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RESEARCH

Selecting indicator-diatom taxa and its application for simplifying trophic diatom index for biological water quality assessment

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Background: Trophic diatom index (TDI) should have been conducted annually by 'water environment conservation act' since 2008 in South Korea. However, its application had difficulties in identification such as fine structures and diversity of diatoms. In this study, it is proposed that a new water quality assessment method called simplified-TDI (S-TDI) for reducing the difficulties of identification using indicator-diatom taxa.
Results: The indicator-diatom taxa were selected using relative abundance and frequency of occurrence. The one criterion was above 10% of the minimum relative abundance of diatom species occurred in each station and the other was above 2% of the minimum frequency of occurrence of diatoms among the total station. Appeared species in 1,999 stations for study was 412 and 115 species were selected as the indicator-diatom taxa. The criterions were verified using statistic assessment method as below similarity, paired sample t-test and root mean square error (RMSE). Species excluded from indicator-diatom taxa were identified two methods. They were identified up to genus level and received the average sensitivities and indicator values of the genus in TDI. Even they were not able to be identified genus level, identified them to "unidentified group" and received the average sensitivities and indicator values of total diatom species in TDI. This method was newly proposed as S-TDI in this study. To verify the application of S-TDI, its values were compared with original TDI values from 60 sampling stations considering various aquatic environments. The similarity between S-TDI and original TDI values were very high degree of 0.999, the paired sample t-test indicated no statistically significant difference and RMSE was 1.52 below the threshold.
Conclusions: These results showed S-TDI can be applied to the water quality assessment instead of original TDI.

Keywords: diatom, indicator-diatom taxa, trophic diatom index, water quality assessment

Introduction

Diatoms lived in different water environmental conditions and well divided tolerances and preferences for various environmental factors (Dixit et al. 1992; Smol and Stoermer 2010). Diatoms were changed in environment conditions in water system and reflected changes of water environment for the time of growth (Lee 1997). For this reason, water quality assessment using diatom should been carried out annually in South Korea by 'Water Environment Conservation Act'. in addition, the diatom must be investigated in water quality assessment by Water Environment Conservation Act of Article 8-2.

However, one of the difficulties in water quality assessment using diatoms is the large number of species (Berthon 2011). VanLandingham (1967) published an 8-volume books that compiled over 44,000 diatom species including their synonyms. AlgaeBase (2021), an algal search website, had registered 14,442 diatom species in February 2023. In South Korea, National Institute of Biological Resources (2023) under the Ministry of Environment (MOE) published the National Species List of Korea reported 2,243 diatom species. The other difficulty is the fine structures used in identification of diatoms. They had fine structures such as striae and raphe, and these were important characteristics for taxa of diatoms in identification. For these reasons, identification of diatoms was required researchers skilled in taxonomy (Bueno et al. 2017; Sánchez et al. 2019).

Various studies were conducted to solve the difficulties of water quality assessment using diatoms. Lavoie et al. (2009) proposed a simplified version of the Indice Diatomées de l'Est du Canada (IDEC) diatom-based water quality index by reducing the taxonomic resolution for identification. They used the relative abundance of diatom taxa to simplify the list of diatom species used in the original IDEC and used this simplified list for improving IDEC (Lavoie et al. 2014). Berthon et al. (2011) proposed a simplified water quality assessment method using the life-forms of diatoms of rivers in southeastern France. However, they didn't suggest how to deal with species which had the same life-form, while they had different pollution sensitivities.

In South Korea, although water quality assessment was carried out using trophic diatom index (TDI; Kelly and Whitton 1995), study about simplifying the TDI was not common.

The purpose of this study is improving the TDI by reducing the difficulties in identification of diatoms using indicator-diatom taxa selected based on relative abundance and frequency of occurrence, and verifying whether the TDI using simplified method can be applied to actual water quality assessment.

Material and Methods

Sampling stations and data collection

Sampling stations for selecting the indicator-diatom taxa were the biomonitoring stations of the Nakdong River basin provided by the 'Water Environment Information System (2021) (https://water.nier.go.kr/)' in the National Institute of Environmental Research (NIER) of the MOE for the years 2019 to 2021. Total sampling stations were 1,014 including 300 stations in 2019, 357 stations in 2020 and 357 stations in 2021. Each of stations was investigated two times per year and, therefore, the data were collected 2,028. Among these, 1,999 datasets were used excluding the data which were ambiguous in identification, or no data due to drying stream or inaccessible area.

