



sucipto hariyanto <sucipto-h@fst.unair.ac.id>

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## Editorial system registration

1 message

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Mon, Aug 2, 2021 at 12:30 PM

To: sucipto-h@fst.unair.ac.id



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Your password is: **987siux6g1**

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Editorial Office

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sucipto hariyanto <sucipto-h@fst.unair.ac.id>

## Acknowledgement of Submission (#GJESM-2108-3815)

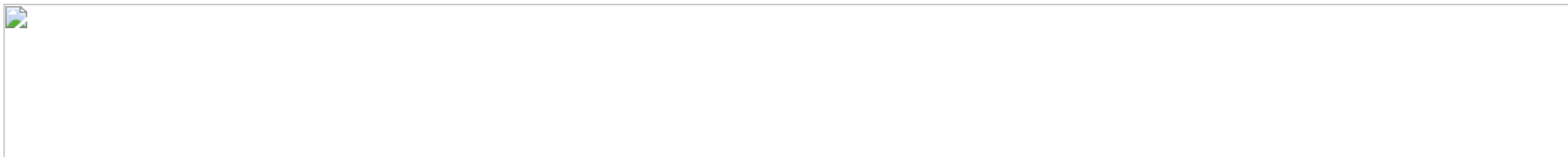
2 messages

Global Journal of Environmental Science and Management <no-reply@sinaweb.net>

Mon, Aug 2, 2021 at 2:59 PM

To: sucipto-h@fst.unair.ac.id

Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id, gjesm.publication@gmail.com, sivakumar.gjesm@gmail.com



# Global Journal of Environmental Science and Management

| Quarterly Publication |

Author Resubmit:

Manuscript ID: GJESM-2108-3815

Manuscript Title: **Characteristics assessment of dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto**

I wish to acknowledge receiving the above-mentioned manuscript. It should be noted that the manuscript will be reviewed for possible publication in the Global Journal of Environmental Science and Management (GJESM). The regular review process includes the following items:

- 1- Checking the manuscript English language
- 2- Format/Style check based on the GJESM Paper Template
- 3- Plagiarism check (less than 15% duplicates)

10/20/23, 12:26 PM

Airlangga University Mail - Acknowledgement of Submission (#GJESM-2108-3815)

4- Reaching at the blind peer reviews the manuscript at least by two potential reviewers in the field of the study, if it passes the three above-mentioned items.

Please be sure that the submitted manuscript has not been published or submitted elsewhere prior to GJESM decision.

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GJESM Journal is mainly indexed in Web of Science and Scopus. The editorial initial screening of the submitted manuscript will immediately forward you within 1 to 2 days so that you can review it beforehand for the further peer review. The peer review is completed through a fast-tracked model in about 2 to 3 weeks up to the entire publication.

It should also be noted that according to the new GJESM editorial decision, all articles submitted after March 1, 2021 will apply the Article Processing Charge (APC). Therefore, to publish open access in the GJESM Journal, international authors are eligible to pay APC: USD500.00 (for internal authors: 1.000.000 Tomans), when it passes positively through the peer reviews. Otherwise, you must immediately withdraw your submitted paper.

Finally, the GJESM Publisher has decided to award an annual prize to the most innovative paper of the year as USD 1000.

I wish to take this opportunity to thank you for sharing your work with GJESM Journal.

Truly yours,

Professor J. Nouri

Editor in Chief

**Global Journal of Environmental Science and Management**

---

**sucipto hariyanto** <sucipto-h@fst.unair.ac.id>  
To: okik hendriyanto <okik.hendriyanto.cahyonugroho-2019@fst.unair.ac.id>

Mon, Aug 2, 2021 at 6:45 PM

[Quoted text hidden]



sucipto hariyanto <sucipto-h@fst.unair.ac.id>

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## Manuscript Needs Resubmission (#GJESM-2108-3815)

1 message

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Wed, Aug 4, 2021 at 3:09 PM

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Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id, gjesm.publication@gmail.com



Manuscript ID: GJESM-2108-3815

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto**,

Thank you very much for your interest in publishing your work in the **Global Journal of Environmental Science and Management**. Your manuscript recorded above cannot be considered for possible publication in the presented form.

Therefore, we suggest you revise your attached manuscript CAREFULLY where you can find it through the website system which some editorial comment is proposed on the proposed file. Thus, after correction, resubmit it through your own dashboard on the journal website system. To resubmit your manuscript, log into <https://www.gjesm.net/> and enter as "Author", where you will find your manuscript title listed under "Manuscript needs to be resubmitted".

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Professor J. Nouri

10/20/23, 12:28 PM

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Editor-in-chief

**Global Journal of Environmental Science and Management,**



## Acknowledgement of Submission (#GJESM-2108-3815)

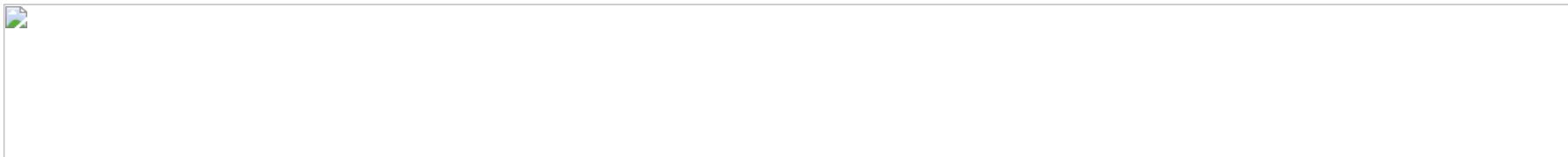
4 messages

**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Wed, Aug 4, 2021 at 1:58 PM

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Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id, gjesm.publication@gmail.com, sivakumar.gjesm@gmail.com



# Global Journal of Environmental Science and Management

| Quarterly Publication |

Author Resubmit:

Manuscript ID: GJESM-2108-3815

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Truly yours,

Professor J. Nouri

Editor in Chief

**Global Journal of Environmental Science and Management**

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Wed, Aug 4, 2021 at 3:08 PM

To: [sucipto-h@fst.unair.ac.id](mailto:sucipto-h@fst.unair.ac.id)

Cc: [okhecah@gmail.com](mailto:okhecah@gmail.com), [ganden-s@fst.unair.ac.id](mailto:ganden-s@fst.unair.ac.id), [gjesm.publication@gmail.com](mailto:gjesm.publication@gmail.com), [sivakumar.gjesm@gmail.com](mailto:sivakumar.gjesm@gmail.com)

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Wed, Aug 4, 2021 at 7:27 PM

To: [sucipto-h@fst.unair.ac.id](mailto:sucipto-h@fst.unair.ac.id)

Cc: [okhecah@gmail.com](mailto:okhecah@gmail.com), [ganden-s@fst.unair.ac.id](mailto:ganden-s@fst.unair.ac.id), [gjesm.publication@gmail.com](mailto:gjesm.publication@gmail.com), [sivakumar.gjesm@gmail.com](mailto:sivakumar.gjesm@gmail.com)

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Thu, Aug 5, 2021 at 1:59 AM

To: [sucipto-h@fst.unair.ac.id](mailto:sucipto-h@fst.unair.ac.id)

Cc: [okhecah@gmail.com](mailto:okhecah@gmail.com), [ganden-s@fst.unair.ac.id](mailto:ganden-s@fst.unair.ac.id), [gjesm.publication@gmail.com](mailto:gjesm.publication@gmail.com), [sivakumar.gjesm@gmail.com](mailto:sivakumar.gjesm@gmail.com)

[Quoted text hidden]



sucipto hariyanto <sucipto-h@fst.unair.ac.id>

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**RE: Submitted Manuscript #3815 Edited**

1 message

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**GJESM Journal** <gjesm.publication@gmail.com>  
To: sucipto-h@fst.unair.ac.id  
Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id

Thu, Aug 5, 2021 at 2:18 AM

Dear Author(s),

Your attached file is just gone for the fast-tracked review. The reason we attach the primary final revision for you is because every time you change the name of the file as well as change the editorial format such as the Tables titles and Figures captions must be in font 10p.

Thus, just treat the attached file (#3815 Edited) which is also archived at your dashboard for your possible acceptance further process.

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Editorial Team  
Global Journal of Environmental Science and Management (GJESM)

Tel.: +9821- 26105110  
Fax: +9821- 26105110

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 **3815 Edited.docx**  
1219K





sucipto hariyanto <sucipto-h@fst.unair.ac.id>

---

**RE: Your 3 proposed Reviewers**

1 message

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>  
To: sucipto-h@fst.unair.ac.id

Sat, Aug 14, 2021 at 10:00 PM



GJESM-2108-3815

**Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Dear Author(s),

As the GJESM Journals are reviewers reviewed your paper positively, if one of your assigned reviewers responds as soon as possible, your the case will be forwarded to you for your action.

Editorial Office



sucipto hariyanto <sucipto-h@fst.unair.ac.id>

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## Manuscript Needs Major Revision (#GJESM-2108-3815 (R1))

1 message

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Mon, Aug 16, 2021 at 1:55 PM

To: sucipto-h@fst.unair.ac.id

Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id, gjesm.publication@gmail.com



Manuscript ID: GJESM-2108-3815 (R1)

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho,Sucipto Hariyanto,Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto**

Your manuscript has now been peer reviewed and virtually resulted as **Major Revision**. As numerous technical comments have pointed out by the reviewers, please revise your manuscript carefully according to the reviewers' comments as well as the editor notifications, within 10 days.

