

Analysis of Trace Iron (Fe) in Sea Water by Using Inductively Coupled Plasma Mass Spectrometry with Solid Phase Chelate Extraction Technique

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Abstract

Field experiments on recovering from barren sea ground called "Isoyake" by providing Fe in sea water to restore seaweed beds on barren ground, has been carried out. Fe supply source, a mixture of steelmaking slag and compost (humic wood chips), were laid underground in shoreline. A certain amount of restoration of seaweed beds has been confirmed at the locations for several years. Quantitative determination of trace amount of Fe in sea water is necessary to validate the effect of iron source. However, to analyze dissolved Fe in sea water was difficult due to its trace concentration and the interference by high concentration of salts in the analytical samples. We established new analysis method of Fe in sea water, by treating sea water with solid phase chelate extraction and analyzed by inductively coupled plasma mass spectrometry with cool plasma condition. This method was successfully applied for the sea water sample taken from experimental site. Other water quality parameters in the experimental site were also investigated. Fe concentration was significantly highest around the Fe supply source and was decreased with the distance. Although the concentrations of dissolved silica, Mg and Ca were correlated to electrical conductivity of the sample water, Fe concentration was not. According to those results, the concentration of dissolved Fe from steelmaking slag was confirmed, and Fe might be contiguously supplied to the sea water from Fe supply source and promoted the growth of seaweeds. Therefore, applying a mixture of steelmaking slag and compost as Fe supply source is an effective technique to supply dissolved iron to Isoyake area.

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1. Introduction

In Japanese coastal areas, the marine whitening phenomena known as “isoyake,” or sea desertification, is a problem. When it occurs, beneficial algae fail to come into being or grow, eventually causing the seabed to be covered with calcareous algae. Isoyake has brought about a serious problem through decreased marine resources, such as seaweed and fish, for coastal fisheries.¹⁾ Isoyake is thought to be caused by a combination of factors, including a rise in seawater temperature and the resultant change in ocean currents, and damage to seaweed caused by plant-eating animals, etc. Therefore, it is considered that the cause of isoyake differs from region to region according to the topography and ocean environment, including the ecosystems and suchlike of each individual locality. With the aim of improving the above conditions, various measures to restore lost algae grounds have been studied.²⁻⁴⁾

A change in nutrients in marine areas, specifically a decline in the concentration of Fe, which is a nutrient indispensable for the growth of algae,⁵⁾ has been reported as another major factor in the occurrence and progress of isoyake.^{6,7)} Therefore, attempts have been made to create new algae grounds in marine areas by supplying Fe as required.^{8,9)} When Fe is supplied to a marine area, it is promptly oxidized by the oxygen dissolved in the seawater and is precipitated in the form of a hydroxide. At this stage, its usefulness to marine life is considered to decline markedly.¹⁰⁾ The challenge then is to supply dissolved Fe in a stable state. As a measure to meet the challenge, we developed and applied fertilizers as the sources of Fe for some 20 marine areas throughout the country that suffer from isoyake caused by Fe deficiency in seawater. The fertilizer is a mixture of Fe-containing steelmaking slag and compost that produces dissolved Fe in the form of humic complexes in stable states. Our efforts to improve the environment of marine areas are producing tangible results.^{11,12)}

For example, on the Shaguma Coast, Mashike-cho, Hokkaido, one of the experimental marine areas, in autumn 2004 we buried our fertilizer along the shoreline of an actual marine area where isoyake had progressed noticeably. Since then, the algae grounds have been reviving and expanding over the area, mainly along the shoreline.^{13,14)} As the Vivary® Series including the Steel Slag Hydrated Matrix (SSHM) block, the fertilizer mentioned above has been approved and registered under the application technology certification system established by the Japan Fisheries Science and Technology Association for materials and equipment for the development and restoration of fishing grounds. In addition, the safety of the fertilizer has been certified under the steelmaking slag product safety confirmation & certification system established by the National Federation of Fisheries Cooperative Associations. Thus, public recognition of our fertilizer has been rising.

