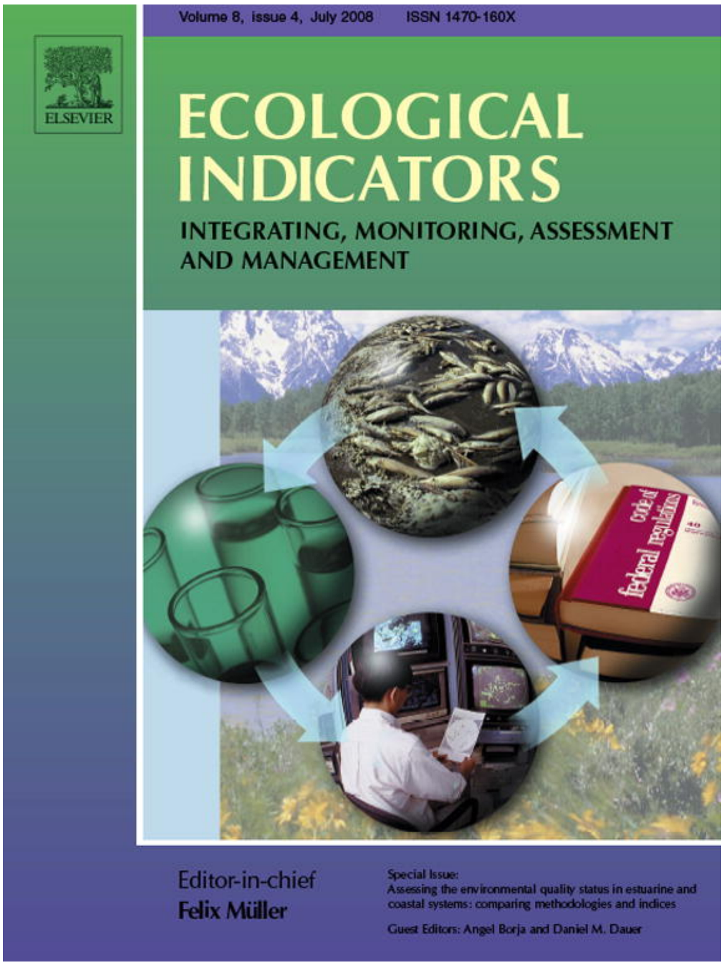
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Depth-related patterns in benthic community condition along an estuarine gradient in Chesapeake Bay, USA

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ABSTRACT

We tested whether macrobenthic community condition varies significantly with water depth in a variety of regions of Chesapeake Bay, USA. Benthic community condition was characterized using the Benthic Index of Biotic Integrity (B-IBI) previously developed for the Bay. We applied two water depth thresholds intended to emphasize the ecological importance and/or anthropogenic impacts upon shallow-water regions. The first threshold of 2 m emphasizes restoring and supporting submerged aquatic vegetation while the second threshold of 4 m emphasizes the zone of maximum anthropogenic impact upon natural ecosystem functions. An *a priori* expectation is that benthic community condition may worsen with increasing depth, specifically in regions (1) where water column stratification at depth results in prolonged low dissolved oxygen levels or (2) where net deposition at depth results in higher levels of hydrophobic, sediment-bound contaminants. Samples collected from a major tributary of Chesapeake Bay, the York River estuary, spanned the entire salinity range from tidal freshwater to polyhaline. We also tested the shallow-water depth thresholds using data from the Virginia Mainstem of Chesapeake Bay and the Southern Branch of the Elizabeth River. These two polyhaline regions are characterized as having the best and worst benthic community condition in Chesapeake Bay. At the scale of the entire tidal York River system, there were no significant differences in benthic community condition with water depth. However, two salinity regions, low mesohaline and polyhaline, had significant depth effects with the shallowest water depth zone significantly different from the other two depth regions. For the low mesohaline region benthic community condition was worse at the shallowest depth and for the polyhaline region the shallowest depth was better comparing the three depth regions. No depth-related differences in the B-IBI were found for the two additional Chesapeake Bay strata, the Virginia Mainstem characterized with the lowest levels of benthic community degradation and for the Southern branch of the Elizabeth River, characterized by the highest levels of benthic community degradation. We conclude that the ecological state of Chesapeake Bay subtidal benthic communities is adequately characterized by randomly sampling all depths without further stratification into shallow and deeper regions.