Should the reviewers and editor be satisfied with your amendments, you will be notified the acceptance of your manuscript for publication.

Please do not hesitate to contact us if you have any further inquiries regarding your manuscript revision.

Wishing you every success in your future endeavors.

Truly yours,

**Managing Editor**

**Global Journal of Environmental Science and Management**

Dear Author(s),

- 1- Your manuscript review has now been performed as a fast-tacked peer reviewed exceptionally and virtually resulted as Minor Revision where the three reviewers' comments can be found through your own dashboard in the website system (#3815). Therefore you must satisfy the both reviewers by your accurate and completed revisions.
- 2- You will also find an "Author Query Form" that you have to add the manuscript reviewer' inquiries and then response each item into the "Author's response" column carefully and correctly in order to be recognized by the reviewers and editor.
- 3- REMEMBER, any alteration and corrections must be done JUST on the attached final modified manuscript file (#3815 Edited), highlighting with RED paint in order to be recognized by the Editor and Reviewers
- 4- After completion of your paper revisions, please resubmit your revised manuscript as well as the completed Author Query Form file through your own dashboard back (#3815) where you had already submitted your manuscript in the system as soon as possible to avoid any further delays for the latter processing as your paper may be included at the forthcoming issue publication as the last released package.
- 5- Please also finalize your manuscript English content, otherwise, the manuscript processing will be delayed.
- 6- Should the reviewers and editor be satisfied with your amendments, you will be notified to receive the Galley Proof and related copyright forms.
- 7- You will have 5 days' time to return your completed revised manuscript back. In case of not receiving your revised file after the deadline, we assume that you do not like or cannot revise your last stage of article process of publication and therefore your file will be closed with no further action.

Editorial Office

Reviewers Recommendation:

**Reviewer 1:**

Reviewer Comment For Author:

Comments:

1. The author should present in the Abstract the quantity of any units in terms of the resulting values or the resulting data of the four components of dissolved organic matter namely AP-like, HA-like, SMPs-like, and FA-like to make the findings more specific.
2. The Abstract mentions an acronym (FRI) that is not spelled out. According to the GJESM Authors Guideline for Manuscript Preparation, the Abstract should not contain any undefined abbreviation.
3. The manuscript title highlighted water quality where other parameters may be of important considerations such as total suspended solids, turbidity, dissolved oxygen, etc. Any justifications why these parameters are not considered in the study?
4. With "water quality" as one of the mean contents of the manuscript title, a specific review of literature about water quality is helpful to enhance the scientific basis of the study. The literature can be seen in the following link:  
<https://www.innspub.net/wp-content/uploads/2018/04/JBES-Vol-12-No-3-p-201-209.pdf>
5. It is highly suggested to present a GIS-based generated map of the study site to make the presentation more comprehensible.
6. The reviewer assumes to have only two (2) replicates of samples being carried out in the study based on the number of data collected which was done twice per week from January to March 2021. Generally, in biology using one-way ANOVA needs at least three (3) replicates to make it acceptable. Any justification for this query?
7. The manuscript could be more enhanced if the author/s will present a precise description of the study site in terms of climatic, environmental, or bio-physical conditions.
8. The paper would be more robust if the author/s present a discussion about the other sources of dissolved organic matter which may be included in the correction analysis between the phytoplankton abundance and the amount of DOM in the surface water.
9. Some of the labels in Fig. 4: are too small. It is suggested to increase their font size to enhance readability.
10. The labels in Figures 6 and 7 are too small. They should be improved.
11. The discussions mention Figures 6a and 6d as well as Figures 7a and 7d. However, these labels are not found in the respective graphs.
12. The authors have to recheck carefully the language, grammar including the punctuation of the entire manuscript.
13. The conclusion is too generic. A conclusion should highlight the significant findings supported with specific values of the study results.

**Reviewer 2:**

Reviewer Comment For Author:

The manuscript is well written and the methodology was well developed.

Minor recommendations:

1. Improve the description in the legend of the figures. The reader should know that it is Sta. 1, Sta. 2, Sta. 3 and Sta. 4 without having to search in the text.
2. In Figures 4 and 5, improve the quality of the axis labels.

3. It would be interesting to have an image of the study area, since this could contextualize the readers about the characteristics and functioning of the ecosystem. Which could be modulating the behavior found.
4. In Table 1, where the results of the correlation are shown, not only the value of the correlation coefficient should be shown, but also the p-value.

**Reviewer 3:**

Reviewer Comment For Author:

1. Why does surface water have quite high TOC concentration, is it probably due to phytoplankton activities?
2. Figure 4, the author showed I, II, IV, it is confused. The reviewer suggested that I, II, IV should be written as name of region, such as AP-like, etc., as mentioned on the paragraph.
3. How do you define the area of spectrum Figure 4? which reference?
4. Did the author found the other species of phytoplankton? how is the abundance value of those phytoplankton?
5. This study should make a suggestion that further research in the lab scale will be conducted in order to identify the characteristic of organic matter has been released by a kind of phytoplankton species.
6. Table 1 seems a miss typed or TOP, it should be TOC.



sucipto hariyanto <sucipto-h@fst.unair.ac.id>

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## Manuscript Needs Major Revision (#GJESM-2108-3815 (R1))

1 message

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Mon, Aug 16, 2021 at 1:55 PM

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Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id, gjesm.publication@gmail.com



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## Acknowledgement of Revision (#GJESM-2108-3815 (R1))

1 message

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**Global Journal of Environmental Science and Management** <no-reply@sinaweb.net>

Tue, Aug 17, 2021 at 7:46 PM

To: sucipto-h@fst.unair.ac.id

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Manuscript ID: GJESM-2108-3815 (R1)

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Date: 2021-08-04

Dear **Dr. Sucipto Hariyanto**

Thank you for submitting the revised file of your manuscript to the **Global Journal of Environmental Science and Management**

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Managing Editor



10/20/23, 12:35 PM

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sucipto hariyanto <sucipto-h@fst.unair.ac.id>

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## Request for Submit/Confirm Galley Proof (#GJESM-2108-3815 (R1))

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Wed, Aug 18, 2021 at 12:34 PM

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Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id, gjesm.publication@gmail.com



Manuscript ID: GJESM-2108-3815 (R1)

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto**

We are pleased to inform you that your paper for the GJESM journal is ready for publication. The page proofs are available at:

<https://www.gjesm.net/>

Attached, you will find three files through the system. 1) Galley Proof, 2) Copyright Release, 3) Conflict of Interest Forms. Please read and revise the Galley Proof carefully, if required to be corrected. Therefore, all new corrections must be highlighted with RED fonts to be recognized by the editor. After the final correction, complete and sign the two copyright forms and immediately return them into PDF format for final steps through your own dashboard via the system. In addition, finalize English correction if required. The contents must be returned immediately for the entire online publication.

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Professor D. Sivakumar

Managing Editor

10/20/23, 12:35 PM

Airlangga University Mail - Request for Submit/Confirm Galley Proof (#GJESM-2108-3815 (R1))

**Global Journal of Environmental Science and Management**

(URGENT PLEASE)!

Dear Author,

Attached through your dashboard in the website system, you will find three files; 1) Galley Proof, 2) Copyright release, 3) Conflict of interest forms.

Please read the Galley Proof carefully and correct any remaining minor corrections. Then, complete and sign the two forms and return them into PDF format through your dashboard (#3815) in the system immediately for the final processes.

In addition, finalize the English language correction, if required. The materials must be returned immediately for the entire publication as the forthcoming issue is going to be released.

Editorial Office



sucipto hariyanto &lt;sucipto-h@fst.unair.ac.id&gt;

---

**The Final Stage of Publication (APC payment for Paper # 3815)**

1 message

---

**Jafar Nouri** <nourijafar@gmail.com>  
To: sucipto-h@fst.unair.ac.id  
Cc: okhecah@gmail.com, ganden-s@fst.unair.ac.id

Wed, Aug 18, 2021 at 8:33 PM

(URGENT MESSAGE)

Dear Author(s),

1- First it is necessary to mention that in the time of spiking the hyperlinks into the references, most of the reference parts were deleted by your mandatory action. You were lucky that we found out and recovered them. You may view from your first Galley Proof as well as the finalized file as attached!

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Manuscript ID: GJESM-2108-3815 (R1)

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto****Acceptance Letter**

This is to confirm that after technical and in-house evaluation, the above mentioned manuscript is finalized and recommended by the Editorial Board Committee to be accepted for publication in the Global Journal of Environmental Science and Management (GJESM).

It is necessary to mention that GJESM is an open access, double-blind, peer reviewed quarterly publication, which is indexed and cited in the well-known world databases mainly at the [Web of Science](#), [Scopus](#), [SJR \(Q2\)](#), [EBSCO](#), [ProQuest](#), [Ulrichsweb](#), [Cabi](#), [Agricola](#) and [Chemical Abstract](#). The title is committed to the Committee on Publication Ethics (COPE) and meets the highest ethical standards in accordance with ethical rules (COPE).

