5-MHz acoustic-backscatter measurements of *Cochlodinium polykrikoides* blooms in Korean coastal waters

Eunhye Kim, Hyungbeen Lee, Jungyul Na, Jee Woong Choi, and Donhyug Kang


The toxic, chain-forming dinoflagellate *Cochlodinium polykrikoides* blooms annually in the coastal waters of Korea and has a serious impact on coastal fisheries and the surrounding ecosystem. Measurements of the acoustic volume-backscattering strength of *C. polykrikoides* blooms were made using 5 MHz signals at sites located off the coast of Geumo Island, Korea. An optical microscopic method was used to estimate the abundance of phytoplankton in coastal areas from water samples collected simultaneously with the acoustic measurements. Background scattering strengths at sites where there were no blooms were less than $-42$ dB re 1 m$^{-1}$. In contrast, backscattering strengths measured in bloom areas increased rapidly, showing a high correlation with dinoflagellate concentrations. The measured volume-backscattering strengths were compared with those predicted from a fluid-sphere scattering model; the results implied that the chain-forming cells should be viewed as a single sphere with an equivalent radius. The results suggest that it may be possible to apply an acoustic technique to the real-time detection of harmful algal blooms.

**Keywords:** acoustic volume-backscattering strengths, harmful algal blooms, real-time monitoring.

Received 7 August 2009; accepted 30 May 2010; advance access publication 4 August 2010.

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**Introduction**

Harmful algal blooms (HABs), defined as the rapid growth and accumulation of toxic microscopic algae, have serious impacts on human health and local economies as well as on living marine resources. Recently, the incidence of HABs has been increasing worldwide (Rhodes et al., 2001; Lee and Lee, 2005; Rabalais et al., 2009). One of the causative HAB species in Korean coastal waters is the chain-forming, toxic dinoflagellate *Cochlodinium polykrikoides*, and it blooms densely and frequently, especially from July through October, off Korea’s southern coasts, seriously affecting commercial fisheries.

Many techniques have been suggested for monitoring and detecting HABs. The microscopic examination of water samples is one traditional method. Although it has the advantage of identifying species composition, it is time-consuming and cannot be used to detect HABs in their initial bloom stages. Spectrophotometric and fluorometric analyses of Chl $a$ can be used to provide the biomass estimates of phytoplankton (Holm-Hansen et al., 1965; Marker et al., 1980; Dijkman et al., 1999). However, these analytical methods are not in situ techniques for monitoring HABs in real time. Satellite remote sensing has been widely used in Korea since the 1990s to monitor red tides, which cause the sea surface to appear discoloured. Although this technique can be used to monitor large areas, it is impossible to use remote sensing to detect HABs at low cell densities because of insufficient sea-surface discoloration in early-bloom stages. Several techniques for the real-time in situ measurements of HABs have recently been developed. For example, an optical phytoplankton discriminator was used to detect *Karenia* spp. in mixed phytoplankton communities by Robbins et al. (2006). Further, laser in situ scattering and transmissometry (LISST), which was developed to measure the size distribution of suspended particles, has been applied to monitor the short-term spatial and temporal changes in an *Alexandrium taylorii* bloom (Angles et al., 2008).

Here, the real-time acoustic-backscatter measurements from *C. polykrikoides* aggregations are presented as an alternative method for detecting and monitoring HABs. Acoustic observations have been used widely to investigate the abundance and distribution of zooplankton, which are in the order of centimetres to millimetres in size, with frequencies of several tens to hundreds of kilohertz (Greenlaw, 1977; Foote and Stanton, 2000; Holliday et al., 2003). In contrast, most phytoplankton organisms have cell sizes in the range 1–200 $\mu$m (Medwin and Clay, 1998), so ultrasonic frequencies in MHz need to be used. Blanc et al. (1998, 2000) measured the volume-scattering strengths of the diatom *Skeletonema costatum*, which has an equivalent radius in the range 1–6 $\mu$m, using 2.6-MHz acoustic signals in the laboratory. They then compared the results with those predicted by the fluid-sphere scattering model suggested by Johnson (1977). Here, we present backscattering strengths measured at a frequency of 5 MHz in the upper layer of the coastal waters where *C. polykrikoides* blooms. The measurements are compared with those predicted by the fluid-sphere scattering model using the single-cell size of *C. polykrikoides* and the equivalent cell size of a sphere the same size as the chain-forming cells.
Transducer calibration