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1. Introduction

1.1. *Macrobenthic communities*

Macrobenthic communities are widely used to characterize the ecological condition of coastal and estuarine ecosystems due to their predictable response to many kinds of natural and anthropogenic stress (Pearson and Rosenberg, 1978; Dauer, 1993; Tapp et al., 1993; Wilson and Jeffrey, 1994; Dauer et al., 2000; Borja et al., 2003). Benthic communities are especially useful in detecting and evaluating the impacts of low dissolved oxygen events and aquatic contamination because exposure to anoxia/hypoxia is greatest in near-bottom waters and hydrophobic anthropogenic contaminants typically accumulate in sediments. Benthic organisms with limited mobility cannot avoid adverse conditions and better reflect local environmental conditions compared to most pelagic fauna (Gray, 1979). The diversity of physiological tolerances, life history strategies, feeding modes, and trophic interactions make sedimentary macrobenthic communities effective estimators of environmental condition (Pearson and Rosenberg, 1978; Rhoads et al., 1978; Boesch and Rosenberg, 1981; Bilyard, 1987; Dauer, 1993; Dauer et al., 2000).

In the Chesapeake Bay, the benthic monitoring program consists of both fixed-point stations and probability-based samples. The fixed-point stations are used primarily for the determination of long-term trends (e.g., Dauer and Alden, 1995; Dauer, 1997) and the probability-based samples for the determination of the areal extent of degraded benthic community condition (Llansó et al., 2003; Dauer and Llansó, 2003). The probability-based sampling design consists of equal replication of random samples among strata and is, therefore, a stratified simple random design (Kingsford, 1998). The probability-based benthic sampling program consists of 10 strata that represent (1) major tidal rivers (James, York, Rappahannock, Potomac, Patuxent), (2) major salinity regions of the mainstem of the Bay (tidal freshwater/oligohaline, mesohaline, polyhaline), (3) Maryland western tributaries (urban-impacted), and (4) Maryland eastern tributaries (agricultural-impacted) (see Fig. 1 in Dauer and Llansó, 2003). Benthic community condition is assessed using the benthic index of biotic integrity (B-IBI) that evaluates the ecological condition of a sample by comparing values of key benthic community attributes to reference values expected under undegraded conditions in similar habitat types (Weisberg et al., 1997).

1.2. *Habitat/typological delineation of estuarine/ transitional waters*

Estuaries or transitional waters are spatially and temporally complex. Assessing ecological condition using macrobenthic communities can be optimized, or may require, spatial or temporal stratification of the ecosystem for determination of ecological status (Gibson et al., 2000). Typology and habitat determination are methods for stratifying estuarine or transitional ecosystems into functional units that minimize the natural variance of these dynamic systems and allow more robust assessment of anthropogenic impacts. For example, in developing a Benthic Index of Biotic Integrity (BIBI) for Chesapeake Bay, Weisberg et al. (1997) stratified the benthos into communities occupying seven habitat types that emphasized salinity and/or sediment type. Similar stratification approaches have been used in the development and/or application of benthic community indices elsewhere in the USA (Van Dolah et al., 1999; Eaton, 2001; Llansó et al., 2002a,b). The European Water Frame Directive requires stratification of coastal and transitional water bodies into different types and the determination of reference condition for each water body type (Bald et al., 2005; Borja, 2005, 2006; Chainho et al., in press).