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Manuscript ID: GJESM-2108-3815 (R1)

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto**

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Manuscript ID: GJESM-2108-3815 (R1)

Manuscript Title: **Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality**

Authors: Okik Hendriyanto Cahyonugroho, Sucipto Hariyanto, Ganden Supriyanto

Dear **Dr. Sucipto Hariyanto**

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Please note that from now on, in case of your current published article gets more citations, in different sources specially in the Scopus and Web of Science journal articles as well as the social media, your future submitted manuscripts will have a better positive impact on easier acceptance in the GJESM Journal. In this regard, We hope to receive your highly cited scientific articles in the future.

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**REVIEWER RECOMMENDATIONS AND  
COMMENTS & RESPONSES TO  
REVIEWER COMMENTS**

## REVIEWER COMMENTS

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2- You will also find an "Author Query Form" that you have to add the manuscript reviewer' inquiries and then response each item into the "Author's response" column carefully and correctly in order to be recognized by the reviewers and editor.

3- REMEMBER, any alteration and corrections must be done JUST on the attached final modified manuscript file (#3815 Edited), highlighting with RED paint in order to be recognized by the Editor and Reviewers

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Reviewers Recommendation:

**Reviewer 1:**

Reviewer Comment For Author:

Comments:

1. The author should present in the Abstract the quantity of any units in terms of the resulting values or the resulting data of the four components of dissolved organic matter namely AP-like, HA-like, SMPs-like, and FA-like to make the findings more specific.

2. The Abstract mentions an acronym (FRI) that is not spelled out. According to the GJESM Authors Guideline for Manuscript Preparation, the Abstract should not contain any undefined abbreviation.

3. The manuscript title highlighted water quality where other parameters may be of important considerations such as total suspended solids, turbidity, dissolved oxygen, etc. Any justifications why these parameters are not considered in the study?
4. With “water quality” as one of the main contents of the manuscript title, a specific review of literature about water quality is helpful to enhance the scientific basis of the study. The literature can be seen in the following link:  
<https://www.innspub.net/wp-content/uploads/2018/04/JBES-Vol-12-No-3-p-201-209.pdf>
5. It is highly suggested to present a GIS-based generated map of the study site to make the presentation more comprehensible.
6. The reviewer assumes to have only two (2) replicates of samples being carried out in the study based on the number of data collected which was done twice per week from January to March 2021. Generally, in biology using one-way ANOVA needs at least three (3) replicates to make it acceptable. Any justification for this query?
7. The manuscript could be more enhanced if the author/s will present a precise description of the study site in terms of climatic, environmental, or bio-physical conditions.
8. The paper would be more robust if the author/s present a discussion about the other sources of dissolved organic matter which may be included in the correlation analysis between the phytoplankton abundance and the amount of DOM in the surface water.
9. Some of the labels in Fig. 4: are too small. It is suggested to increase their font size to enhance readability.
10. The labels in Figures 6 and 7 are too small. They should be improved.
11. The discussions mention Figures 6a and 6d as well as Figures 7a and 7d. However, these labels are not found in the respective graphs.
12. The authors have to recheck carefully the language, grammar including the punctuation of the entire manuscript.
13. The conclusion is too generic. A conclusion should highlight the significant findings supported with specific values of the study results.

### **Reviewer 2:**

Reviewer Comment For Author:

The manuscript is well written and the methodology was well developed.

Minor recommendations:

1. Improve the description in the legend of the figures. The reader should know that it is Sta. 1, Sta. 2, Sta. 3 and Sta. 4 without having to search in the text.
2. In Figures 4 and 5, improve the quality of the axis labels.
3. It would be interesting to have an image of the study area, since this could contextualize the readers about the characteristics and functioning of the ecosystem. Which could be modulating the behavior found.
4. In Table 1, where the results of the correlation are shown, not only the value of the correlation coefficient should be shown, but also the p-value.

### **Reviewer 3:**

Reviewer Comment For Author:

1. Why does surface water have quite high TOC concentration, is it probably due to phytoplankton activities?

2. Figure 4, the author showed I, II, II, IV, it is confused. The reviewer suggested that I, II, II, IV should be written as name of region, such as AP-like, etc., as mentioned on the paragraph.
3. How do you define the area of spectrum Figure 4? which reference?
4. Did the author found the other species of phytoplankton? how is the abundance value of those phytoplankton?
5. This study should make a suggestion that further research in the lab scale will be conducted in order to identify the characteristic of organic matter has been released by a kind of phytoplankton species.
6. Table 1 seems a miss typed or TOP, it should be TOC.

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#### Reviewer # 1:

Query	Review Details Required	<b>Author's Response</b> (Author <b>MUST</b> show the place of performed corrections in the revised manuscript at this column)
1.	The author should present in the Abstract the quantity of any units in terms of the resulting values or the resulting data of the four components of dissolved organic matter namely AP-like, HA-like, SMPs-like, and FA-like to make the findings more specific.	The quantity of unit fluorescence spectra is arbitrary unit (A.U). It has been added on the abstract of the revised manuscript. (Page 1)
2.	The Abstract mentions an acronym (FRI) that is not spelled out. According to the GJESM Authors Guideline for Manuscript Preparation, the Abstract should not contain any undefined abbreviation.	The acronym of FRI is Fluorescence Regional Integration, it has been added on the abstract of the revised manuscript. (Page 1)
3.	The manuscript title highlighted water quality where other parameters may be of important considerations such as total suspended solids, turbidity, dissolved oxygen, etc. Any justifications why these parameters are not considered in the study?	The title highlighted water quality, however the parameter such as total suspended solids, turbidity, dissolved oxygen, etc. are not included in the manuscript because this manuscript focused on dissolved organic matter, since the author assumed that phytoplankton could give effect on the quantity and quality of organic matter in the aquatic. However, this study measured the parameters of water quality, such as total organic carbon.
4.	With "water quality" as one of the mean contents of the manuscript title, a specific review of literature about water quality is helpful to enhance the scientific basis of the study. The literature can be seen in the following link: <a href="https://www.innspub.net/wp-content/uploads/2018/04/JBES-Vol-12-No-3-p-201-209.pdf">https://www.innspub.net/wp-content/uploads/2018/04/JBES-Vol-12-No-3-p-201-209.pdf</a>	The literature suggestion has been added in the revised manuscript. Allochthonous and autochthonous with effluent organic matter are the source of dissolved organic matter (DOM) in the surface water, since allochthonous could be generated from the upstream, midstream and the downstream. The upstream was found to be covered with perennial vegetation; the midstream is used for agriculture and covered with least forest; the downstream was mainly used for residential and utilized for different forms of agriculture (Dumago et al., 2018) (Page 2)

5.	It is highly suggested to present a GIS-based generated map of the study site to make the presentation more comprehensible.	The reviewer suggestion is accepted. A GIS-based generated map of the study site has been added as Figure 1 of the revised manuscript. Therefore, the existing figure has shifted into labels shifted into the following number. (Page 3)
6.	The reviewer assumes to have only two (2) replicates of samples being carried out in the study based on the number of data collected which was done twice per week from January to March 2021. Generally, in biology using one-way ANOVA needs at least three (3) replicates to make it acceptable. Any justification for this query?	The sample was collected twice per week means sampling time has been conducted two times per week. Each sampling time taken three samples or three replicates. We made mistake in type two replications. That has been revised in the last paragraph of material and methods. (Page 4)
7.	The manuscript could be more enhanced if the author/s will present a precise description of the study site in terms of climatic, environmental, or bio-physical conditions.	A description of the study site in terms of climate, environmental condition has been added in Table 1 of the revised manuscript. Table 1 is a new table. (Page 3)
8.	The paper would be more robust if the author/s present a discussion about the other sources of dissolved organic matter which may be included in the correction analysis between the phytoplankton abundance and the amount of DOM in the surface water.	The correction analysis between DOM, which has been released by the phytoplankton abundance, and the amount of DOM in the surface water did not include in this study. However, the reviewer suggestion will be implemented in the future work, as it has been added in the conclusion of the revised manuscript. (Page 12)
9.	Some of the labels in Fig. 4: are too small. It is suggested to increase their font size to enhance readability.	The labels in Figure 4 (rename as Figure 5) has been revised. (Page 7)
10.	The labels in Figures 6 and 7 are too small. They should be improved.	The labels in Figure 6 (rename as Figure 7) and Figure 7 (rename as Figure 8) has been improved. (Page 9 – 10)
11.	The discussions mention Figures 6a and 6d as well as Figures 7a and 7d. However, these labels are not found in the respective graphs.	The labels “a, b, c, d” has been added in the Figure 7 and Figure 8 of the revised manuscript. (Page 9 – 10)
12.	The authors have to recheck carefully the language, grammar including the punctuation of the entire manuscript.	The authors did recheck the language, grammar and the punctuation in the manuscript.
13.	The conclusion is too generic. A conclusion should highlight the significant findings supported with specific values of the study results.	The conclusion has been added significant findings supported with spesific values of the study results. (Page 12)