In addition, 60 sampling stations were directly investigated considering the TDI value to reflect the various ecological conditions of rivers and streams. These were conducted to investigated whether the 'simplified trophic diatom index (S-TDI)' proposed simplified method in this study can be replaced the TDI in actual field water quality assessment (Fig. 1).

Diatom sampling method and calculation of the TDI

The diatom samples were collected by scraping a surface

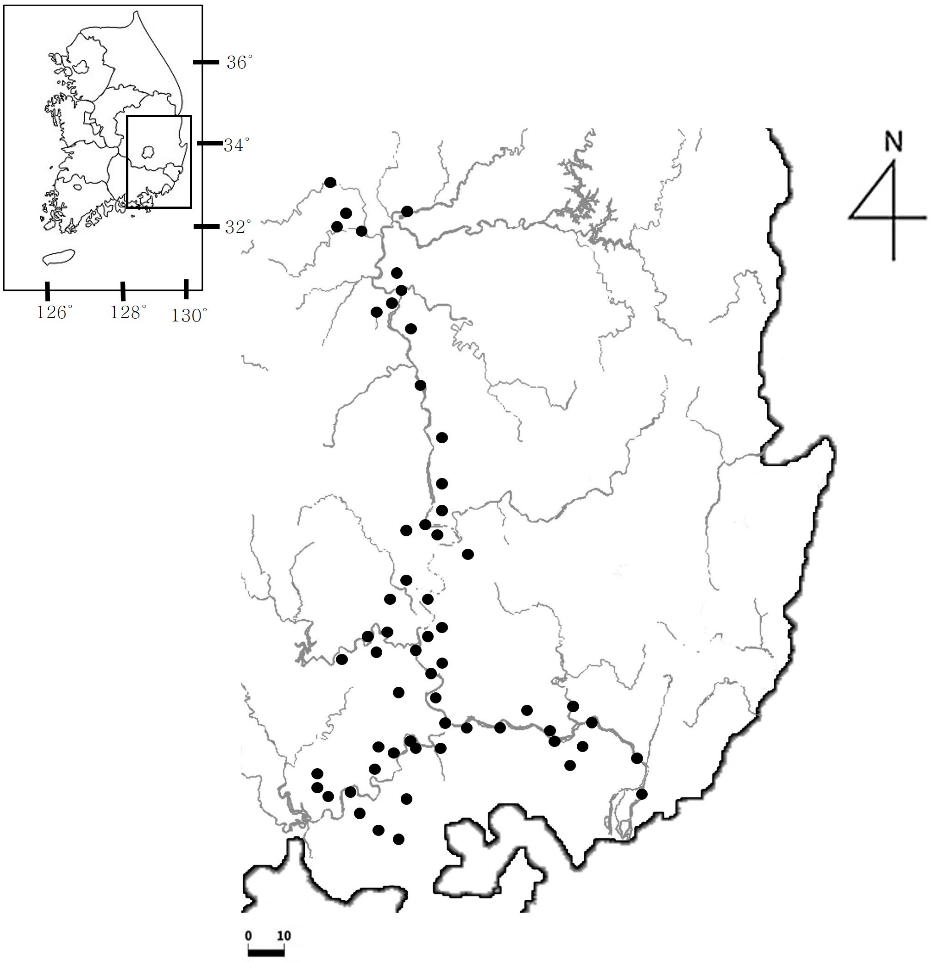

Fig. 1 The 60 sampling stations selected to investigate the possibility for application of TDI newly proposed in this study from the Nakdong River (Two sites were surveyed in both 2021 and 2022, a total of 58 sites are shown on the map). TDI: trophic diatom index.

area parallel to the water surface with a brush from a stone located at a depth of 20 to 30 cm. Permanent diatom slides were prepared by cleaning the samples using the $KMnO_4$ method (Hendey 1974) and mounting them with Pleurax. identification and counting for calculating the relative abundance of the diatoms was used a light microscope (Axioscope 2.0; ZEISS, Jena, Germany) and randomly selected microscopic views were observed through 1,000x magnification.

TDI was calculated using the relative abundance (%) of appeared species obtained from microscopic counting, and the assigned pollution sensitivity and indicator values of each appeared species. The method for calculating the TDI value and the grading criteria for indicating the environmental state follow the MOE of South Korea, as shown in Table 1 (NIER 2019).