The US Clean Water Act (CWA) enacted in 1972 (variously amended since), was intended to "...restore and maintain the chemical, physical, and biological integrity of the Nation's water... for the protection and propagation of fish, shellfish, and wildlife and provides for recreation in and on the water..." Each State must establish designated uses to be achieved and specify water quality criteria necessary to protect the State's waters. Typically State designated uses include potable water supplies; propagation of fish, shellfish and wildlife; recreation; agricultural and industrial supplies; and navigation. Recently, estuarine ecosystems have been further stratified based upon designated water use criteria (USEPA, 2004a,b) and the importance of shallow water (Reilly et al., 1996). In assessing restoration efforts in Chesapeake Bay, five designated water use strata were defined—migratory spawning and nursery, shallow-water, open-water, deep-water and deep-channel (USEPA, 2003a,b). The shallow-water designated use category was "Designed to protect underwater bay grasses and the many fish and crab species that depend on the shallow-water habitat provided by underwater bay grass beds. Waters with this designated use support the survival, growth and propagation of rooted, underwater bay grasses necessary for the propagation and growth of balanced, indigenous populations of ecologically, recreationally and commercially important fish and shellfish inhabiting vegetated shallow-water habitats." The depth zone designated as shallow water is based exclusively upon support of submerged aquatic vegetation (SAV) and goes from the upper intertidal to the 2 m (below mean low water) contour.

Based upon extensive consideration of multiple ecological functions and values, and input from environmental managers and regulators from numerous state and federal agencies, Reilly et al. (1996) defined shallow water as "the area of maximum interaction between human activities and biological resources. It includes the intertidal zone down to four meters below mean low water (MLW) where critical functions such as biological productivity and ecological balance must be reconciled with human activities." These two shallow-water depth thresholds reflect different intents. The EPA 2 m threshold emphasizes restoring and supporting submerged aquatic vegetation (Batiuk et al., 1992, 2000), while the Reilly et al. (1996) 4 m threshold emphasizes the zone of maximum anthropogenic impact upon natural ecosystem functions.

In an adaptive monitoring approach (Ringold et al., 1996) consideration of creating separate shallow-water strata within the existing Chesapeake Bay benthic monitoring strata should be addressed. Specifically we ask whether the available benthic data indicate a differential benthic community response of shallow water versus deep water subtidal regions in regard to major natural gradients, e.g., the estuarine salinity gradient or anthropogenic impact gradients, e.g., in highly urbanized versus heavily forested regions. In this study we used data from the York River of Chesapeake Bay and examined patterns in benthic community condition at the scale of the entire river and by salinity regions within the river as defined by the Venice classification system. We also examined patterns of benthic community condition with depth from two additional locations: the Southern Branch of the Elizabeth River and the lower Mainstem of the Chesapeake Bay that respectively, have the highest and lowest levels of benthic community degradation in Chesapeake Bay (Dauer and Llansó, 2003).

2. Materials and methods

Samples of macrobenthic communities from three regions of Chesapeake Bay were used in our analyses—the York River, the Southern Branch of the Elizabeth River and the Virginia Mainstem of Chesapeake Bay (Fig. 1). The York River was selected because in 1995 samples were collected in very shallow subtidal water depths at 0.5 m (MLW) along the entire