**Reviewer # 2:**

<b>Query</b>	<b>Review Details Required</b>	<b>Author's Response</b>
1.	Improve the description in the legend of the figures. The reader should know that it is Sta. 1, Sta. 2, Sta. 3 and Sta. 4 without having to search in the text.	The description in the legend of the figure, such as Sta.1, Sta.2, Sta.3, and Sta.4 has been improved into Station 1, Station 2, and Station 3. It has been added in the revised manuscript. (Page 5, 6, 8, 9, 10)
2.	In Figures 4 and 5, improve the quality of the axis labels.	The quality of Figure 4 (rename as Figure 5) and Figure 5 (rename as Figure 6) has been improved. (Page 7 – 8)
3.	It would be interesting to have an image of the study area, since this could contextualize the readers about the characteristics and functioning of the ecosystem. Which could be modulating the behavior found.	An image of the study area has been added as Figure 1 in the revised manuscript. (Page 3)
4.	In Table 1, where the results of the correlation are shown, not only the value of the correlation coefficient should be shown, but also the p-value.	Table 1 (rename as Table 2) has shown the results of the correlation coefficient and the p-value. It has been added on the revised manuscript. (Page 11)
5.		
6.		
7.		
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**Reviewer # 3:**

<b>Query</b>	<b>Review Details Required</b>	<b>Author's Response</b>
1.	Why does surface water have quite high TOC concentration, is it probably due to phytoplankton activities?	Surface water has quite high TOC concentration, it is affected by allochthonous and autochthonous sources. Allochthonous could be generated through watershed discharge into surface water, while autochthonous was produced by microorganism activities in water bodies. This information has been explained in the introduction of the revised manuscript. (Page 2)
2.	Figure 4, the author showed I, II, II, IV, it is confused. The reviewer suggested that I, II, II, IV should be written as name of region, such as AP-like, etc., as mentioned on the paragraph.	The labels "I, II, III, IV" has been revised into " Aromatic protein-like; Fulvic acid -like, Soluble microbials products, humic acid-like" as shown in Figure 4 (rename as Figure 5) of the revised manuscript. (Page 7)
3.	How do you define the area of spectrum Figure 4? which reference?	The area of spectrum has been defined according to the reference of Chen et al., 2003. It has been mentioned in the revised manuscript. (Page 6)
4.	Did the author found the other species of phytoplankton? how is the abundance value of those phytoplankton?	This study was found the other phytoplankton species, such as Oscillatoria sp. (1002 cell/L), Scenedemus sp. (390 cell/L), Spyrogyra sp. (238 cell/L), Synedra sp. (270 cell/L), Terpsione sp. (202 cell/L) and Mougeotya sp. (262 cell/L).
5.	This study should make a suggestion that further research in the lab scale will be conducted in order to identify the characteristic of organic matter has been released by a kind of phytoplankton species.	The reviewer suggestion has been added in the conclusion of the revised manuscript. (Page 12)
6.	Table 1 seems a miss typed or TOP, it should be TOC.	"TOP" has been revised into "TOC" in the Table 1 (rename as Table 2) of the revised manuscript. (Page 11)
7.		
8.		
9.		
10.		

## Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality

### ABSTRACT:

**BACKGROUND AND OBJECTIVES:** Dissolved organic matter has a fundamental role in supporting phytoplankton abundance and growth in aquatic environments. However, these organisms produce dissolved organic matter with varied quantities or characteristics depending on the nutrient availability and the species composition. Therefore, this study aims to assess the characteristic of dissolved organic matter on surface water and its correlation with phytoplankton abundance for monitoring water quality.

**METHODS:** The sample was obtained at four Kali Surabaya river stations for further dissolved organic matter analysis and phytoplankton species analysis. The analysis was presented through bulk parameters of total organic, ultraviolet at 254 nm wavelength, specific ultraviolet absorbance value, and fluorescence spectroscopy using excitation-emission matrices with fluorescence regional integration analysis.

**FINDINGS:** The results showed the bulk parameters of dissolved organic matter at all stations were significantly different, as Station 1 and 2 were higher, while 3 and 4 had a lower concentration. Furthermore, the fluorescence spectroscopy identified four components of dissolved organic matter at all stations, namely aromatic proteins-like, humic acid-like, soluble microbial by-products-like, and fulvic acid-like, which is the unit of fluorescence spectra in arbitrary unit. Also, stations 1 and 2 were grouped in the high percentage fluorescence regional integration of humic substance (fulvic acid-like and humic acid-like), while 3 and 4 were classified in the high percentage fluorescence regional integration of non-humic substances (aromatic proteins-like and soluble microbial by-products-like).

**CONCLUSION:** The main phytoplankton species, namely Plectonema sp., Pinularia sp., Nitzschia sp., Navicula sp., had the highest abundance at Stations 1, 3, and 4, respectively. A strong correlation between dissolved organic matter analysis and phytoplankton abundance led to the usage of these methods for monitoring surface water quality.

**KEYWORDS:** *Correlation; Dissolved organic matter; Fluorescence spectroscopy; Phytoplankton.*

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<b>NUMBER OF REFERENCES</b> 55	<b>NUMBER OF FIGURES</b> 8	<b>NUMBER OF TABLES</b> 2
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**RUNNING TITLE:** Assessing of characteristic dissolved organic matter and its correlation.

### INTRODUCTION

Human, industrial and agricultural activities have significantly changed aquatic ecosystems due to high organic and inorganic wastewater discharge. This runoff has appeared in the eutrophication of rivers and tributary (Conley *et al.*, 2009; Bhattacharya and Osburn, 2017) causing blooming phytoplankton and consequently, and the environmental issues (Paerl *et al.*, 2008; Heisler *et al.*, 2008; Biggs, 2000). It is eminent that phytoplankton community dynamics (i.e., taxonomic composition, abundance, and biomass) regard the quantity of inorganic phosphorus and nitrogen in the aquatic surrounding (Cao *et al.*, 2016; Cuvin-Aralar *et al.*, 2004). Furthermore, the impact of the organic pollutants contributes to

the quantity or quality of dissolved organic matter in surface water. Allochthonous and autochthonous with effluent organic matter are the source of dissolved organic matter (DOM) in the surface water, since allochthonous could be generated from the upstream, midstream and the downstream. The upstream was found to be covered with perennial vegetation; the midstream is used for agriculture and covered with least forest; the downstream was mainly used for residential and utilized for different forms of agriculture (Dumago *et al.*, 2018). In addition, biogeochemical cycles will affect the quality and quantity of DOM from the surrounding environment. Also, DOM has an essential role in supporting phytoplankton abundance and growth in aquatic surroundings (Kissman *et al.*, 2017; Burpee *et al.*, 2016) due to its usage as an organic nutrient source. It can be used by these micro-organisms as a source of nitrogen, phosphorus, and carbon when inorganic phosphorus and nitrogen are unavailable (Burpee *et al.*, 2016). The primary producers were proposed as an important source that influences its composition in surface water (Biddanda and Benner, 1997). Conversely, DOM can be produced by phytoplankton (Thornton, 2014), with varied characteristics and quantity which are mostly dependent on nutrient availability (Myklestad, 1995), composition of phytoplankton type (Biddanda and Benner, 1997), and bacterial interaction (Ramanan *et al.*, 2016). According to previous studies, various types of DOM have been found and released by different taxonomic groups of phytoplankton (Fukuzaki *et al.*, 2014; Romera-Castillo *et al.*, 2010). Phytoplankton production, microbial metabolism, residue from microbial degradation after their death and other processes, release protein-like materials as one of DOM components (Liu *et al.*, 2019; Mangal *et al.*, 2016). The fluorescence spectroscopy fingerprints, identified the signals of protein-like and humic-like materials released from extracellular *Microcystis aeruginosa* (Ziegmann *et al.*, 2010). In addition, the DOM which is closely related to the phytoplankton community dynamics, mainly consist of humic-like and protein-like materials (Suksomjit *et al.*, 2009; Zhang *et al.*, 2014) and exhibits their blooming (Altman and Paerl, 2012; Hounshell *et al.*, 2017). The qualitative and quantitative methods for characterizing organic matter analysis have been implemented to clarify the types of DOM transformation through the treatment process or in source water and their following removal. For example, using the bulk parameters of dissolved organic carbon (DOC) concentration, UV/vis at 254 nm wavelength to measure the aromaticity degree of organic matter and specific ultraviolet absorbance (SUVA) (Edzwald *et al.*, 1985; Lai *et al.*, 2015; Hidayah *et al.*, 2017), high-performance size exclusion chromatography (HPSEC) with ultraviolet detector (UVD) or an on-line organic carbon detector (OCD) (Jiao *et al.*, 2014; Lai *et al.*, 2015), fluorescence spectroscopy as well as fluorescence excitation-emission matrices (FEEM) (Hidayah *et al.*, 2017; Ho *et al.*, 2019). These procedures have been previously applied in observing the contribution of phytoplankton degradation to DOM as chromophoric by using fluorescent spectroscopy (Zhang *et al.*, 2009), to characterize DOM excreted by phytoplankton (Chari *et al.*, 2013), and to reveal its relationship with the community (Liu *et al.*, 2021). The use of bulk parameters and fluorescent spectroscopy methods, simultaneously for characterizing organic matter considering the phytoplankton abundance, have been rarely observed. Therefore, resulting in poor implementation of optimal water quality control measures. Furthermore, using these techniques to characterize organic matter and its correlation with phytoplankton abundance for monitoring surface water quality seems to urgently need implementation. Hence, this study aims to assess the characteristic of dissolved organic matter on surface water, as well as its correlation with phytoplankton abundance using the bulk parameters and fluorescence spectroscopy to monitor surface water quality. This study was conducted in the Kali Surabaya River, Surabaya, Indonesia, in 2021.