Selection of the indicator-diatom taxa

The indicator-diatom taxa were selected to simplify the TDI used for water quality assessment in South Korea. The relative abundance and frequency of occurrence of diatoms observed in the Nakdong River basins were used for the selection. The frequency of occurrence was calculated as a percentage of sites where each species was found out of all the sampling stations.

The indicator-diatom taxa were selected by two-steps process. The first step was to select indicator-diatom taxa based on the relative abundance of each species observed sampling stations. Following Lavoie et al. (2009), ten cutoff levels for selecting the indicator-diatom taxa based on relative abundance were set at 2% intervals from 2% to 20%. Species with a maximum relative abundance above the cutoff level at least once in the sampling stations were selected as the indicator-diatom taxa provisionally. The second step was to select species which selected in first step based on frequency of occurrence. Ten frequency of occurrence cutoff levels were set at 1% intervals from 1% to 10%. Species with a maximum frequency of occurrence above the cutoff level at least once in the sampling stations were finally selected as the indicator-diatom taxa (Fig. 2).

Table 1 The grades and environmental status criteria of the TDIm (NIER 2019)

Grade	Environmental status	Trophic diatom index (TDI)
A	Very good	90 ≤ – ≤ 100
B	Good	70 ≤ – < 90
C	Fair	50 ≤ – < 70
D	Poor	30 ≤ – < 50
E	Very poor	0 ≤ – < 30

$TDI = 100 - \{(WMS \times 25) - 25\}$.
WMS: weighted mean sensitivity.
$WMS = \sum_{i=1}^{n} Ai \cdot Si \cdot Vi / \sum_{i=1}^{n} Ai \cdot Vi$
n: number of total species; *Ai*: abundance of species *i* in sample; *Si*: pollution sensitivity (1–5) of species *I*; *Vi*: indicator value (1–3) of species *I*.
TDIm: modified trophic diatom index.

Statistical analyses

Similarity analysis, paired sample t-test, and scatter plot were performed using IBM SPSS Statistics 20 to select indicator-diatom taxa and to compare TDI and S-TDI. The R program (Ver. 4.2.2) was used to calculate the root mean square error (RMSE) for error comparison.

Terminology

TDI and modified trophic diatom index (TDIm)

TDI originally refers to a water quality assessment method using diatoms proposed by Kelly and Whitton (1995). In South Korea, TDI was partially modified the pollution sensitivities and indicator values of diatom species appropriately for the South Korean rivers and streams, and it called TDIm (Kim 2012; NIER 2019). In this study, TDI refers to TDIm.

S-TDI

S-TDI is a newly proposed water quality assessment method in this study using "the indicator-diatom taxa" for simplifying TDI.

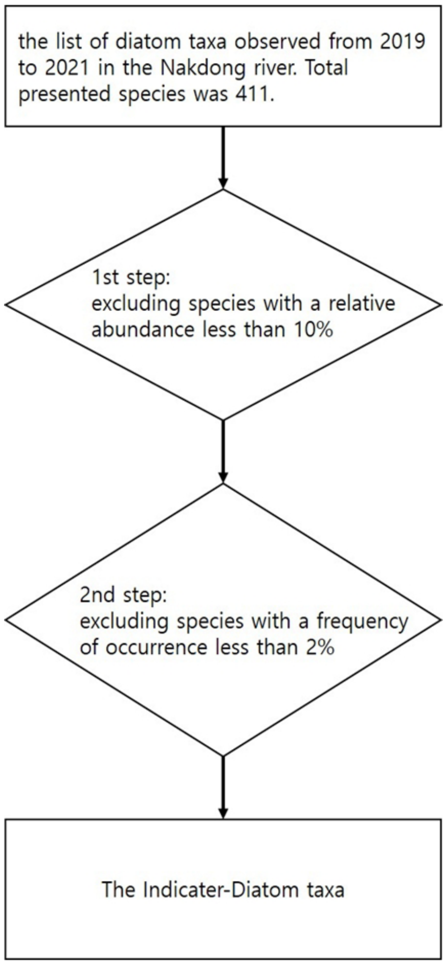

Fig. 2 Over view of selecting process of the indicator-diatom taxa.

Indicator-diatom taxa

Indicator-diatom taxa is a group of 115 major diatom species selected based on the relative abundance and frequency of occurrence. These species were selected to reduce the difficulties in identification for calculating the TDI.

TDI is a method of water quality assessment based on the pollution sensitivities and indicator values of observed all species. However, identifying all epilithic diatom species is very difficult because they have fine structures and a large number of species.