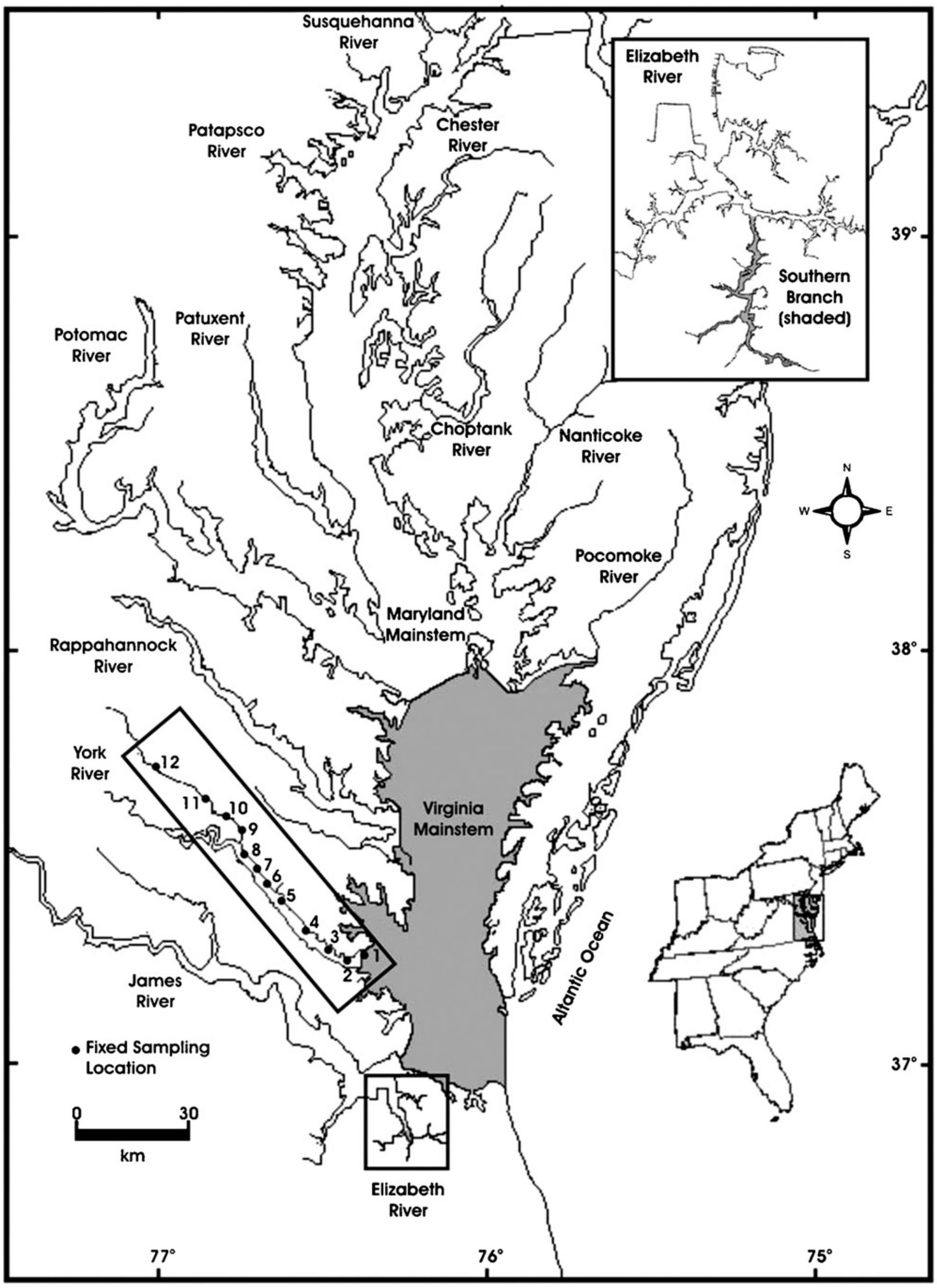

Fig. 1 – Chesapeake Bay, USA. The three study regions are indicated. The York River system with the 12 fixed-point stations indicated, the Southern Branch of the Elizabeth River (see shaded portion of insert) and the Virginia Mainstem (shaded area).

estuarine gradient from tidal freshwater to polyhaline regions. These data supplement existing data from the Chesapeake Bay Benthic Monitoring Program and provide a unique opportunity to test in a more robust manner whether shallow-water benthic community condition differs from conditions in deep-water regions within salinity regions and/or the entire estuarine gradient. Benthic community data from two additional regions with the worst (Southern Branch of the Elizabeth River) and best (Virginia Mainstem) benthic community condition in Chesapeake Bay were used to examine whether depth-related patterns exist in a highly impacted region compared to a minimally impacted region. All benthic samples were sieved on a 0.5 mm screen and preserved in the field. Samples were sorted, enumerated and identified to the lowest possible taxon. Ash-free dry weight biomass was determined for each taxon.