## **MATERIALS AND METHODS**

### *Data collection*

This study used water from the Kali Surabaya River in Surabaya city, a surface water source for public supply. The position of station 1 to station 4 is as shown in Table 1 and Fig. 1. The sample was collected twice per week from January to March 2021, and the DOM analysis, as well as phytoplankton abundance was measured through the bulk parameters and fluorescence spectroscopy. The parameters include TOC, UV<sub>254</sub>, SUVA value, while fluorescence spectroscopy identified aromatic proteins-like (AP-like), humic acid-like (HA-like), soluble microbial products-like (SMPs-like), and fulvic acid-like (FA-like). As this study targeted on dissolved organic matter in source water, 0.45 m filter

paper was used to filter the collected source water (Millipore Corporation, USA) to eliminate suspended particles before analysis the parameters. Furthermore, the ultraviolet absorbance at 254 nm (UV<sub>254</sub>) and total organic carbon (TOC) concentration of the water was measured for common physicochemical characteristics based on Standard Methods procedures (APHA *et al.*, 2012).

Table 1: The study sampling location characteristics

No.	Sampling station	Coordinate	Climate	Environmental condition
1	Rolag Telu dam	7°26'40" S 112°27'25" E	- Tropical - Sunny weather - Temperature 29°C	- Downstream of the Brantas river - Stagnant water - No residential
2	Wringin Anom district	7°24'21" S 112°30'27" E	- Tropical - Sunny weather - Temperature 29°C	- Agricultural land - There are residential - There are domestic activities (bathing, washing, latrine)
3	Cangkir district	7°22'04" S 112°37'47" E	- Tropical - Sunny weather - Temperature 29°C	- Industrial area - Densely populated - Temporary dump site
4	Karang Pilang drinking water company inlet	7°20'54" S 112°40'51" E	- Tropical - Sunny weather - Temperature 29°C	- Industrial area - There are residential - There are domestic activities (bathing, washing, latrine)



Fig. 1: Geographic location of the study area in the Kali Surabaya River, Indonesia

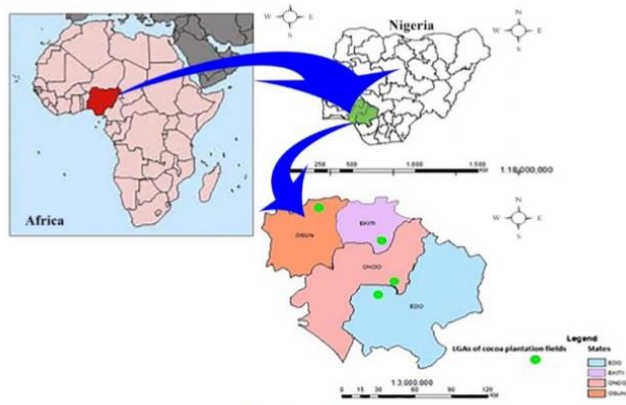
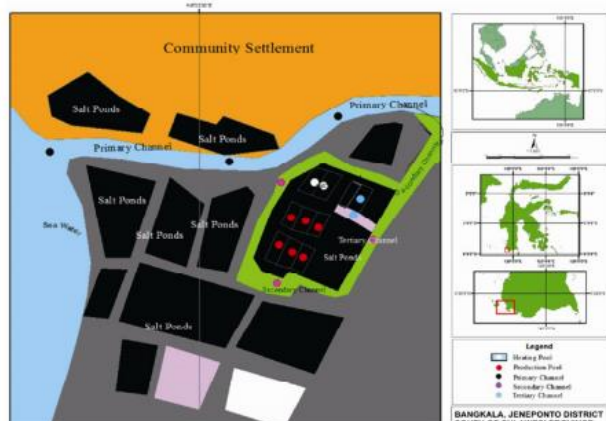
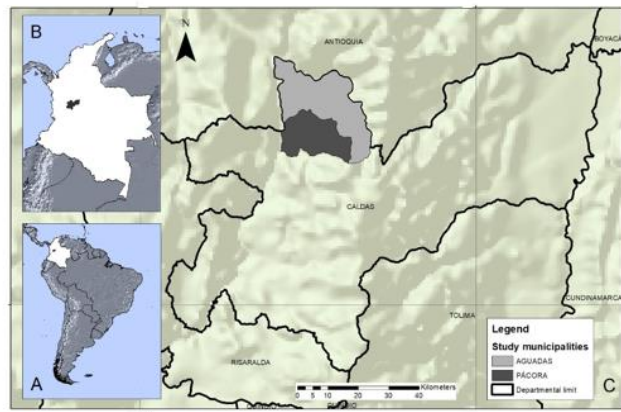
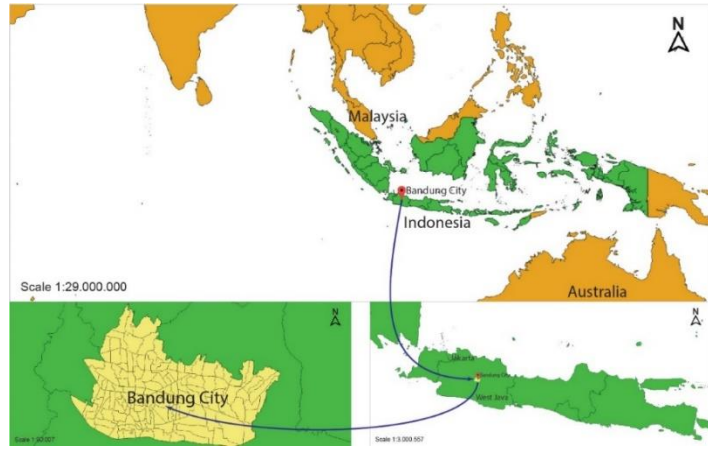


Fig. 1: Locations of the study area



4 Sample Maps



TOC was quantified using TOC-500 Model (Shimadzu, Japan), while  $UV_{254}$  was detected by UV/vis spectrophotometer Model U-2001 (Hitachi, Japan). The SUVA value showed the dissolved organics were contained in hydrophilic fraction as calculated from measurements of  $UV_{254}$  and DOC samples. Perkin Elmer LS-55 spectrometer with excitation-emission wavelength pair was used to measure the fluorescence in the source water. Moreover, the excitation-emission matrix (EEM) were resulted for each sample by skimming overexcitation (Ex) wavelengths between 230 and 400 nm at an interval of 10 nm with emission (Em) wavelengths between 300 and 547.5 nm at 0.5 nm interval (Murphy *et al.*, 2013; Hidayah *et al.*, 2017). Counting of fluorescence regional integration (FRI) analysis was used to provide the cumulative fluorescence reaction of organic matter with identical characteristic in selected regions by integration beneath EEMs (Chen *et al.*, 2003). The phytoplankton sampling was conducted using a plankton net mesh size 60  $\mu$ m as much as 100 liters. Meanwhile, its identification was carried out in the laboratory using a binocular microscope with 10 x 10 magnification (AmScope B100B-MS). Also, the abundance was calculated using Sedgewick-Rafter Counting Chamber for three replications (Marienfeld GmbH).

#### *Analytical framework*

The Kolmogorov-Smirnov, one-way Analysis of Variance (ANOVA), and Pearson correlations were applied utilizing SPSS Statistics 17.0 software (IBM, Armonk, NY, USA). The Kolmogorov-Smirnov test opposed the empirical cumulative distribution function of bulk parameters data and the results of FRI analysis with the distribution expected when the data were standard. When the observed difference is adequately significant, the test will reject the hypothesis of bulk parameters data, the results of FRI analysis data, and phytoplankton abundance normality. However, when the p-value of this test is less than 5%, it can be concluded that the bulk parameters data, the results of FRI analysis data, and phytoplankton abundance are non-normal. The one-way ANOVA was applied to determine whether any statistically significant differences between the means of bulk parameters and the results of FRI data. It was also used to determine at least two groups of the parameters data as the results of FRI analysis were different. In addition, The Pearson correlation coefficients measured the strength of the linear relationship variables among TOC,  $UV_{254}$ , SUVA value, AP-like, FA-like, SMPs-like, HA-like, and phytoplankton abundance.