It is estimated that Fe eluted from steelmaking slag dissolves in stable states by complexing with humic matters, thereby helping to promote the growth of algae and microorganisms. In order to verify the above estimation and clarify the duration of the effect of Fe and the biological mechanism of Fe, it is essential to measure the concentration of Fe dissolved in the actual marine area. However, the concentration of Fe in coastal seawater is estimated to be only in the order of several $\mu\text{g/L}$, even though it is higher than that in ocean water. As a result, and due to the fact that the salt concentration of seawater is high, it is difficult to measure the concentration of Fe in seawater using conventional methods. For this reason, the effect of the Fe concentration in experimental marine areas on the recovery of algae grounds therein is not clearly understood.¹²⁾ In the present study, therefore, we established a method for trace analysis of Fe separated

from the matrix of seawater and investigated the water qualities of the actual marine area along the Shaguma Coast, Mashike Town, Hokkaido. On the basis of the investigation results, we discuss the effect of applying our fertilizers.^{15,16)}

2. Method of Analyzing Trace Amount of Fe in Seawater

2.1 Separation of Fe from seawater matrix

When a seawater sample is directly admitted into inductively coupled plasma mass spectrometer (ICP-MS), atomic absorption photometer or some other analyzer, a considerable proportion of the salt contained in the sample often precipitates in the analyzer and interferes with the analysis. Also, 1, 10-phenanthroline absorption photometry, which has long been used for Fe analysis, is insufficiently sensitive for the purposes of our study. Therefore, we considered it necessary to selectively extract Fe—our target substance—from the seawater. We decided first to separate Fe from the matrix of seawater and then to carry out a high-sensitivity analysis of the Fe using ICP-MS.

In order to separate out Fe, we adopted the solid-phase chelate extraction method. As the chelate, Nobias CHELATE-PA1 (Hitachi High-Technologies Corporation) that selectively adsorbs Fe and other metallic elements was selected.¹⁷⁾ As shown in Fig. 1, the chelate is composed of a resin base on which ethylenediaminetriacetic and iminodiacetic groups are modified. It selectively adsorbs transition metals, alkaline earth metals and alkali metals in that order. We discussed conditions for separating Fe from artificial seawater by using the above solid phase chelate and to analyze Fe.

The artificial seawater used to discuss conditions for the Fe analysis was ultrapure water (Millipore, specific resistance of 18.3 M Ω) with the Aquamarine reagent (Yashima Pure Chemicals Co., Ltd.) added. As the nitric acid for elution and cleaning, TAMAPURE 100 (Tama Chemicals Co., Ltd.) was used. The other reagents used were special-grade reagents (Wako Pure Chemical Industries, Ltd and Kanto Chemicals Co., Ltd.). These reagents were not subjected to any special treatment such as refining.

2.2 Influence of pH on Fe recovery ratio

Iron (Fe) was added to both the ultrapure water and artificial seawater until the Fe concentration became 0.1 mg/L. Then, the ratio of Fe recovery from the pure water and seawater, respectively, was measured. The measurement results are shown in Fig. 2. The Fe recovery ratio (i.e., the degree to which the added Fe is absorbed to the solid phase chelate) was measured by varying the pH value of each