Ultrasonic transducers with megahertz resonance frequency are often operated in the nearfield region of the transducer because of the range limitations caused by attenuation in the sound-propagation medium (Ma et al., 1987; Downing et al., 1995). Therefore, there is value in investigating the range-dependence of the transmitted signal in the nearfield region. Calibration experiments were performed to estimate the receiving voltage sensitivity (RVS), beam width of a 5-MHz transducer (A308S, PANAMETRICS), along with the transmission loss in the nearfield. The transducer was set at the middle depth of the water tank (0.73 m × 0.36 m × 0.44 m). A needle hydrophone (TNU001A, NTR Systems, Inc., RVS = 242.05 dB re 1 V μPa⁻¹) was mounted at the same depth and moved out at intervals of 0.5 mm along the on-axis direction of the transducer from 0.06 to 0.5 m. Continuous wave (CW) signals with 0.5-μs pulse length generated by a pulser/receiver (5072PR, PANAMETRICS) were transmitted. The signals were received by the needle hydrophone, amplified by 30 dB with a preamplifier (PFS017A, NTR Systems, Inc.), and digitized at a sampling rate of 100 MHz.

The critical distance between the nearfield and the far field is defined as $\pi a^2/l$, where $a$ and $l$ represent the radius of the radiating face area of the transducer and the acoustic wavelength, respectively (Thorne et al., 1991; Medwin and Clay, 1998), which was estimated to be ~1 m. Therefore, in a range of <0.5 m, the squared voltage expressed in the unit of dBV does not follow the curve of the spherical spreading loss (Figure 1), which is specified as $20 \log r + \alpha r + K$, where $K$ is an arbitrary decibel offset.

Field measurements

Acoustic measurements were made in summer 2008 in an experimental area off the coast of Geumo Island in southern Korea, where HABs have been observed frequently in summer in recent years (Figure 3). Experiments were conducted between 11:00 and 16:00 local time (UTC +9 h), because *C. polykrikoides* ascends to a depth range of 0–5 m by day to photosynthesize (Park et al., 2001). The sea state was calm, and the windspeed was <4 m s⁻¹ during the course of the measurements.

The 5-MHz transducer was deployed at a depth of two metres from the side of a small fishing vessel to avoid the scattering effect caused by microbubbles near the sea surface, the concentration of which decreases exponentially with depth, with an e-folding depth of ~1.5 m (Crawford and Farmer, 1987). In all, 50 repetitions of 0.5-μs CW pulses at time intervals of 10 ms were transmitted, and the process was repeated every 20 s, with the transducer being towed at a steady speed of two knots. The routes of the vessel taken on 16 July, 4 August, and 6 August, and the locations of the discrete sampling points (hereafter, sites) from which the acoustic and environmental data were obtained, are shown in Figure 3.

Conductivity–temperature–depth casts were made just before the acoustic measurements to monitor sound speed, water temperature, and water density at each site. Water samples were collected using a Van Dorn sampler deployed at a depth of two metres at each site at the same time as the acoustic measurements were taken. The concentration of suspended particulate matter (SPM) and the distribution of plankton in the water samples were recorded.
To determine the SPM concentration, water samples (1000 ml) were filtered through preweighed glassfibre filters (Whatman, GF/C). The filters were dried for 12 h at 100°C, then reweighed. The concentration of suspended matter in the water sample was estimated from the increase in filter weight.