## **RESULTS AND DISCUSSION**

### *The bulk parameters of dissolved organic matter in the river segment.*

The distribution data for the bulk parameters of dissolved organic matter in the river segment as tested by Kolmogorov–Smirnov showed the TOC concentration ( $P > 0.15$ ),  $UV_{254}$  concentration ( $P > 0.15$ ), and SUVA value ( $P > 0.15$ ) was normal. Furthermore, the normal distribution data was performed using ANOVA testing to know the differences in mean concentrations of TOC and  $UV_{254}$ , as well as SUVA value. ANOVA with the Tukey 95% confidence interval also determined whether there were statistically significant or non-significance differences. The results indicated statistically significant differences in the mean concentration of the bulk parameters among the river segment with a p-value of 0.011, 0.001, and 0.004 in TOC,  $UV_{254}$ , and SUVA values, respectively. Moreover, enough evidence was provided, which concluded that the average of the bulk organic matter parameters at all stations was significantly different. The Tukey analysis classified the bulk parameters concentration at each station into two main groups. Station 1 and 2 were grouped in the high concentration, while 3 and 4 were classified in the bulk parameters' low concentration, which means the former had averages significantly different from the latter. The average TOC concentration for stations 1 and 2 was about a value 10.1-11.7 mg/L, while 3 and 4 were between 9.8-10.9 mg/L. The average  $UV_{254}$  concentration for stations 1 and 2 was in the range of 10.1-11.7 mg/L, while 3 and 4 were in between 9.8-10.9 mg/L. The average  $UV_{254}$  concentration for stations 1 and 2 was in the range of 0.65-0.8/cm, while 3 and 4 were 0.39-0.65/cm. The average SUVA concentration of stations 1 and 2 was 5.3-6.4 L/mg/m, while 3 and 4 were 4.0-5.3 L/mg/m. Furthermore, statistical box plot analysis presented the pattern for the bulk parameters of dissolved organic matter in the surface water. Figs. 2, 3, and 4 show a box plot of the average concentration of TOC,  $UV_{254}$ , and average SUVA value respectively. Fig. 2 shows that the highest

average TOC concentration occurred at Station 2 with a varying range. In comparison, the lowest average TOC concentration with a low range occurred at station 4. In addition, the results showed the average concentration from the highest to the lowest was found at stations 2, 1, 3, and 4. The surface water used in this study contained 7.36 – 15.50 mg/L TOC concentration, which was typically associated with the DOC range. River water has a typical concentration about 2 to 10 mg/L of dissolved organic carbon, which was much higher than groundwater and seawater. Variation in average concentrations of TOC indicated various physical or ecological drivers, chemical processes, spatial changes, which can significantly affect on organic matters dynamics (Maie et al., 2006). The organic matter compositional changes could be induced by biophysical controls, such as changes in composition, which likely result in bioavailability, photoreactivity, nutrient cycling, or chelating capacity and can affect carbon fluxes consequentially ecological drivers not accounted for (Jaffe, 2008). In addition, the hydrology dynamics of surface runoff contributed to the surface water stream (Hood et al., 2006).

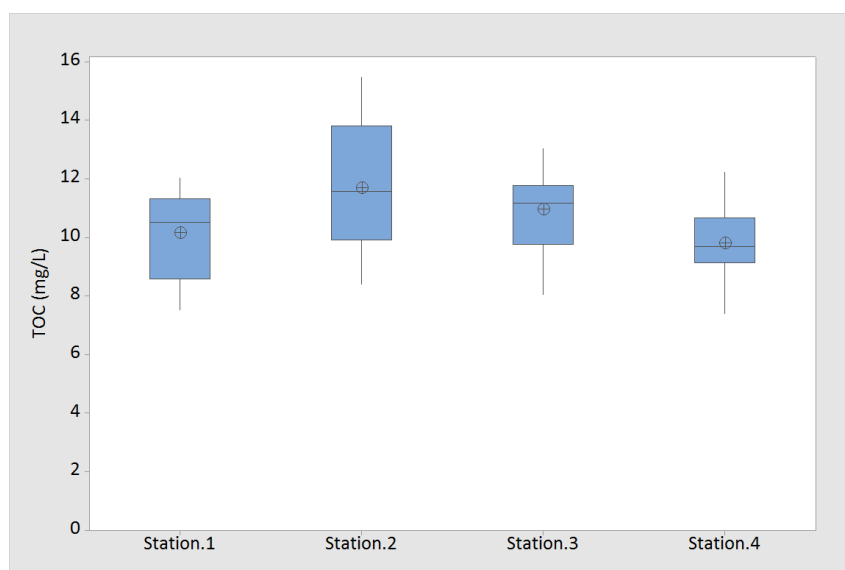


Fig. 2: The average TOC concentration in the river segment at various stations.

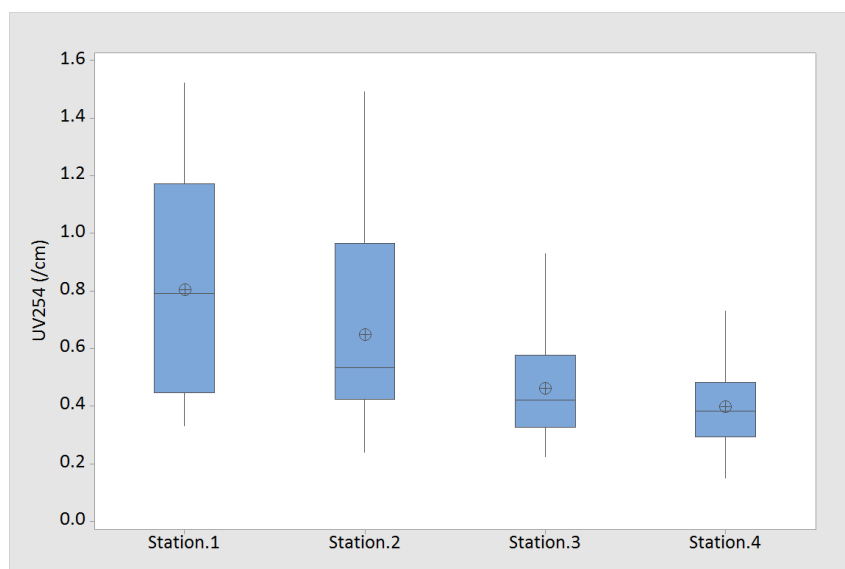


Fig. 3: The average UV<sub>254</sub> concentration in the river segment at various station

Fig. 3 describes the concentration of  $UV_{254}$ , which corresponded to the organic compounds with an aromatic structure, double bonds of C=C (Matilainen *et al.*, 2011). In this study, the concentration of  $UV_{254}$  for surface water was 0.148 – 1.524/cm, which was within the typical range of river (0.085 – 0.4/cm) (Edzwald *et al.*, 1985). The results showed that the average highest aromatic compound was detected at Station 1, while Station 4 had the average lowest concentration. Therefore, Station 1 contained higher humic matter with conjugated C=C double structural bonds than the others. Meanwhile, Station 4 contained lower humic matter than the others. As well known, organic compounds of humic matter contain unsaturated carbon bonds (double or triple) or aromatic rings in their molecular structure. Hence, it absorbs an amount of UV light through the water sample (Her *et al.*, 2002).

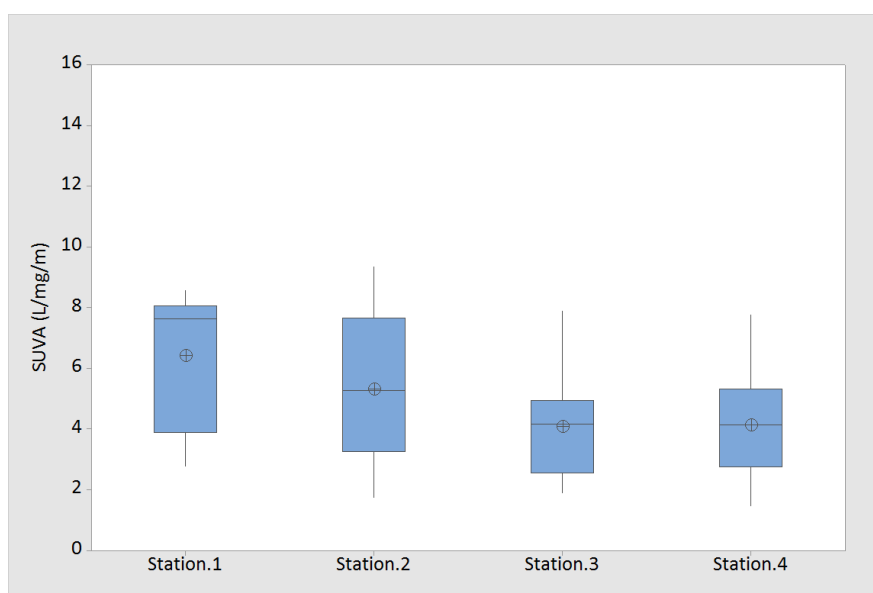


Fig. 4: The average SUVA value in the river segment at various station

Fig. 4. Shows the hydrophobicity of organic matter characteristic or specific UV-absorbance (SUVA) value. The results revealed a value between 1.45 – 9.36 L/mg/m. However, it was mostly higher than 4 along the river segment, which means that the organic matter is mainly consists of humic, hydrophobic, and high molar mass organic material. According to Edzwald and Tobiason (2011), SUVA is a parameter of the organic matter composition in water. Source water with SUVA values  $\geq 4$  indicated that natural organic matter composed mainly of humic or hydrophobic matter, while those  $< 2$  contained mainly non-humic or hydrophilic natural organic matter. The results were consistent with the high concentration of  $UV_{254}$  (0.148 – 1.524/cm). The values typically ranged from 1.0 to 6.0 L/mg/m for surface water. However, values greater than 6.0 were revealed for interstitial waters dominated by a solid terrestrial signature (Jaffe *et al.*, 2008). According to previous studies, these higher values can be as a result of the absorption at 254 nm from colloids, iron, or other components in the sample (Weishaar *et al.*, 2003; Hudson *et al.*, 2007). Combining the bulk parameters of TOC,  $UV_{254}$ , and SUVA value led to characterize the organic matter in the river. Station 2 was mainly composed of the highest TOC with lower aromatic and hydrophobic than 1, and vice versa. Also, station 4 was mainly composed of lower bulk parameters than 3. Therefore, 2 contained more aliphatic organic matter that does not absorb at 254 nm than the others. The lower SUVA value among all stations indicated the mixtures of aquatic humics, hydrophobic and hydrophilic, and molecular weights of organic matter.