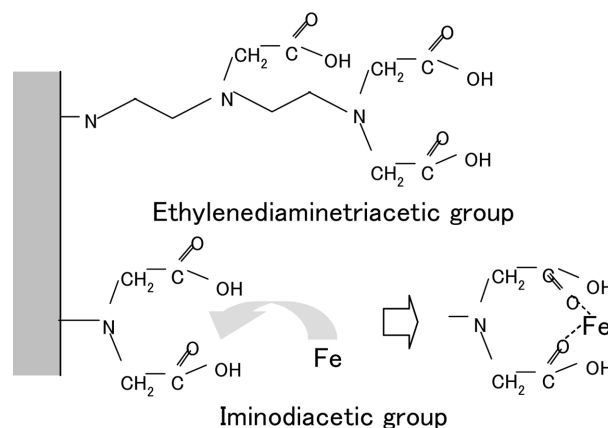


Fig. 1 Chemical structure of chelate in this study

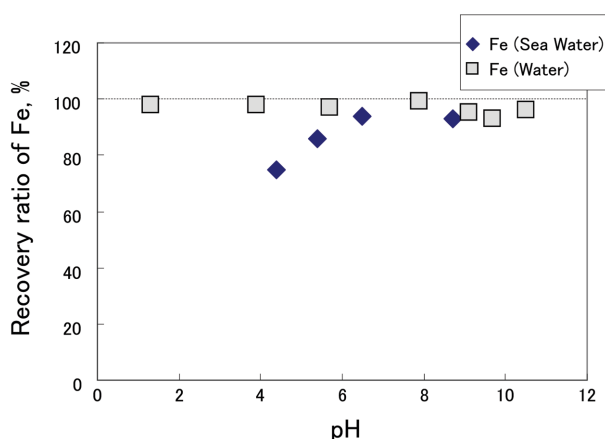


Fig. 2 Effect of pH on recovery ratio of Fe in sea water and ultrapure water

sample. In the case of the ultrapure water with Fe added, the Fe recovery ratio was nearly 100% over a wide pH range, from acid to alkali. With the artificial seawater with Fe added, it was confirmed that the Fe recovery ratio shows a tendency to drop as the pH value of the sample decreases. The drop in the Fe recovery ratio when the sample is acidic and has a high salt concentration is considered ascribable to the decline in chelate stability owing to the difference between the ion intensity and the pH of the chelate used in the present study. Since the pH value of the experimental marine area is around 8, it does not seem to pose any problem in the analysis of natural seawater, except in an unusual environment such as when acid wastewater with a high salt concentration flows into the marine area.

2.3 Influence of EDTA concentration on Fe recovery ratio

Fig. 3 shows the measurement results for the Fe recovery ratio when EDTA (ethylenediaminetetraacetic acid) was gradually added to ultrapure water whose Fe concentration was 0.1 mg/L. It is already understood that when EDTA is added to Fe-containing solution, the Fe forms Fe-EDTA complex.

When the amount of EDTA addition reached a certain level, the Fe in the sample could no longer be extracted by the chelate used in the present study. The reason for this is thought to be as follows. Since the chelate used in the present study has ligands similar to that of EDTA, its ability to extract Fe is almost the same as the ability of EDTA to form a complex. As a result, the Fe-EDTA complex is stable, preventing the Fe from being extracted.

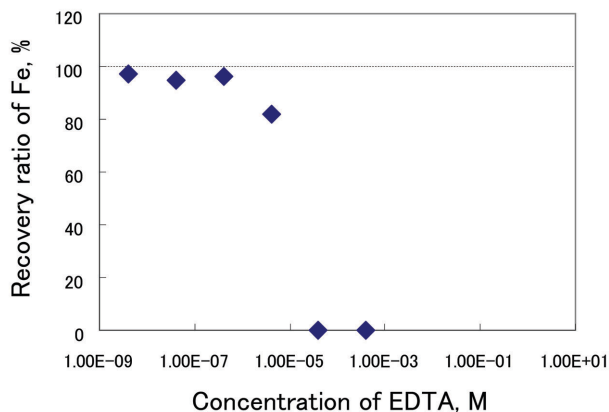


Fig. 3 Effect of EDTA on recovery ratio of Fe

In addition to EDTA, various substances capable of forming complexes were studied. As a result, it was confirmed that surface active agents contained in detergents did not interfere with the extraction of Fe even when added in substantial quantities. However, those substances that form strong complexes with Fe (e.g., tannic acid) reduced the Fe recovery ratio to nearly zero depending on the quantity added. The implication is that with seawater samples in which huge volumes of domestic wastewater flow into the marine area, the analysis results might be lower than the true values. Therefore, it is considered necessary first to measure the contents of complexing substances by gas chromatography or liquid chromatography, and then to decompose the complexes by acid treatment, etc.

Considering the possible existence of humic substances, we also measured the Fe recovery ratio when the sample had fulvic acid (standard reagent of the International Humic Substances Society) added. The result was that the Fe recovery ratio far exceeded the amount of addition. The reason for this is thought to be that the fulvic acid reagent added contains soluble Fe, which was extracted by the chelate. Thus, we estimated that it would be possible to extract the fulvic acid-Fe complex.

2.4 Elution profile

After Fe is extracted by the solid phase chelate, the Fe adsorbed to the chelate is eluted via a suitable eluting solution. We studied the relationship between the amount of eluting solution and the extraction (recovery) ratio of Fe in that process. When the amount of eluting solution is large, it adversely affects the analysis because of the reagent blank and increases the chance of sample contamination. Therefore, it is important to confirm the relationship between the recovery ratio of Fe and the amount of eluting solution.