S-TDI is calculated using only indicator-diatom taxa, instead of all the diatom species present at the sampling site. Diatom species observed at the sampling site but not included in the indicator-diatom taxa were evaluated using a separate simple method, which will be described later.

Results

The 1st step for selection of the indicator-diatom taxa

A Pearson correlation analysis was conducted to compare the similarity between the groups of the TDI recalculated using species selected according to the cutoff levels based on relative abundance and the TDI calculated using the original method. Similarity ranged from 0.984 to 0.999, with a significance level of $p < 0.01$. The similarity showed the greatest change at the cutoff levels of 16%–18%, decreasing from 0.991 to 0.986, showing a reduction of 0.005. There was no change in the similarity at the cutoff levels of 2%–4%. In the cutoff levels of 4%–10% and 12%–16%, the similarity decreased by 0.001 for every 2% increase in relative abundance.

RMSE was calculated to compare the error values between the groups. RMSE represents average difference between the groups and analyze the effects of indicator-diatom taxa to TDI. Therefore, threshold was set the 5% of the maximum shift in RMSE following Lavoie et al. (2009) and it was 5.

The RMSE values were below the threshold at all cutoff levels and showed a tendency to increase as the relative abundance increased. The RMSE value showed the greatest increase from 2.00 to 2.96 after the 10% cutoff level (Table 2).

The scatter plots of the groups showed singular values and the cutoff levels were increased, the number of singular values were increased (Fig. 3).

The cutoff level based on the relative abundance used to simplify the diatom list in TDI was determined to minimize the significant impact on RMES. This study focuses more on reducing the error between assessments using fewer species and traditional assessments, rather than just reducing the number of species needed for water quality assessment.

Although the RMSE were under the threshold at all cutoff levels, the RMSE value increased the most from 2.00 in cutoff level of 10%, to 2.96 at 12%. Therefore, 10% was selected as the cutoff levels based on relative abundance for selecting the indicator-diatom taxa.

As a result, the total number of species presented in this study was 411. However, it was simplified to 158 species by selecting species with relative abundance cutoff level of 10%.

2nd step for selection of the indicator-diatom taxa

From the previously selected 158 species, the frequency of occurrence was calculated for each diatom species to select the indicator-diatom taxa. Ten frequency of occurrence cutoff levels were set at 1% intervals from 1% to 10%. Species with a maximum frequency of occurrence above the cutoff level at least once in the sampling stations were selected. Ten frequency of occurrence cutoff levels were set at 1% intervals from 1% to 10%.

The Pearson correlation analysis was conducted to compare the similarity between the groups of the TDI recalculated using species selected according to the cutoff levels based on frequency of occurrence and the TDI calculated using the original method.

The cutoff level based on frequency of occurrence changed from 2% to 3%, the similarity changed from 0.974 to 0.956. These results showed the greatest change of similarity (Table 3).

RMSE was calculated to compare the error values between the groups. The threshold was set the same with the first step as 5. The RMSE exceeded threshold with 6.42 at the 3% cutoff level and showed the greatest change from 4.90 to 6.42 (Table 3).

The scatter plots were conducted to compare the similarity between the groups of the TDI recalculated using species selected according to the cutoff levels based on frequency of occurrence and the TDI calculated using the original method. The singular values which were not ob-

Table 2 The changes of similarity and RMSE according to cutoff level based on relative abundance

	Cutoff levels based on relative abundance (%)									
	2%	4%	6%	8%	10%	12%	14%	16%	18%	20%
Similarity	0.999	0.999	0.998	0.997	0.996	0.993	0.992	0.991	0.986	0.984
RMSE	0.69	0.91	1.24	1.75	2.00	2.96	3.13	3.40	4.29	4.62

RMSE: root mean square error.