Water samples from sites where *C. polykrikoides* was blooming were covered with aluminium foil and transported immediately to the laboratory (within 2 h) without the use of a fixing solution. However, water samples from sites where there was no *C. polykrikoides* bloom were fixed in situ with Lugol’s solution.

Cell counts for seawater samples with abundant *C. polykrikoides* cells were conducted using 1-ml subsamples taken directly from well-stirred, 500-ml water samples. However, the fixed, 500-ml water samples with less abundant *C. polykrikoides* cells were condensed to 5 ml, from which 1-ml subsamples were taken. Cell counts were then made using a Sedgwick Rafter chamber at ×100–400 magnification under an optical microscope (Nikon/Olympus BX50).

For Chl *a* analysis, water samples were filtered onto 47-mm GF/F Whatman filters, and the filters were immersed in 90% acetone–water (v/v) solution and centrifuged. Centrifuge extracts were analysed with a spectrophotometer (Mecasys Optizen 2120UV).

**Measurement results**

The sonar equation used to estimate the volume-backscattering strength in the far field is defined by Urick (1983) as

\[ S_v = RL - SL + 2TL - 10 \log_{10} V, \]  

where \( S_v \) is the volume-backscattering strength (in dB re 1 m\(^{-1}\)) over the insonified volume \( V \), RL the received intensity level (in dB re 1 μPa), and SL the source level (in dB re 1 μPa at 1 m). TL is the one-way transmission loss (in dB) from the source to the insonified volume. With SPL\(_n\) measured during the calibration process, Equation (1) is converted to backscattering strength in the nearfield:

\[ S_v = RL - \text{SPL}_n + TL_2 - 10 \log_{10} V, \]  

where TL\(_2\) is the transmission loss from the insonified volume to the transducer, which is assumed to be 20 log\(_{10}\) \( r + \sigma r \). \( V \) is associated with the equivalent beam angle \( \psi \) of the transducer and the pulse length \( \tau \) of the source waveform and is calculated (Urick, 1983; Simmonds, 1984) from

\[ V = c \tau \psi^2 / 2, \]  

where \( c \) is the compressional wave speed in water. The equivalent beam angle was estimated from the measured beam pattern (Figure 2b) using the method suggested by Reynisson (1998). Scattering strengths were estimated from 400 segments of the backscattered signals corresponding to a range 0.30–0.45 m from the source. To avoid overestimation through contamination from signals scattered from other sources such as microbubbles, turbulence from vessel movement, and zooplankton, the intensity levels of each segment for 50 pings were compared with 2.58 \( \sigma \) (where \( \sigma \) is the s.d.), and deviant signals were rejected based on Chauvenet’s criterion (Taylor, 1997). The remaining intensity segments were averaged to estimate RL. Finally, a background-noise correction was made to minimize contamination from the system plus ambient noise, which was measured before volume-scattering measurements and bandpass-filtered with a 3-dB bandwidth of 4 MHz centred at 5 MHz. The measured background noise levels were 1–3 dB less than the RL obtained from the signals scattered from normal seawater without *C. polykrikoides* blooms. As the backscattered signals are the random signals with an amplitude distribution following a Rayleigh probability density function, Equation (1) can be expressed as
density function, the intensity average for 50 realizations is expected to have a Gaussian distribution with an s.d. of 0.8 dB. Then, because the backscattering strengths were the average values from 400 segments, the error range \( \Delta x \) was reduced to 0.1 dB. As the systematic error \( \Delta y \) was estimated to be 0.3 dB by the transducer-calibration experiment, the total measurement error was expected to be less than \( +0.3 \) dB, which can be estimated using \( \sqrt{\Delta x^2 + \Delta y^2} \), under the assumption that the intensity of the backscattering signals and system noise are independent of each other and are both governed by the Gaussian distribution.