The sample used was the ultrapure water with ferric nitrate solution added until the Fe concentration became 0.1 mg/L. The pH value of the sample was adjusted to about 7 by adding ammonium acetate. The eluting solution used was 3M high-purity nitric acid (High-purity reagent diluted with the ultrapure water). Fig. 4 shows the relationship between the amount of eluting solution and the extraction ratio of Fe. Since the Fe recovery ratio remains almost the same when the amount of eluting solution is 4 ml or more, we considered that for our experimental system, 4 ml was the optimum amount of eluting solution.

2.5 Analytical procedure and quantitiveness

On the basis of the above results, we measured the concentration of Fe in actual seawater samples. The measurement procedure is schematically shown in Fig. 5. First, the chelate is activated, cleaned in

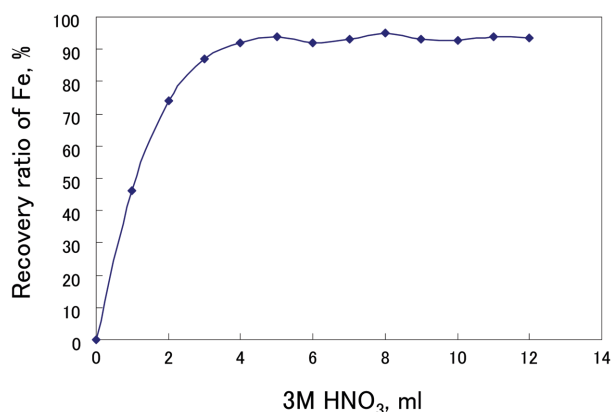


Fig. 4 Elution profile of Fe extracted by solid phase chelate

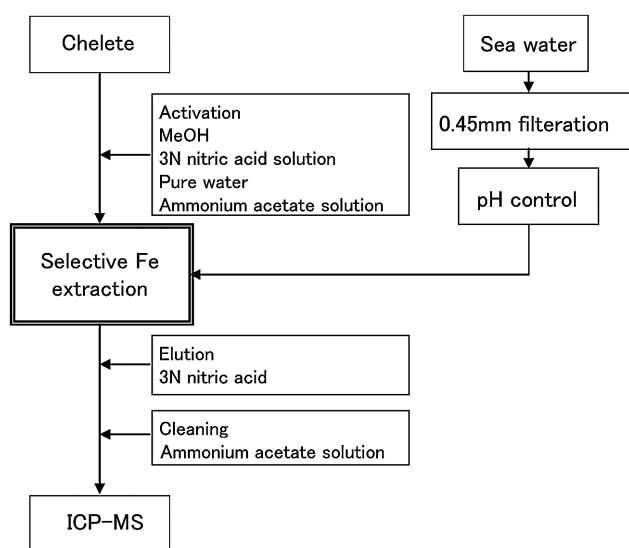


Fig. 5 Analytical procedure

nitric acid solution, and treated with ammonium acetate. Then, a seawater sample, which is passed through a 0.45 μm pore filter and whose pH is adjusted to about 7 by ammonium acetate, is subjected to selective Fe extraction by the solid phase chelate. The extracted Fe in the sample is eluted in nitric acid solution and the Fe concentration of the eluting solution is measured using ICP-MS to determine the Fe concentration of natural seawater.

ICP-MS is a technique for measuring the mass and quantity of elements. The principle of this technique is as follows. The sample solution is turned into mist by a nebulizer and let into a high-temperature, inductively coupled plasma to excite and ionize the elements contained in the solution. The ions of each element obtained are let into a vacuum via sampling and skimmer cones, and focused on the mass spectrometer through an ion lens. Among the isotopes of Fe, ^{56}Fe is the most abundant and should be measurable with a high degree of sensitivity. However, when ^{40}Ar and ^{16}O combine to form interfering ions, they have the same mass as ^{56}Fe and interfere with the measurement of Fe. Therefore, with the ICP-MS system equipped with a quadrupole mass spectrometer, it is possible to measure only low concentrations on the order of several $\mu\text{g/L}$.