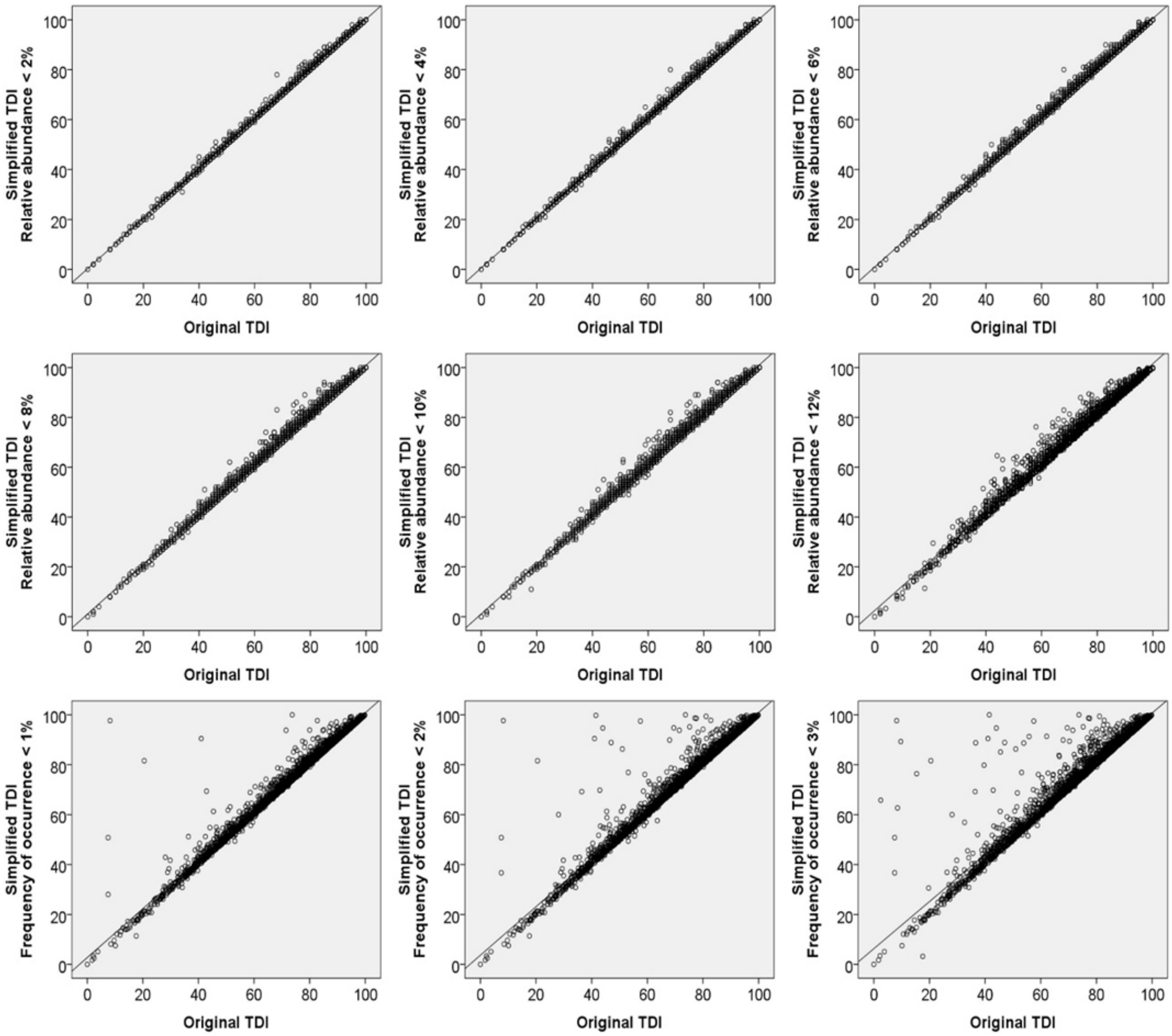

Fig. 3 Identification of singular values using scatter plots between the original TDI as the reference and simplified TDI calculated using selected species based on different criterion and cutoff levels. Original TDI means the TDI calculated by the method of NIER (2019). TDI: trophic diatom index.

Table 3 The changes of similarity and RMSE according to cutoff level based onfrequency of occurrence

	Cutoff levels based on frequency of occurrence (%)									
	1%	2%	3%	4%	5%	6%	7%	8%	9%	10%
Similarity	0.985	0.974	0.956	0.954	0.951	0.947	0.939	0.934	0.934	0.929
RMSE	3.79	4.90	6.42	6.62	6.81	7.16	7.88	8.20	8.26	8.56

RMSE: root mean square error.

served in the first step's scatter plot were appeared with large error (Fig. 3).

The RMSE calculated using diatom group selected by cutoff level based on frequency of occurrence had large error in comparison with the RMSE calculated using diatom group selected by cutoff level based on relative abundance.

The frequency of occurrence of 2% was below the critical value, it was selected as the cutoff levels based on frequency of occurrence for selecting the indicator-diatom taxa. As a result, a total of 115 species were finally selected as the indicator-diatom taxa (Table S1).