Phytoplankton abundance at each site is presented in Table 1 (see also Figure 4). Diatoms were the most abundant species at sites 1–4, ranging from \( 0.27 \times 10^5 \) to \( 0.55 \times 10^5 \) cells l\(^{-1}\), whereas dinoflagellates were rare at these sites. Diatom abundances at sites 5–7 were more than twice as high as at sites 1–4, representing \( \sim 98\% \) of the total phytoplankton numbers, except at site 6. For the samples obtained on 6 August 2008, total phytoplankton numbers were \( 3.38 \times 10^5 \) cells l\(^{-1}\) and dinoflagellate abundance was twice that of diatoms, the volume-backscattering strength was greater than \( 240 \) dB re 1 m\(^2\). At sites 8 and 9, where red tides have been seen, backscattering strengths were greater than \( 237 \) dB re 1 m\(^2\), showing a rapid increase with the abundance of phytoplankton. The results of the backscattering-strength analyses corresponding to cell numbers \( >2.0 \times 10^5 \) cells l\(^{-1}\) showed that backscattering strengths increased with a slope of \( \sim 7 \) dB per decade increase in cell numbers.

Comparison with theory

To investigate the acoustic behaviour of HABs, the measured backscattering strengths were compared with those predicted by a

![Figure 4](https://academic.oup.com/icesjms/article-abstract/67/8/1759/606728)

**Figure 4.** Numerical abundance of diatoms (black) and dinoflagellates (white) during the acoustic measurements.

![Figure 5](https://academic.oup.com/icesjms/article-abstract/67/8/1759/606728)

**Figure 5.** Chl-a concentration as a function of the abundance of dinoflagellates and total phytoplankton.

Table 1. Environmental data collected from the various sites (1–9) and the phytoplankton abundance determined by microscopic observation.

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<th>Parameter</th>
<th>Track A</th>
<th>Track B</th>
<th>Track C</th>
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<td>Date</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>16 July 2008</td>
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<tr>
<td>Local time</td>
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<tr>
<td>11:15</td>
<td>12:14</td>
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<td>14:18</td>
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<tr>
<td>Water temperature (°C)</td>
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<td>–</td>
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<td>25.4</td>
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<tr>
<td>Water density (g cm(^{-3}))</td>
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<tr>
<td>–</td>
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<td>1.021</td>
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<tr>
<td>Chl a (µg l(^{-1}))</td>
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<tr>
<td>3.8</td>
<td>2.4</td>
<td>0.9</td>
<td>3.8</td>
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<tr>
<td>SPM (mg l(^{-1}))</td>
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</tr>
<tr>
<td>7.4</td>
<td>10.5</td>
<td>7.8</td>
<td>5.4</td>
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<tr>
<td>Phytoplankton abundance (( \times 10^5 ) cells l(^{-1}))</td>
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<tr>
<td>Total</td>
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<td>0.55</td>
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<tr>
<td>Diatom</td>
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<td>0.53</td>
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<tr>
<td>Dinoflagellate (Cochlodinium)</td>
<td>0.03</td>
<td>0.02</td>
<td>0.01</td>
</tr>
<tr>
<td>Cell number of dominant chain</td>
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</table>

Comparison with theory

To investigate the acoustic behaviour of HABs, the measured backscattering strengths were compared with those predicted by a...
fluid-sphere scattering model (Johnson, 1977). The mean of the equivalent spherical radius of the dinoflagellates was estimated to be 16.0 μm with an s.d. of 2.5 μm, corresponding to the Rayleigh scatter, because the product of the acoustic wave number and the equivalent radius is <1. Note that although G. catenatum accounted for ~30% of total phytoplankton abundance during blooms, it was assumed that its acoustic behaviour was the same as that of C. polykrikoides, because the species are morphologically similar. The fluid-sphere scattering model for $ka < 1$ ($k$ and $a$ represent the acoustic wave number and the equivalent radius of the cell, respectively) is given by