2.1. Fixed shallow-water stations of the York River

As part of an unpublished study comparing the distribution of intertidal and shallow subtidal benthic communities of the York River, 12 sites were selected approximately equidistantly from the polyhaline mouth of the river through the tidal freshwater region (Fig. 1). Only locations with an unvegetated intertidal zone were selected to reduce or eliminate high levels of detritus in samples. At each of the 12 sites, the macrobenthic community was sampled using three paired sets of intertidal and subtidal replicates located at 0.5 m above and below the mean tidal level. The intertidal sample locations at each of the 12 locations were selected using random horizontal distances (selected *a priori* using random number generator software) from the nearest intertidal salt marsh stand. Subtidal samples were taken directly offshore of each intertidal location at the appropriate water depth. Data for the intertidal samples were not used in our study. Samples for macrobenthos were collected using a hand-held PVC coring device. The coring device had a 10 cm internal diameter (surface area of 78.5 cm^2) and was pushed into the sediment to a depth of 25 cm. All samples were collected in 1995. A total of 36 samples collected at 0.5 m (MLW) were used in our study.

2.2. Probabilistic (random) stations

The Chesapeake Bay Benthic Monitoring Program has collected macrobenthic community data on a bay-wide scale since 1985. In 1996, a probability-based sampling program was added. Ten strata were defined (see Fig. 1 in Dauer and Llansó, 2003) and 25 random sites were allocated to each stratum. Each year 25 new locations are selected for each of the 10 strata. By combining probabilistic allocation of samples and applying the Chesapeake Bay Benthic Index of Biotic Integrity (see Section 2.3) an integrated assessment of the Bay's overall condition can be made (Brown et al., 2005). In addition, the condition of each stratum and an unbiased comparison of conditions between strata (e.g., tributaries) of the Chesapeake Bay is provided by estimating the areal extent of degradation at each of these spatial scales (e.g., see Table 1 in Dauer and Llansó, 2003). In the York River, 3 years of random samples (1996–1999, $n = 75$) were used in the comparisons with the 0.5 m (MLW) samples (see Section 2.1).

To further clarify interpretations of depth-related patterns of benthic community condition in the York River, we also tested such patterns in two additional regions of Chesapeake Bay—the Southern Branch of the Elizabeth River and the Virginia Mainstem of the Bay. In both regions, salinity ranges from high mesohaline to polyhaline with average bottom water values (±S.E.) for the Southern Branch and Virginia Mainstem of 19.8 ± 0.2 and 22.4 ± 0.3, respectively. These regions were chosen because they represent the regions of the worst and best benthic community condition. As presented by Dauer and Llansó (2003), the areal extent of degradation using 1999 data was 47% for the entire Chesapeake Bay, 92% for the Southern Branch of the Elizabeth River and 20% for the Virginia Mainstem.

The Elizabeth River, a highly urbanized and industrialized watershed, is heavily contaminated with PAHs and heavy metals (Helz and Hugget, 1987; Hall et al., 2002; Walker and Dickhut, 2001; Conrad and Chisholm-Brause, 2004; Walker et al., 2004) and has a degraded benthic community condition (Hawthorne and Dauer, 1983; Dauer, 1993; Dauer et al., 1993, 2000) with the worst levels of degradation in the Southern Branch of the river (Dauer and Llansó, 2003). The data we used are part of the Elizabeth River Benthic Monitoring Program initiated in 1999 (Dauer, 2000). In 1999, the macrobenthic communities of the Elizabeth River were sampled using five strata each of which was sampled at 25 sites with the Southern Branch constituting one of the five strata (Dauer, 2000; see Fig. 2 in Dauer and Llansó, 2003). In subsequent years, the entire watershed (combining the five original strata) was sampled at 25 randomly selected sites. Data for the 3-year period 1999–2001 were used in our analyses including three tidal creeks of the Southern Branch that were sampled as separate strata with 25 random samples—Scuffletown Creek sampled in 1999, Jones and Gilligan creeks sampled in 1999 and Paradise Creek sampled in 2001 (for locations see Fig. 2 in Dauer and Llansó, 2003). This resulted in 111 samples for the Southern Branch of the Elizabeth River.

The Virginia Mainstem of Chesapeake Bay has no significant levels of chemical contamination, eutrophication or bottom low dissolved oxygen events (Dauer et al., 1993) and, consequently, the condition of the benthic communities is characterized as the best for the Chesapeake Bay (Dauer and Llansó, 2003). In order to attain at least 10 samples for the $\leq$2.0 m depth interval, 4 years of data were used (1999–2002). For the years 1996–1998 none of the 25 random samples collected each year were $\leq$2.0 m in water depth. This resulted in 100 samples for the Virginia Mainstem of Chesapeake Bay.