Selecting identification method of species excluded from the indicator-diatom taxa

The species excluded from selecting the indicator-diatom taxa needed the simplified process for the TDI. Therefore, three methods were suggested in this study. The first method is to count the species excluded from the list of indicator-diatom taxa at the genus-level not the species-level. The genus-level pollution sensitivities and indicator values were used by calculating the average of the species-level pollution sensitivities and indicator values used in TDI presented by NIER (2019).

The second method is to count the species excluded from the indicator-diatom taxa as a single unidentified group. This method uses the average of the pollution sensitivity and indicator value of all species used in TDI presented by NIER (2019) to calculate the pollution sensitivities and indicator values of the unidentified group. Pollution sensitivity was calculated as 4 points, and indicator value was calculated as 2 points. The unidentified group was referred to as "Diatom (other)" in this study.

The third method is not to count the species excluded from the indicator-diatom taxa. That is, only the indicator-diatom taxa were included in counting for TDI without the unidentified taxa group.

To compare the similarity between the TDI calculated based on the assessment process of species excluded from the indicator-diatom taxa and the TDI calculated using the original method, Pearson correlation coefficients were performed (Table 4).

Table 4 Changes in TDI similarity according to counting methods for excluded species in indicator-diatom taxa

	Genus	Diatom	Exclusion
Similarity	0.999	0.995	0.972
RMSE	1.09	1.99	4.90

This table compared the similarity based on the original TDI. Original TDI: The TDI calculated by the method of NIER (2019).
Genus TDI: The TDI calculated by counting species other than indicator-diatom taxa at the genus level; Diatom TDI: The TDI calculated by counting non indicator-diatom taxa as unidentified group; Exclusion TDI: The TDI calculated without counting species other than indicator-diatom taxa; TDI: trophic diatom index.

The method with the highest similarity to the TDI calculated using the original methods was the genus-level counting method, with a correlation coefficient of 0.999. The method of counting the species as a single unidentified group had a correlation coefficient of 0.995, and the method of not counting and excluding had the lowest correlation coefficient of 0.972. All three methods showed a significance level of $p < 0.01$.

RMSE was calculated to compare the errors between TDI calculated using three methods and TDI calculated using the original method. The threshold was set the same with the first step as 5. The genus-level counting method had the lowest RMSE of 1.09, a single unidentified group counting method had an RMSE of 1.99, and the method of not counting and excluding had the largest RMSE of 4.90 (Table 4).

In the comparison of scatter plots, the method which have the least number of singular values was the genus-level counting method, while the method which have the largest number of singular values was the method of not counting (Fig. 4).

All three methods did not exceed the threshold of RMSE. However, the method of not counting showed the highest number of singular values in the scatter plot. Therefore, the main method for counting species that were excluded from the selection of indicator-diatom taxa is the genus-level counting method. However, for diatoms that are difficult to identify even at the genus-level, a method of counting them into a single unidentified group and counting them is also used in combination.

Using the S-TDI for the actual field test on the water quality assessment

S-TDI is the newly proposed method of water quality assessment using diatoms in this study. S-TDI is the simplified method that uses the indicator-diatom taxa to identifying and counting species. In addition, species not included

