170 to 16 × 10^3	b	Erard-Le-Denn et al. (1993)
A. minutum	Spain	Scuba divers using core sampler	*	Sonication and sieving (100–10 μm), Ludox TM density gradient	>3270	a	Garcés et al. (2004)
A. minutum and *A. tamarense*	Sweden	Box corer	0–1	Sonication and sieving (100–25 μm)	n.a.	b	Godhe and Mcquoid (2003)
A. minutum and *A. tamarense*	India	sediment sucker and gravity corer	0–2	Sonication and sieving (100–25 μm)	n.a.	b	Godhe et al. (2000)
A. catenella	South Africa	Van Veen grab	_	Sonication and sieving (100–25 μm)	138	c	Joyce and Pitcher (2004)
A. catenella	South Africa	Van Veen grab	_	sonication and sieving (125–25 μm)	Up to 175	c	Joyce et al. (2005)
A. tamarense	Korea	Hand corer	0–2	Sonication and sieving (100–20 μm)	43 to 185	d	Kim et al. (2002)
A. catenella	France	Eckman grab sampler	0–3	sonication and Sieving (100–20 μm), Ludox CLX density gradient or MPN	Up to 175	b	Laabir et al. (2004)
A. tamarense	Celtic sea	Multicorer or Shipek grab	0–0.5	Sieving (118–20 μm), acid treatment,sodium polytungstate density gradient	n.a.	b	Marret and Scourse (2002)
A. tamarense/catenella	Japan	KK type corer	0–5	HCl and HF treatments, sieving (125–20 μm)	7	b	Matsuoka et al. (2003)
A. fundyense	USA	Craib corer	0–1	Sonication and sieving, primuline staining direct count method	Up to 400	a	McGillicuddy et al. (2003)
A. tamarense/catenella	Japan	Handy piston corer	*	Sonication and sieving (125–20 μm), primuline staining direct count method	4 to 3353	b	Mizushima and Matsuoka (2004)
A. minutum and *A. tamarense*	Sweden	Box corer	0–1	Sonication and sieving (100–25 μm)	n.a.	b	Persson et al. (2000)
A. tamarense	USA	Grab corer and mini-piston corer	0–2	HCl and HF treatment and sieving (125–10 μm)	n.a.	a	Pospelova et al. (2004)
A. tamarense	Japan	Gravity core sampler	0–3	Sonication and sieving (100–20 μm), primuline staining direct count method	800 to 1300	c	Tsujino et al. (2002)
A. tamarense/catenella	China	Gravity corer	0–2	HCl and HF treatment, sonication and sieving (125–20 μm)	Up to 398	b	Wang et al. (2004a)
A. tamarense/catenella	China	n.a.	*	HCl and HF treatment, sonication and sieving (125–20 μm)	Up to 503	b	Wang et al. (2004b)
A. tamarense/catenella	China	n.a.	0–2	Sonication and sieving (125–20 μm)	Up to 81	b	Wang et al. (2004c)
A. tamarense and *A. catenella*	Japan	Gravity corer or grab sampler	0–3	Sonication and sieving (150–20 μm), primuline staining direct count method	Up to 869	a	Yamaguchi et al. (1996)

*Vertical profile; **Resting cyst density in: cysts cm^{-3} (a), cysts g^{-1} dry sediment (b), cysts g^{-1} wet sediment (c), cysts g^{-1} of sediment (d); n.a., not available data.