There are several methods of coping with the above problem. Examples: (1) Use a high-resolution ICP-MS equipped with a high resolution mass spectrometer, or (2) Using collision cell or reaction cell technologies to destroy the interfering ions.¹⁷⁾ In the present study, we adopted (3) cold plasma technique that uses a low-temperature plasma to restrain the formation of interfering ions. Specifically, the high-frequency output that is ordinarily in the range 1.0 to 1.2 kW was set at about 0.6 kW. Compared with Methods (1) or (2), Method (3) that we adopted has some drawbacks. For example, our method, which uses cold plasma, is inferior in terms of sensitivity and is susceptible to interference from acid matrices contained in the analytical solution. However, we considered that our method would not pose any special problem in view of the fact that even though the sensitivity is not very high, our method can measure Fe in the order of several $\mu\text{g/L}$ and the matrix of the analytical solution subject to chelate separation and acid elution is the same regardless of the sample used. As ICP-MS systems, we used an SPQ-9000 and an SPQ-9500 made by SII NanoTechnology Inc.

The calibration curve used to determine the Fe in seawater samples

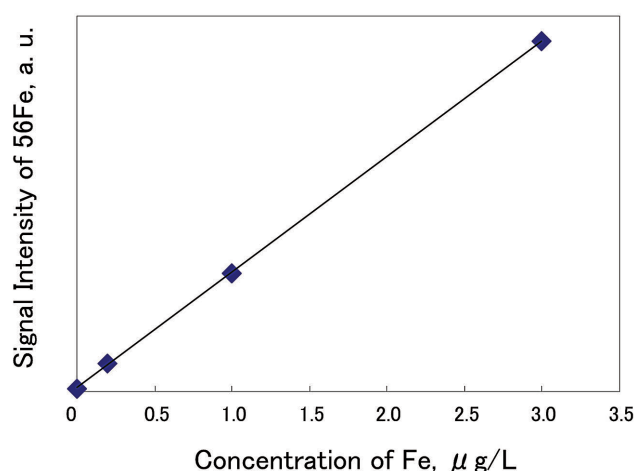


Fig. 6 Calibration curve for Fe determination

Table 1 Analytical results of Fe in nearshore seawater reference material for trace metals

	($\mu\text{g/L}$)	
	Certified value	Measurement result
CASS-04	0.713 ± 0.058	$0.6_9 \pm 0.0_1$
CASS-05	1.44 ± 0.11	$1.4_3 \pm 0.0_1$

is shown in Fig. 6. With the correlation coefficient and blank equivalent concentration being 0.999 and 0.02 $\mu\text{g/L}$, respectively, it was confirmed that the calibration curve had good linearity, suggesting that it could be used to determine Fe concentrations of seawater samples in the order of several $\mu\text{g/L}$. Using the calibration curve, we analyzed Canada's seawater certified reference materials CASS-4 and CASS-5. The results obtained agreed well with the certified values as shown in Table 1.

3. Application to Actual Marine Areas

3.1 Outline of experiment on application of slag fertilizer in marine area

Our analytical method was used in an experiment on the application of slag fertilizer to a marine area conducted along the Shaguma, Mashike-cho, Hokkaido. Fig. 7 schematically shows the experimental zones. Two experimental zones (A, B) and a control zone (E) were installed at intervals of about 100 m along the shoreline. In each of the experimental zones, the fertilizer whose effect was to be verified was buried. About 200 kg of the fertilizer was put in each bag made of coconut fiber and a total of 39 bags (3×13), or about 8 tons, was buried in each experimental zone.

The fertilizer for Experimental Zone A was a 1:1 mixture of humic matter made from used wood chips and steelmaking slag, and the fertilizer for Experimental Zone B was steelmaking slag. In 2004, in each experimental zone, the bags of fertilizer were buried in a ditch measuring 1 m in width, 26 m in length and 0.8 m in depth. In June of 2007, about three years after the fertilizer was buried, we investigated the water qualities of the experimental marine area. At that time, seaweed was flourishing all over the experimental marine area, indicating that the algae were being restored.¹³⁾

3.2 Experimental results and discussions

Samples of clear seawater at points 3 m, 5 m, 10 m, 25 m and 50 m offshore from the coastline of Experimental Zone A, the reference zone, Experimental Zone B, and the midpoint between the two ex-