2.3. Chesapeake Bay Benthic Index of Biotic Integrity

The Benthic Index of Biotic Integrity (B-IBI) was developed to assess the condition of macrobenthic communities of the Chesapeake Bay (Weisberg et al., 1997; Alden et al., 2002). The index defines expected conditions at reference sites relatively free of anthropogenic stress and then assigns values for various descriptive metrics by comparison with observations at these reference sites. The result is a multi-metric index of biotic condition frequently referred to as an index of biotic integrity (IBI). Selection of benthic community metrics and metric scoring thresholds were habitat-dependent but by using categorical scoring, comparisons between habitat types were possible. Overall, the index correctly distinguished degraded sites from reference sites 93% of the time. The results of Alden et al. (2002) indicated that the B-IBI is sensitive, stable, robust, and statistically sound.

2.4. Data analysis

For tests with three depth intervals one-way ANOVAs and a post hoc Duncan's test were used. When only two depth intervals were tested, a *t*-test with a Bonferroni correction for multiple tests was used. Sample assignment to salinity regimes was based on a modified Venice system classification with the tidal freshwater and oligohaline regions combined because of low sample number. The salinity regions were tidal freshwater and oligohaline (0.5–5.0 psu), low mesohaline (≥5.0–12.0 psu), high mesohaline (≥12.0–18.0) and polyhaline (≥18.0 psu). All statistical analyses were conducted using SPSS© (v. 14) statistical analytical software.

3. Results

At the scale of the entire tidal York River system, shallow-water benthic community condition showed no significant differences (Table 1). However, two salinity regions, low mesohaline and polyhaline, had significant depth effects (ANOVA) with the shallowest depth (0.5 m) significantly different from deep-water regions using either the 2 or 4 m shallow-water threshold. However, there were contrasting differences. For the low mesohaline region benthic community condition was worse at the shallowest depth and for the polyhaline region the shallowest depth was better comparing the three depth regions.

For the two salinity regions with significant depth-related differences, i.e., the low mesohaline and the polyhaline regions, we also tested whether the combined shallow-water region (0.5 m plus the 0.5–2.0 m or 0.5 m plus the 0.5–4.0 m) differed significantly from the deep-water region (either >2.0 m or >4.0 m). For the low mesohaline region, the B-IBI value (mean ± one standard error) of 1.8 ± 0.42 for the combined shallow-water region (depths ≤2.0 m) was lower but not significantly different from the value of 2.7 ± 0.15 for the deep-water water region (depths > 2.0 m) (*t*-test, $P = 0.09$; Bonferrroni correction for multiple comparisons requires a $P < 0.01$ for a nominal value of $P = 0.05$). Similarly, using the 4.0 m threshold for the low mesohaline region, the B-IBI value of 2.0 ± 0.40 for the combined shallow-water region (depths ≤4.0 m) was lower but not significantly different from the value of 2.7 ± 0.15 for the deep-water region (depths > 4.0 m) (*t*-test, $P = 0.16$; Bonferrroni correction for multiple comparisons requires a $P < 0.01$ for a nominal value of $P = 0.05$). For the polyhaline region, the B-IBI value of 2.9 ± 0.10 for the combined shallow-water regions (depths ≤2.0 m) was significantly different and higher than the value of 2.3 ± 0.13 for the deep-water region (depths > 2.0 m) (*t*-test, $P = 0.001$; Bonferrroni correction for multiple comparisons requires a

Table 1 – Average B-IBI values ± one standard error (sample size in parentheses) of 0.5 m samples compared to random samples collected shallower or deeper than the two depth thresholds (2.0 or 4.0 m)