$$\sigma = k^4 a^4 \left[ \frac{1 - gh^2}{3gh^2} + \frac{1 - g}{1 + 2g} \right]^2,$$

where $\sigma$ is the backscattering cross section of an individual cell, and $g$ and $h$ are the density and sound-speed ratios of the cell to the water medium, respectively (Johnson, 1977). The volume-backscattering strength $S_v$ is defined as

$$S_v = 10 \log_{10}(N\sigma),$$

where $N$ is the number of cells per unit volume. Van Ierland and Peperzak (1984) reported a density range between 1.08 and 1.12 g cm$^{-3}$ for the dinoflagellate Peridinium sp., using density-gradient centrifugation. Kamiykowski et al. (1992) documented a density range of 1.06–1.09 g cm$^{-3}$ for six dinoflagellate species using a NaCl-adjusted, iso-osmotic Percoll and the same method as that of Van Ierland and Peperzak (1984). Forman and Warren (2010) suggested that it may be important to use a range of values for $g$ and $h$ to convert acoustic data into estimates of the abundance of marine organisms. As it was not possible to measure the density of C. polykrikoides, a density-contrast range of 1.04–1.10 (mean: 1.07) was assumed, using 1.02 g cm$^{-3}$ as the water density. Holliday and Pieper (1980) and Blanc et al. (2000) predicted volume-scattering strengths using density and sound-speed contrasts 4–8% higher than the measured values to conduct a better comparison with the scattering strengths measured from crustaceans with thin carapaces (e.g. copepods, euphausiids, and sergestid shrimps) and phytoplankton with siliceous thecae (e.g. diatoms), respectively. However, C. polykrikoides and G. catenatum belong to the group of unarmoured dinoflagellates (Okaichi, 2004), so no modification of density or sound-speed contrasts was considered. Cochlodinium polykrikoides and G. catenatum are chain-forming dinoflagellates that create chains of 2, 4, 8, and sometimes up to 16 cells during blooms (Carrada et al., 1991; Kim et al., 2007). The number of cells forming the dominant chain at each site is shown in Table 1. Chain-forming cells can be considered as single bodies under the assumption that the sound-scattering behaviour of a non-spherical body smaller than the acoustic wavelength is equivalent to that of a sphere of the same volume (Medwin and Clay, 1998). The radius of an equivalent sphere is estimated as $\sqrt[3]{Na}$. Because the backscattering coefficient is proportional to the sixth power of the radius, that of chain-forming cells becomes much higher in proportion to $N^2$. Finally, the backscattering strength (hereafter referred to as the chain-scattering model) can be estimated from

$$S_v = 10 \log_{10}\left( \sum_{i=1}^{5} M_i N_i \sigma \right) = 10 \log_{10}\left( \sum_{i=1}^{5} M_i \sigma \right),$$

where $\sigma$ is the backscattering cross section of the chain considered as a single body, $N_i$ the number of cells in a chain (1, 2, 4, 8, or 16), and $M_1$ its number.

Comparisons of the measured backscattering strengths and model predictions are shown in Figure 6. Using a density-contrast range of 1.04–1.10 and Equation (5), the sound-speed contrast $h$ must be in the range 1.4–1.6 to achieve agreement between the measurements and model predictions. These values are much higher than those reported in the literature, including those for zooplankton and phytoplankton, which range from 1.00 to 1.07 (Medwin and Clay, 1998; Blanc et al., 2000; Mukai et al., 2004). However, comparisons with the backscattering strengths predicted by the chain-scattering model with a $g$ range of 1.04–1.10 give a sound-speed contrast range of 1.03–1.10 for sites 6, 8, and 9. The squares in Figure 6 indicate the backscattering strengths predicted by the chain-scattering model when $h = 1.06$ for the mean density-contrast value ($g = 1.07$), which is the best fit to the measurements. However, using the same values of $g$ and $h$, the simple fluid-sphere scattering model [Equation (5)] yields predictions that are much lower than the values measured, as shown by the solid line in Figure 6.