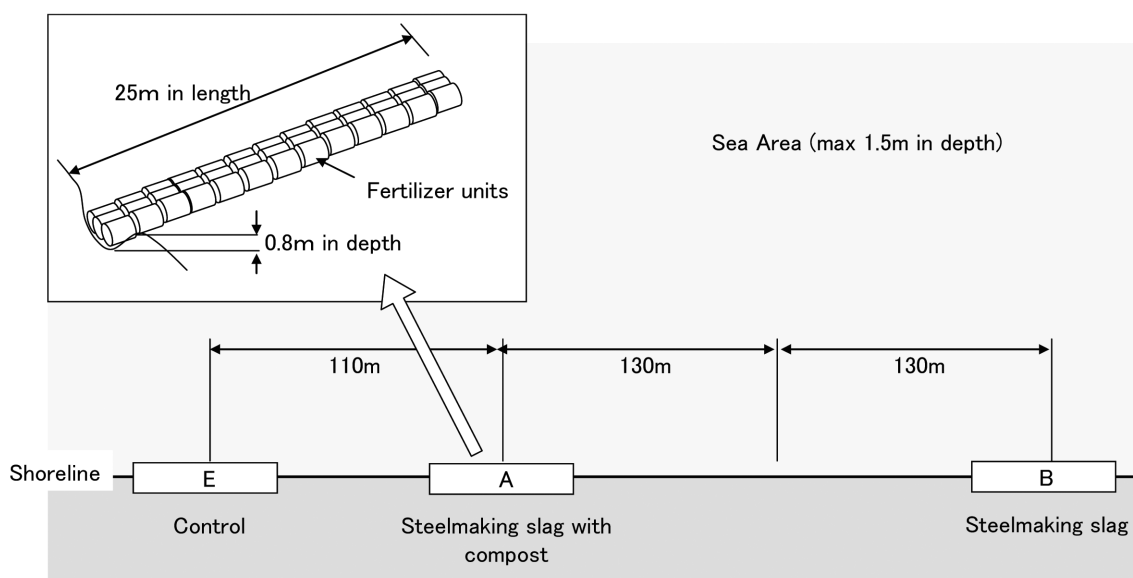


Fig. 7 Schematic of experimental site

perimental zones (a total 20 sampling points) were collected in polyethylene bottles from water depths between 50 cm and 100 cm. The collected seawater samples were speedily transported in a cold-storage to the laboratory for analysis. The water qualities examined were, in addition to the Fe concentration measured by the method described in the preceding section, the pH value, electrical conductivity (EC), total nitrogen (T-N), total phosphorus (T-P), Ca and Mg measured using the method specified in JIS K 0101-1998,¹⁸⁾ and dissolved silica (D-Si) specified in JIS K 0102-1998.¹⁹⁾

The analytical results of the water qualities of the samples collected from the 20 points are shown in **Table 2**. EC was 44.2 ± 2.8 mS/cm on average, with the minimum and maximum values being 39.0 and 48.5, respectively, suggesting the influence of inflows of freshwater, such as river water and spring water, into the experimental marine area, while D-Si was 0.70 mg/L on average. However, the maximum value of D-Si was about five times larger than the minimum value. If freshwater was the principal supply source for D-Si, there is the possibility that D-Si was influenced by freshwater as in the case of EC. On the other hand, Fe was $5.9 \mu\text{g/L}$ on average, showing substantial difference between the minimum and maximum values. The Fe recovery ratio obtained by the method used in the present study is not influenced by freshwater. Therefore, there is the possibility that the concentration distribution of dissolved Fe cannot

always be explained by the influence of an inflow of freshwater alone.

Fig. 8 shows the horizontal distribution of each of the water quality items examined at the seawater sampling points. In the figure, the X-axis represents the shoreline along which the fertilizer was buried while the Y-axis represents the distance from the shoreline, and the analysis results for the individual sampling points have been complemented by the Bi-Liner method. As shown in Fig. 8 (a), a concentration distribution of Fe with $18.1 \mu\text{g/L}$ as its peak was observed in the neighborhood of Experimental Zone A in which the fertilizer composed of humic matter and steelmaking slag had been buried. In the neighborhood of Experimental Zone B, too, in which the fertilizer composed of steelmaking slag alone had been buried, a concentration distribution of Fe having a peak of $10 \mu\text{g/L}$ and a concentration gradient offshore was observed. We consider that both distributions suggest the possibility that Fe was supplied from the fertilizer. It should be noted, however, that a region in which the Fe concentration was relatively high (5 to $10 \mu\text{g/L}$) was observed in the offshore direction from Control Zone E.