Region	Depth regions			Test statistic and P value
	0.5 m	0.5–2.0 m	>2.0 m	
(A) Shallow-water depth threshold of 2.0 m				
Entire York River	2.9 ± 0.12(36)	2.6 ± 0.10(22)	2.5 ± 0.12(53)	F = 2.684; P = 0.073
Tidal freshwater/oligohaline	3.5 ± 0.29(6)	3.0(1)	3.4 ± 0.31(7)	F = 0.205; P = 0.818
Low mesohaline	1.3 ± 0.13(3) a	2.6 ± 0.8(2) b	2.7 ± 0.13(6) b	**F = 9.294; P = 0.008**
High mesohaline	2.6 ± 0.17(9)	2.5 ± 019(10)	2.4 ± 0.12(24)	F = 0.344; P = 0.711
Polyhaline	3.1 ± 0.09(18) a	2.6 ± 0.25(9) b	2.3 ± 0.13(16) b	**F = 9.640; P < 0.001**
Southern Branch	*	2.0 ± 0.06(72)	2.0 ± 0.10(39)	t = 0.088; P = 0.930
Virginia Mainstem	*	3.0 ± 0.19(14)	3.3 ± 0.08(86)	t = −1.614; P = 0.110
Region	**Depth intervals**			**Test statistic and P value**
	0.5 m	0.5–4.0 m	>4.0 m	
(B) Shallow-water depth threshold of 4.0 m				
Entire York River	2.9 ± 0.12(36)	2.6 ± 0.10(38)	2.5 ± 0.12(37)	F = 2.989; P = 0.055
Tidal freshwater/oligohaline	3.5 ± 0.29(6)	3.3 ± 0.25(2)	3.4 ± 0.27(6)	F = 0.111; P = 0.896
Low mesohaline	1.3 ± 0.13(3) a	2.7 ± 0.48(3) b	2.7 ± 0.15(5) b	**F = 9.247; P = 0.008**
High mesohaline	2.6 ± 0.17(9)	2.6 ± 0.13(20)	2.3 ± 0.16(14)	F = 1.088; P = 0.347
Polyhaline	3.1 ± 0.09(18) a	2.6 ± 0.17(13) b	2.2 ± 0.17(12) b	**F = 10.290; P = < 0.001**
Southern Branch	*	2.0 ± 0.06(88)	2.0 ± 0.13(23)	t = −0.269; P = 0.788
Virginia Mainstem	*	3.1 ± 0.14(25)	3.3 ± 0.09(75)	t = −1.336; P = 0.185

Shown in the last column are ANOVA and *t*-test results comparing (1) the entire York River, (2) salinity regions within the York River, (3) the Southern Branch of the Elizabeth River and (4) the Virginia Mainstem of Chesapeake Bay. The last two regions have the highest and lowest levels, respectively, of benthic community degradation in Chesapeake Bay (see Table 1 in Dauer and Llansó, 2003). Tidal freshwater and oligohaline regions of the York River were combined due to small sample size. Statistically significant ($P < 0.05$) ANOVAs are indicated by bold figures in the final column. B-IBI values with the different letters are not significantly different using a Duncan's post hoc test ($P < 0.05$).
* No data.

$P < 0.01$ for a nominal value of $P = 0.05$). Similarly, using the 4.0 m threshold for the polyhaline region, the B-IBI value of 2.9 ± 0.10 for the combined shallow-water region (depths $\leq$4.0 m) was significantly different and higher than the value of 2.2 ± 0.17 for the deep-water region (depths >4.0 m) (t-test, $P = 0.001$; Bonferrroni correction for multiple comparisons requires a $P < 0.01$ for a nominal value of $P = 0.05$).

No depth-related differences in the B-IBI were found for the two additional Chesapeake Bay strata, for the Southern Branch of the Elizabeth River, characterized by the highest levels of benthic community degradation or the Virginia Mainstem characterized with the lowest levels of benthic community degradation In Chesapeake Bay (Table 1).