*Characteristic of fluorescence dissolved organic matter in the river segment through volumetric fluorescence distribution.*

Fig. 5. Illustrates the fluorescence excitation-emission matrices (FEEMs) for dissolved organic matter in the river segment at a different station, taken on the first week sampling time. Dissolved organic carbon was classified into four regions based on its excitation/emission wavelengths (Ex/Em), namely Region 1 indicated the aromatic proteins-like (AP-like), such as tyrosine and tryptophan, at Ex/Em <250 nm/<350 nm. Region 2 identified the fulvic acid-like (FA-like) substances at Ex/Em <250 nm/>380 nm, Region 3 was corresponded to the soluble microbial by products-like (SMPs-like) substances at Ex/Em 250-280 nm /<380 nm, while Region 4 was identified as the humic acid-like (HA-like) substances with Ex/Em >280 nm/>380 nm (Chen *et al.*, 2003).

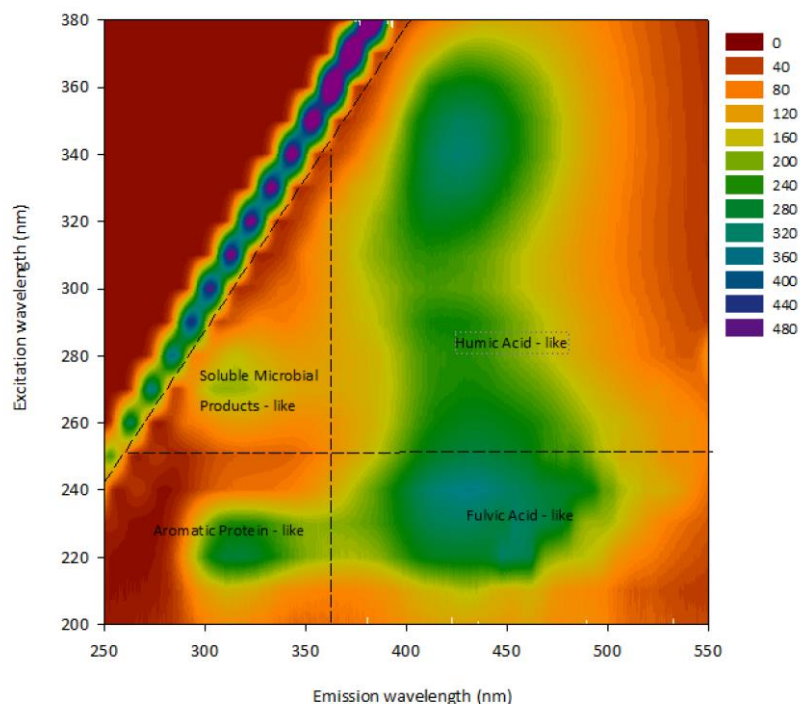


Fig. 5: Spectrum of fluorescence spectrometer analysis in the river segment

This study shows that the fluorescence component from FEEM analysis has consistent results with previous studies (Her *et al.*, 2003; Yao *et al.*, 2016; Moradi *et al.*, 2018; Hidayah *et al.*, 2020). Generally, HA-like and FA-like correlated with aromatic compounds. They mainly exist as carboxylic and phenolic functional groups in natural dissolved organic matter. These fluorescence structures are mostly present as a significant percentage of humic substances, which typically represent over 50% of natural organic matter (Shon *et al.*, 2012). In addition, source water may contain protein-like materials which microbial activities can generate. The amount, characteristics, and properties of dissolved organic matter in the aquatic system depend on their origin and environmental biochemical cycles. Sources of organic matter are classified as allochthonous (generated from a terrestrial watershed) and autochthonous (produced by organism activities, such as phytoplankton activities) (Chari *et al.*, 2013; Zhang *et al.*, 2009; Haraguchi *et al.*, 2019). Terrestrial watershed is mainly composed of humic substances such as fulvic and humic acids as well as humin, which are primarily hydrophobic and rich in aromatic carbon. The autochthonous source material is microbially derived organic, such as algal-derived and effluent organic matter (Kelso and Baker, 2020). Fig. 6 showed the percentage fluorescence response, which was calculated by Fluorescence Regional Integration (FRI) method. The percentage of fluorescence distribution indicated the four fractions quantity of fluorescence organic matter. This study classified the fraction into humic and non-humic substances-like. The first was represented by Region 2 (FA-like) and 4 (HA-like), while the second one by Region 1 (AP-like) and 3 (SMPs-like). Firstly, the results showed the highest total percentage of FRI in Region 2 and 4 was at Station 1 (76.6%), and the lowest total percentage for humic substances-like was at Station 4 (69.2%). Both components are classified as humic substances and are mainly composed of aromatic compounds with high to medium molecular weight (Watson *et al.*, 2018; Hua *et al.*, 2020). Their total percentage

FRI showed a consistent  $UV_{254}$  concentration and SUVA value. Furthermore, Station 1 had the highest bulk parameters, while 4 had the lowest. Secondly, the highest total percentage FRI of Region 1 and 3 (30.8%) was identified at Station 4, with the lowest at 1 (23.4%). This indicated that Station 4 contained abundant proteins substances and microbial-like fluorescence than the others and followed the lowest SUVA value of Station 4 with the highest for Station 1. Region 1 and 3 correlated with high molecular weight protein-like, which had chemical properties related to aromatic amino acids, tryptophan or tyrosine-like (Yamashita *et al.*, 2008; Hua *et al.*, 2020) and low molecular weight microbial humic-like as well as less conjugated double bond organic matter (Nguyen *et al.*, 2013; Hua *et al.*, 2020).

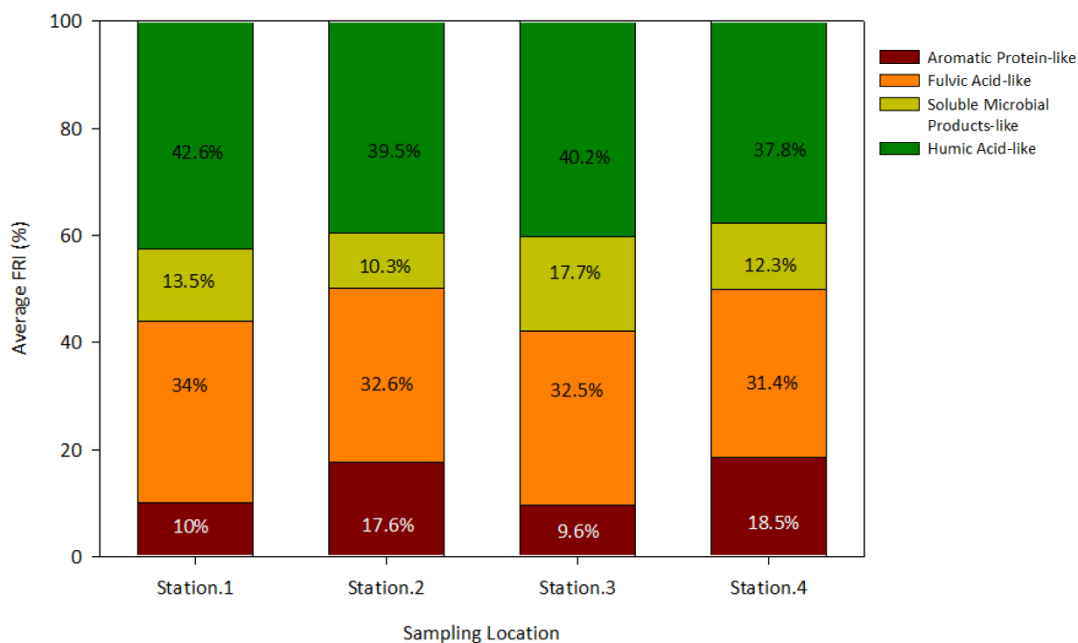


Fig. 6: FRI distribution of fractionated organic matter from the various river segment