The pH value in Fig. 8 (b) showed a tendency to decrease from the shoreline toward the offing. It was somewhat high in Experimental Zone B and somewhat low in Experimental Zone A. Concerning EC in Fig. 8 (c) and D-Si in Fig. 8 (d), they showed similar concentration distributions having a gradient from the bottom left towards the top right. The implication is that D-Si was supplied to the marine area by inflows of freshwater and that EC decreased as the seawater was diluted by freshwater. With respect to Ca (Fig. 8 (e)) and Mg (Fig. 8 (f)), which are generally contained in extremely small amounts in freshwater and which come from seawater, a low concentration region was observed from Control Zone E toward the offing, attesting to the dilution of seawater by inflows of freshwater.

As to nutrient salts, such as T-N, P and D-Si, it is estimated that they will be supplied to the marine area by freshwater during the snow-thawing season and at times when the water temperature rises. We considered that as in the case of those nutrient salts, freshwater might be a source of Fe supply to coastal marine areas. So, with the aim of measuring the influence of freshwater inflows on the Fe distribution observed in the experimental marine area, we studied the

Table 2 Analytical results of sea water taken from experimental site

		Measurement results	
		Average	min - max
pH		8.27 ± 0.08	8.14 - 8.39
EC	mS/cm	44.2 ± 2.8	39.0 - 48.5
T-N	mg/L	0.13 ± 0.02	0.10 - 0.18
T-P	mg/L	0.016 ± 0.009	< 0.01 - 0.04
D-Si	mg/L	0.70 ± 0.28	0.27 - 1.24
Ca	mg/L	445 ± 37	379 - 505
Mg	mg/L	$1,380 \pm 117$	1,157 - 1,558
Fe	$\mu\text{g/L}$	5.9 ± 4.5	< 1 - 18.1