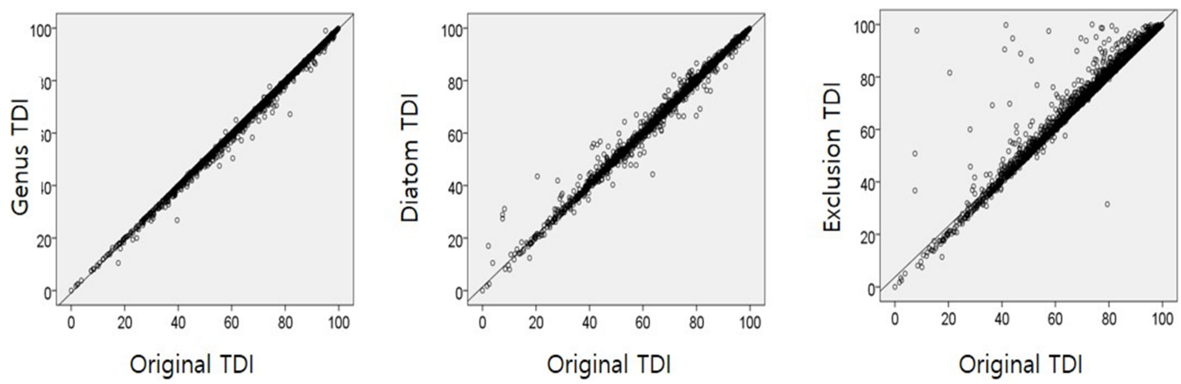

Fig. 4 Identification of singular values using scatter plots between the original TDI as the reference and simplified TDI (Genus TDI, Diatom TDI and Exclusion TDI). Original TDI means the TDI calculated by the method of NIER (2019). Genus TDI means a TDI calculated by counting species other than indicator-diatom taxa at the genus level. Diatom TDI means a TDI calculated by counting non indicator-diatom taxa as one unidentified group. Exclusion TDI means the TDI calculated without counting species other than indicator-diatom taxa. The central line crosses the graph represents the fitted line, which is a line that best represents the relationship between the two variables being compared. TDI: trophic diatom index.

Table 5 Statistical differences, RMSE and similarity between original TDI and S-TDI

	Descriptive statistics			t(*p*)	RMSE	Similarity
	N	M	SD			
Original TDI	60	66.20	19.96	–1.176	1.52	0.999**
S-TDI	60	66.41	19.70			

Original TDI means the TDI calculated by the method of NIER (2019). S-TDI is newly proposed in this study. A statistical measure that indicates the degree of variability in the data. t-value: a statistical measure used to test whether the difference between the means of two groups is statistically significant. In a paired t-test, if the *p*-value is greater than 0.05, it indicates that there is no statistically significant difference. The threshold for RMSE was set to 5. The similarity was calculated through pearson correlation analysis.
TDI: trophic diatom index; S-TDI: simplified trophic diatom index; RMSE: root mean square error; N: number of samples; M: the average of TDI values; SD: standard deviation.
** $p < 0.01$.

in the indicator-diatom taxa are identified to genus-level and the species that are difficult to identify even at the genus-level are counted as a single unidentified group called "Diatom (other)".

TDI and S-TDI were calculated for each of the 60 sampling stations presented in Figure 1 and the results were compared. To compare the similarity between the TDI and S-TDI values, pearson correlation coefficients was conducted. The similarity was 0.999, and the significance level was $p < 0.01$, this result showed a high degree of similarity between the two indices (Table 5).

In addition, a paired sample t-test was conducted to assess the consistency of results between TDI and S-TDI (Table 5). The difference in the mean (M) TDI and S-TDI was 0.21, and a difference in standard deviation (SD) was 0.26. However, the significance level of the t-value indicating the statistical difference between the two indices was $p > 0.05$. This result indicated that TDI and S-TDI were statistically the same group.

Although there was no statistically difference between the two groups, RMSE was calculated to confirm the difference. The RMSE value was 1.52, which was below the threshold (Table 5).

Based on the results of TDI and S-TDI, a scatter plot was created and compared (Fig. 5). The values were distributed near the fitted line. The 95% range of the normal distribution was indicated by two lines near the fitted line, and singular values were not observed outside this range.

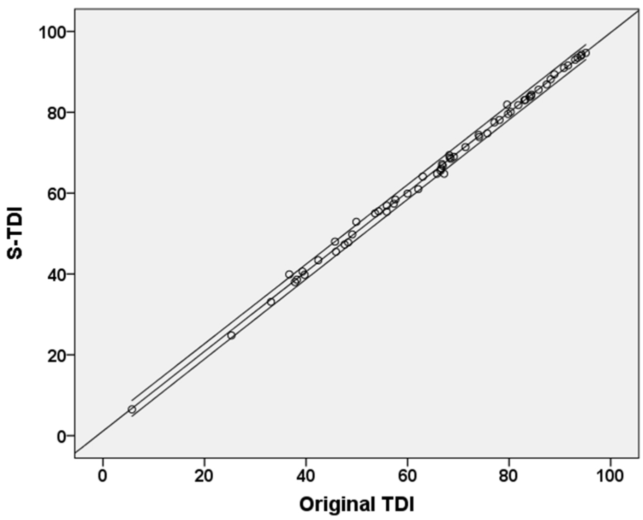

Fig. 5 Identification of singular values using scatter plot between the original TDI as the reference and the S-TDI. Original TDI means the TDI calculated by the method of NIER (2019). S-TDI means a TDI calculated by counting species other than indicator-diatom taxa at the genus level. The central line crosses the graph represents the fitted line, which is a line that best represents the relationship between the two variables being compared. The diagonal lines on both sides of the fitted line represent the 95% confidence interval. TDI: trophic diatom index; S-TDI: simplified trophic diatom index.