Summary and discussion

We have presented the results of volume-backscattering strength measurements made in HABs off the coast of Geumdo Island, southern Korea. The most abundant phytoplankton in this area was the dinoflagellate C. polykrikoides, which accounted for ~70% of total phytoplankton abundance, with another dinoflagellate, G. catenatum, accounting for the balance. The measured backscattering strengths seemed to be well correlated with dinoflagellate concentrations. In normal seawater, where diatoms were most abundant, backscattering strengths ranged between $-42$ and $-46$ dB re 1 m$^{-1}$, which was not proportional to the logarithmic number of cells per unit volume. It is postulated that SPM is a major contributor to the 5-MHz background scattering strengths in normal seawater without blooms of C. polykrikoides (Table 1). When the total abundance of

![Figure 6](https://academic.oup.com/icesjms/article-abstract/67/8/1759/606728/1763)
phytoplankton exceeded $>3.0 \times 10^5$ cells l$^{-1}$, which corresponds to the cases of sites 6, 8, and 9, backscattering strengths increased rapidly, with a slope of $\sim 7$ dB per decade increase in cell numbers (as shown in Figure 6), consistent with the predictions obtained using the chain-scattering model.

Measured backscattering strengths were compared with those predicted by scattering models, and the results implied that the backscattering coefficient of chain-forming phytoplankton was in proportion to the square of the number of cells in the chain, showing the best fit when $h = 1.06$ for the mean density-contrast value ($g = 1.07$) reported in the literature. The model comparisons for the case without consideration for the cell chain, however, required impractically high sound-speed contrast.

Other possible scattering sources, such as microbubbles, turbulence from vessel movement, and zooplankton, may contaminate the results of bloom-causing phytoplankton scattering-strength measurements. The radius of a microbubble with a resonant frequency at 5 MHz is $\sim 0.7$ μm, and the probability that a 0.7-μm bubble occurred in the insonified volume was <5% when the windspeed was < 4 m s$^{-1}$, according to the measurements of aerosol particle distribution reported by Piazzola and Despiau (1997). Moreover, the target strength of a 0.7-μm bubble is estimated to be less than $-86$ dB re 1 m$^2$. Given that the transducer-towing conditions, including towing speed and depth, were very stable over the measurement period, it was assumed that the scattering effect of the turbulence caused by the vessel’s movement was the same for each site and was included in the background level. The dominant species of zooplankton in the southern coastal waters of Korea are copepods, and the average abundance is reported to be 100–300 ind. m$^{-3}$ (Kang et al., 2000). The probability of copepods occurring in the insonified volume, which was estimated to range from $\sim 1.8 \times 10^{-8}$ to $4.0 \times 10^{-8}$ m$^3$, was therefore very low. Although other zooplankton species can flourish when water temperatures rise in summer, their abundances have been reported to be relatively low compared with that of copepods (NFRDI, 2007). Salps are predictors of phytoplankton, and their abundance increases rapidly during blooms, sometimes exceeding that of copepods. However, predator abundance in initial bloom stages is lower than their average abundance, following the classic prey–predator population curve (Kang et al., 2004). Therefore, it was assumed that the effects of other scattering sources were negligible. Nevertheless, to minimize the possibility of overestimation as a consequence of contamination from signals scattered from other sources, deviant signals were rejected based on Chauvenet’s criterion.

The results presented here imply that the acoustic method may be a useful tool for detecting C. polykrikoides blooms in their initial stages. However, further study, including investigations of the density and sound speed of bloom-causing algal species and the abundance of other scattering sources, is needed to understand more fully the acoustic-scattering behaviour of HABs.

Acknowledgements

The study was supported by a “Development of ubiquitous management technologies for marine useful/harmful organisms” grant (No. PE98474), promoted by the Korea Ocean Research and Development Institute.

References


doi:10.1093/icesjms/fsq093