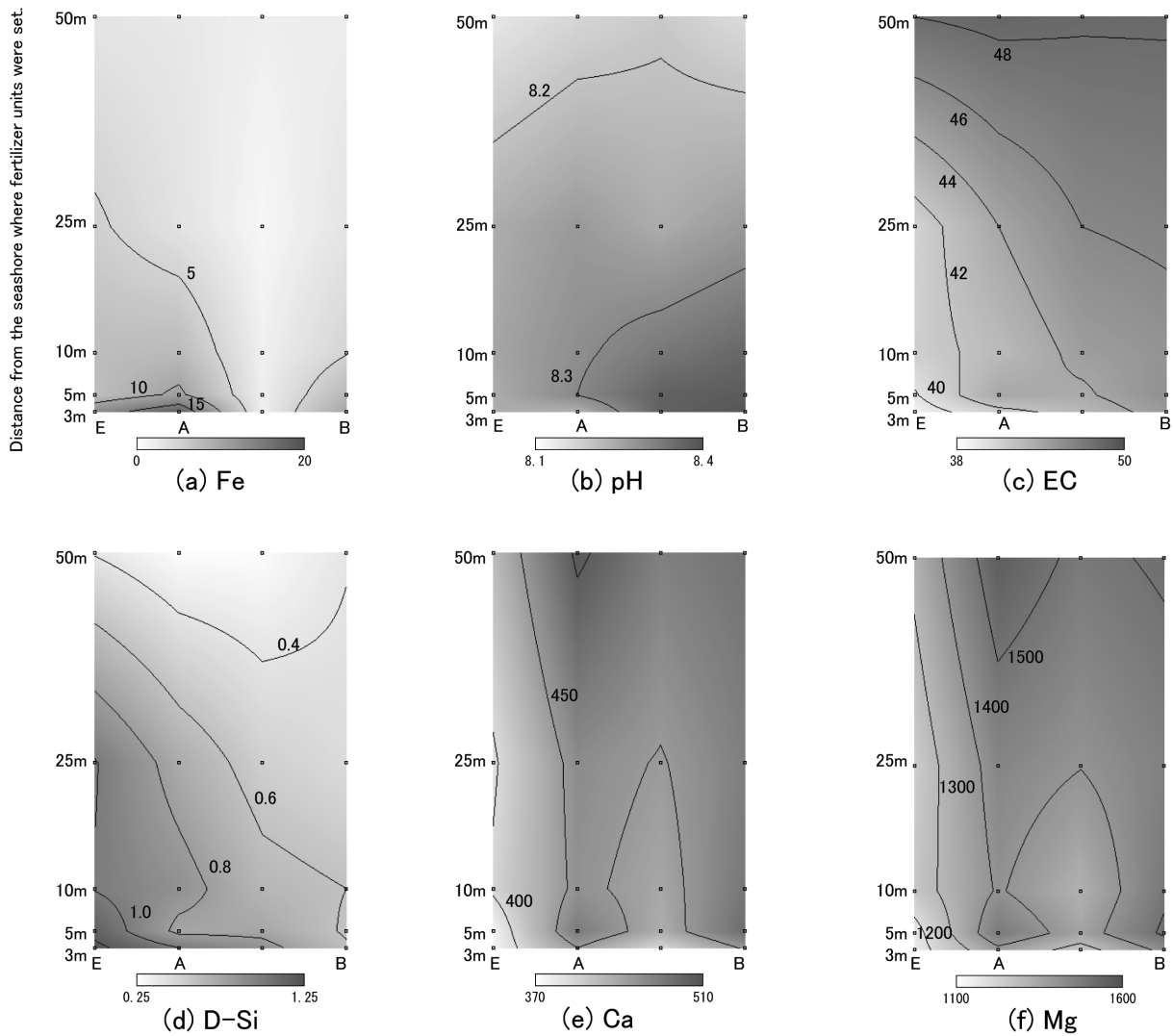


Fig. 8 Horizontal distribution of each parameter at the experimental site

correlations of the water quality parameters. As a result, a definitely negative correlation (correlation coefficient $R = 0.99$) was observed between EC and D-Si. This suggests the possibility that D-Si came from freshwater. Calcium (Ca) and Mg, respectively, show a positive correlation with EC ($R = 0.79$). In addition, there was a definite correlation ($R = 0.99$) between Ca and Mg. Based on these findings, it is estimated that Ca and Mg, which had always existed in the seawater, were diluted by inflows of freshwater.

On the other hand, Fe has only weak correlations with the other parameters— $R = 0.6$ with EC and D-Si, $R = 0.5$ with T-N, $R = 0.4$ with T-P, $R = 0.2$ with Ca and Mg. Thus, the Fe concentration distribution in the experimental marine area cannot be explained by the influence of freshwater inflows. In view of the fact that the concentration of Fe was highest in Experimental Zones A and B in which the fertilizer had been buried, it is reasonable to think that the Fe came from the fertilizers.

4. Conclusion

With the aim of determining the quantities of Fe contained in seawater, we studied a highly sensitive Fe analysis method in which

Fe in seawater is extracted by solid phase chelate. It was possible to analyze the Fe dissolved in seawater using this method, although there is the possibility that the analysis may be interfered with by specific elements (these elements can be identified with suitable methods). Using the solid phase chelate, we also analyzed seawater certified reference materials. The analysis results obtained agreed well with the certified values.

In an experimental marine area, Mashike-cho, Hokkaido, we buried fertilizers composed of humic matter and steelmaking slag along the shoreline with the aim of studying the recovery mechanism for algae grounds and restoring the algae grounds in the coastal marine area where “*isoyake*” was progressing. Recently, we analyzed the water qualities there. As a result, in the experimental marine area where the influence of inflows of freshwater was estimated from the distribution of EC, Fe did not show any significant correlation with other water quality parameters, such as EC, D-Si, Ca and Mg, suggesting that it did not come from freshwater that had flown into the marine area. In addition, it was found that the Fe concentration in the experimental marine area was highest in the neighborhood of the points where the fertilizer had been buried and that the Fe concentra-

tion decreased toward the offing. Those findings strongly suggest that in these experimental marine areas, Fe dissolved from the fertilizer had spread over a wide area. Thus, we estimated that the dissolved Fe does contribute to the reproduction of seaweed.

In present study, we were able to measure the concentration of Fe in a marine area and confirm the possibility that fertilizers composed of steelmaking slag should be able to supply dissolved Fe to the marine area. In order to further demonstrate that our fertilizer (product name: Vivary® Series) is effective in improving the environment of marine areas and promote the utilization of steelmaking slag, we intend to continue monitoring the experimental marine areas, including the dissolved Fe, and implementing biological and environmental impact assessments.

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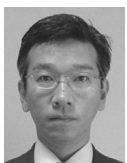
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