Discussion

The diatom-indicator taxa were selected using relative abundance and frequency of occurrence each species. In the selecting process, relative abundance and frequency of occurrence cutoff levels increased, more species were excluded and the ecological informations were missing such as pollution sensitivity and indicator value. Therefore, the TDI values were changed and the similarities decreased and the number of singular value and RMSE increased. These changes were significant especially in second process using frequency of occurrence. These results were similar to those of Lavoie et al. (2009). They compared IDEC (Lavoie et al. 2006) and IDEC recalculated using simplified diatom metrix by statistical methods to provide a user-friendly tool for routine use in diatom-based water quality assessment. They excluded diatom species for simplifying diatom metrix using cutoff levels based on relative abundance and the scatter plot of IDEC and IDEC recalculated using simplified diatom metrix showed increased the number of singular values and RMSE as cutoff levels based on relative abundance increased.

In addition, Lavoie et al. (2009) compared using taxa excluded based on the frequency of occurrence with IDEC using taxa excluded based on relative abundance. IDEC using taxa excluded based on the frequency of occurrence had a large error compared to IDEC using taxa excluded based on relative abundance.

Nijboer and Schmidt-Kloiber (2004), who conducted an ecological assessment using macroinvertebrates based on species diversity, reported that the ecological assessment based on relative abundance did not significantly affect the results, even when low relative abundance species were excluded. However, the species with low frequencies of occurrence often reflect specific ecological conditions. Therefore, excluding species with low frequencies of occurrence were significantly affected the ecological assessment results. Therefore, identification method of species excluded from the indicator-diatom taxa had to be found.

To find the counting method for excluded from indicator-diatom taxa, three methods were compared and the method counting Genus-level was most appropriate method.

Growns (1999) reported that there is no significant difference between genus-level and species-level for ecological assessment results using diatoms. Additionally, Riato et al. (2022) started that ecological assessments based on genus-level counts with genus specific characteristics and those based on species-level counts did not have a significant difference. These results were similar the result of this study. The result showed that the most appropriate method for counting species excluded from the selection of indicator-diatom taxa was the genus-level counting method. In addition, the method of counting as a single unidentified group can be used too.

S-TDI using indicator-diatom taxa, counting genus-level and single unidentified group was proposed in this study. Verifying the application of S-TDI in actual field, 60 sampling stations were selected considering TDI values and grades variously for representing various environmental conditions. The result comparing TDI and S-TDI was indicated that S-TDI can be used to water quality assessment.

Conclusions

In this study, it is proposed that a new water quality assessment method called S-TDI for reducing the difficulties of taxonomical identification. The following results were obtained through the process of selecting indicator-diatom taxa, selected the assessment process of species excluded from the indicator-diatom taxa and the practical application process of S-TDI.

Cutoff level based on the relative abundance is the most appropriate to the exclusion of taxa. However, cutoff level based on the frequency of occurrence lead to singular vales significantly due to losing the ecological informations. Those were associated with excluded species from indicator-diatom taxa were important to water quality assessment and it was necessary to minimize them. Therefore, genus-level identification was appropriate method for minimizing the missing informations.

To compare the S-TDI and original TDI, both methods were conducted in 60 sampling stations considering various aquatic environments and showed similar results.

Due to the difficulty in taxonomic identification caused by the large numbers of species and the micro structures of diatoms, the use of water quality assessment based on diatoms has been somewhat limited. Based on those results, water quality assessment was conducted using indicator-diatom taxa instead of all diatoms and found the potential for practical water quality assessment using S-TDI.

Supplementary Information

Supplementary information accompanies this paper at https://doi.org/10.5141/jee.24.095.

Table S1. The list of the indicator-diatom taxa and genera for calculate the S-TDI.

Abbreviations

IDEC: Indice Diatomées de l'Est du Canada
MOE: Ministry of Environment
NIER: National Institute of Environmental Research
RMSE: Root mean square error
S-TDI: Simplified trophic diatom index
TDI: Trophic diatom index
TDIm: Modified trophic diatom index

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Not applicable.

Authors' contributions

SMK designed the study, carried out the fieldwork, performed the analysis and wrote the manuscript. JHL supervised the research and revised the manuscript.

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Availability of data and materials

Data presented in this study will be available on a fair request to the corresponding author.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The author declares that they have no competing interests.

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