4. Discussion

In designing estuarine and coastal monitoring programs, delineation of habitat types or typology typically includes an initial subdivision of the ecosystem based upon physico-chemical parameters. The most commonly used parameters are salinity gradients, water depth, and sediment type (Rakocinski et al., 1997; Gibson et al., 2000). Depth characterization is important for evaluating spatial patterns of dissolved oxygen, temperature, salinity, water density, and the size of the photic zone. Water depth stratification may be especially significant in coastal areas where bathymetry changes are great, i.e. glacially created systems such as Puget Sound, Washington, USA (Lie, 1974; Llansó, 1999). However, depth-related changes in dissolved oxygen can be important in coastal plain or barrier island estuaries when density stratification of the water column occurs (Malone, 1992; Bishop et al., 2006). In relation to estuarine and coastal benthic community ecology, water flow can vary greatly with water depth (Caeiro et al., 2005) and through patterns of erosion and deposition determine the spatial distribution of sediment types, deposited organic matter, and deposited contaminants associated with fine-sized materials (Conrad and Chisholm-Brause, 2004). Quite clearly potential impacts from land runoff (e.g., nutrients, contaminants, sediments) potentially affect shallow-water benthic habitats more frequently and with greater intensity than deep-water regions. In contrast, deep-water water benthic communities that are below the pycnocline are more frequently affected by low dissolved oxygen events (Holland et al., 1987; Malone, 1992; Dauer et al., 1992) and benthic communities associated with depositional regions (independent of water depth) can have greater exposure to higher sediment contaminant levels (Brown et al., 2000).

In an adaptive monitoring sense (Ringold et al., 1996), the benthic monitoring program of the Chesapeake Bay has been modified from an initial program consisting of fixed-point stations sampled frequently within each year to a program with (1) fixed-point stations (sampled with a reduced within year frequency) and (2) probability-based sampling within 10 strata during a summer index period. The fixed-point stations are used primarily for the determination of long-term trends (e.g., Dauer and Alden, 1995; Dauer, 1997) and the probability-based samples for the determination of the areal extent of degraded benthic community condition (Llansó et al., 2003; Dauer and Llansó, 2003). The latter data provide environmental managers with areal estimates of benthic degradation with known confidence intervals and such data can be used to both set restoration goals and assess restoration success. In an adaptive monitoring approach, we examined whether shallow-water benthic community condition differs significantly from deep-water regions in regard to either important natural estuarine gradients (salinity) or within regions of high (Southern Branch of the Elizabeth River) or low (Virginia Mainstem) anthropogenic impact. Consistent differences in depth-related patterns of benthic community condition would merit consideration of creating separate shallow-water strata within the existing Chesapeake Bay benthic monitoring strata.

Previously, Llansó et al. (2003) examined patterns of water depth and benthic community condition as indicated by the B-IBI in Chesapeake Bay. A logistic regression model was used to describe the relationship between depth and the probability of degraded condition within 67 segments of the Bay. The model has a binomial outcome that distinguishes between two conditions (degraded condition and ''otherwise'') with depth as the independent variable. Only 5 of 67 segments revealed increased probability of degraded benthos with increasing depth and these five regions are known to experience hypoxia or to have significant toxic contamination. In the present study, we examined depth-related differences in benthic community condition using specific shallow-water depth thresholds instead of testing depth as a continuous independent variable as in Llansó et al. (2003). Our study examined differences across a boundary rather than whether there was a significant relationship with depth. A region could have a significant regression between benthic community condition and water depth and still not differ across a specific depth threshold. Also regions with no significant relationships with water depth using a regression approach could still differ across a specific depth threshold. In our study, few depth-related differences in benthic community condition were detected and we conclude that an adaptive monitoring change (sensu Ringold et al., 1996) of stratification into shallow and stratification into shallow and deeper regions regions is not required for the existing probability-based benthic monitoring program.

5. Conclusions

We tested whether there were significant differences in benthic community condition of subtidal regions of Chesapeake Bay relative to two shallow-water thresholds depths of 2 or 4 m (MLW). These thresholds are intended to emphasize the restoration of submerged aquatic vegetation (2 m) or the zone of maximum anthropogenic impact (4 m). Few significant differences were detected and we conclude that the ecological state of Chesapeake Bay subtidal benthic communities is adequately characterized by randomly sampling all depths without further stratification into shallow and deep-water regions.

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