The distribution data for the fluorescence of dissolved organic matter in the river segment was tested by Kolmogorov–Smirnov and the bulk parameters. The results showed distribution data for percentage FRI of Region 1 (AP-like), 2 (FA-like), 3 (SMPs-like), and Region 4 (HA-like) with  $P > 0.000$ ,  $0.007$ ,  $0.000$  and  $0.013$  respectively were normal. Furthermore, Analysis of Variance (ANOVA) testing was carried out to determine the differences in mean percentage FRI for each region. The statistical analysis ANOVA One-Way with the Tukey 95% confidence interval also determined whether statistically significant or non-significance differences in percentage FRI of AP-like, FA-like, SMPs-like, and HA-like among all stations. The results showed statistically significant differences in the mean percentage FRI of all fluorescence organic fractions at all stations with  $p = 0.000$ ,  $0.007$ ,  $0.000$ , and  $0.013$  in AP-like, FA-like, SMPs-like, HA-like, respectively. The results provided enough evidence to conclude that the mean percentage FRI of all fluorescence organic fractions at all stations was significantly different. Moreover, the Tukey analysis classified their percentage FRI at each station into two main groups. Station 1 and 2 were grouped in the high percentage FRI of humic substance-like (FA-like and HA-like), while 3 and 4 were classified in the low percentage. This means the former had an average percentage FRI of FA-like and HA-like, which were significantly different from the latter. In addition, stations 3 and 4 were grouped in the high percentage FRI of non-humic substance-like (AP-like and SMPs-like), while stations 3 and 4 were grouped in the low percentage. This showed both had average percentage FRI of AP-like and SMPs-like, which were significantly different from stations 1 and 2. Moreover, statistical box plot analysis presented the pattern of the fluorescence organic matter in the river segments. Fig. 7a to 7d presented box plot with average percentage FRI of the organic matter. Firstly, a comparison among all fluoresces organic compounds showed the average FRI of HA-like was much higher and much lower for SMPs-like than the others. However, HA-like, located at Region 4 of the fluorescence spectra, had the most extensive range of excitation and emission wavelengths. Therefore, the humic acid

substances-like region had the most extensive volume distribution of FRI when compared to others (Chen *et al.*, 2003). Meanwhile, SMPs-like or Region 3 comprised a dominant percentage of the fluorescence in wastewater treatment plant effluent (Chen *et al.*, 2003) and was closely related to the phytoplankton activities (Liu *et al.*, 2021; Hua *et al.*, 2020). Second, the average percentage FRI of the organic matter indicated different quantities and quality at each station. The non-humic substances-like fluorescence as presented by AP-like, SMPs-like, with statistical analysis, had a higher percentage FRI at stations 3 and 4 than the others. It was likely that Station 3 and 4 had a higher percentage of extracellular biological organic matter fraction than the other river segments. The fraction was supposed to contain soluble microbial products of amino acids and carbohydrates. Tryptophan and tyrosine which are aromatic amino acids, were confirmed as biological activity products in natural systems and exhibited a distribution of fluorescence response similar to AP-like and SMPs-like of this study (Coble 2007; Determann *et al.*, 1998). The humic substances-like fluorescence as presented by FA-like and HA-like had a higher percentage FRI at stations 1 and 2 than others and were tested by ANOVA One-Way. Combining the bulk parameters of TOC,  $UV_{254}$ , SUVA value, and fluorescence spectroscopy convinced the characteristic of organic matter in the river. Station 1 and 2 had high  $UV_{254}$  concentration, SUVA value, high percentage FRI of FA-like and HA-like substances. It was conjectured that stations 1 and 2 were mainly composed of aromatic, hydrophobic, humic substances organic matter, which may be generated from terrestrial systems.

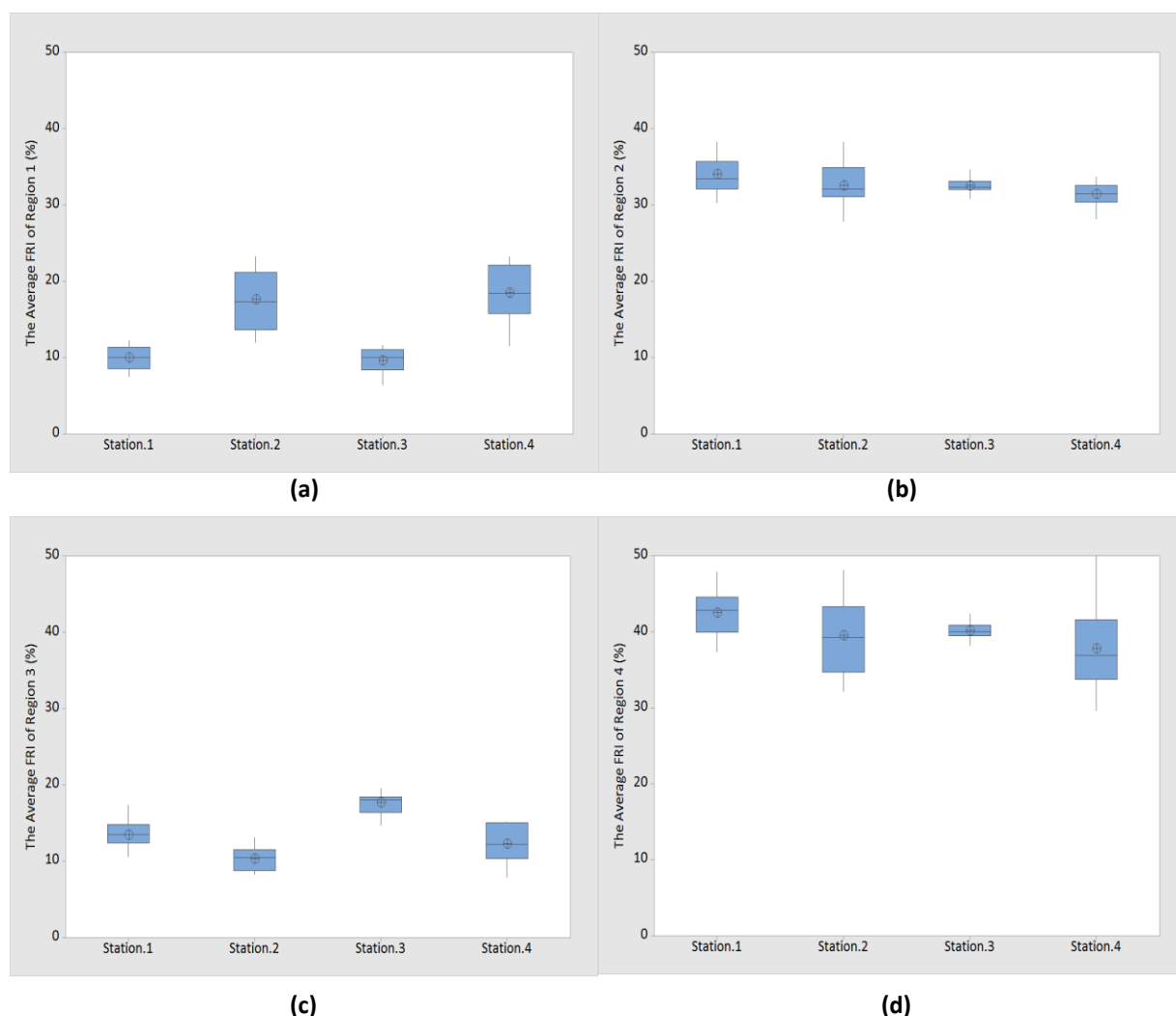


Fig. 7: The average percentage FRI of fluorescence organic matter in the river segment at various stations

Station 3 and 4 had lower UV concentration and SUVA values, with a high percentage FRI of AP-like and SMPs-like than the others. There was a lower SUVA value among all stations indicates in the mixtures of aquatic humics, hydrophobic and hydrophilic, as well as molecular weights of organic

matter. This showed that Station 3 and 4 comprised more autochthonous and sources of organic matter from anthropogenic activities. The river ecosystem, which source is terrestrial, autochthonous, and anthropogenic, provided hotspots for storing, transporting, and transforming organic matter. The sources proportions were primarily and terrestrially derived with increased autochthonous inputs from macrophytes. In addition, the sources of dissolved organic matter are a mixture of terrestrial, autochthonous, or primarily from wastewater effluent (Kelso and Baker, 2020).

*Contribution of phytoplankton abundance to fluorescence dissolved organic matter in the river segment.*

This study discovered four main phytoplankton species with various abundance in the river segments, namely *Plectonema* sp., *Nitzschia* sp., *Navicula* sp., and *Pinularia* sp. The distribution data of the phytoplankton abundance in this segment was tested by Kolmogorov–Smirnov. The results showed a usual distribution data for *Plectonema* sp., *Nitzschia* sp., *Navicular* sp., and *Pinularia* sp. as abundance  $P > 0.000$ , 0.007, 0.000, and 0.013, respectively. Furthermore, ANOVA testing was carried out to determine the differences in the mean phytoplankton abundance of the river segments. The statistical analysis ANOVA One-Way with the Tukey 95% confidence interval determined whether there were statistically significant or non-significance differences in the abundance of the species among all stations. According to the results, there were statistically significant differences in the mean abundance of phytoplankton at all stations with p-value = 0.006 and 0.01 in *Plectonema* sp. and *Nitzschia* sp. abundance, respectively. Meanwhile, the analysis generated p-value = 0.156 and 0.412 for *Navicula* sp. and *Pinularia* sp. abundance, respectively, therefore, classified as only one group of phytoplankton abundance. This showed that there were non-significantly differences in both species abundance among all stations. The Tukey analysis classified *Plectonema* sp. and *Nitzschia* sp. abundance at each station into two main groups. Station 1 and 2 were grouped in the high *Plectonema* sp. and low *Nitzschia* sp. abundance, while Station 3 and 4 were classified in the low *Plectonema* sp. and high *Nitzschia* sp. abundance. Furthermore, the statistical box plot analysis presented the pattern of the phytoplankton abundance in the river segments.

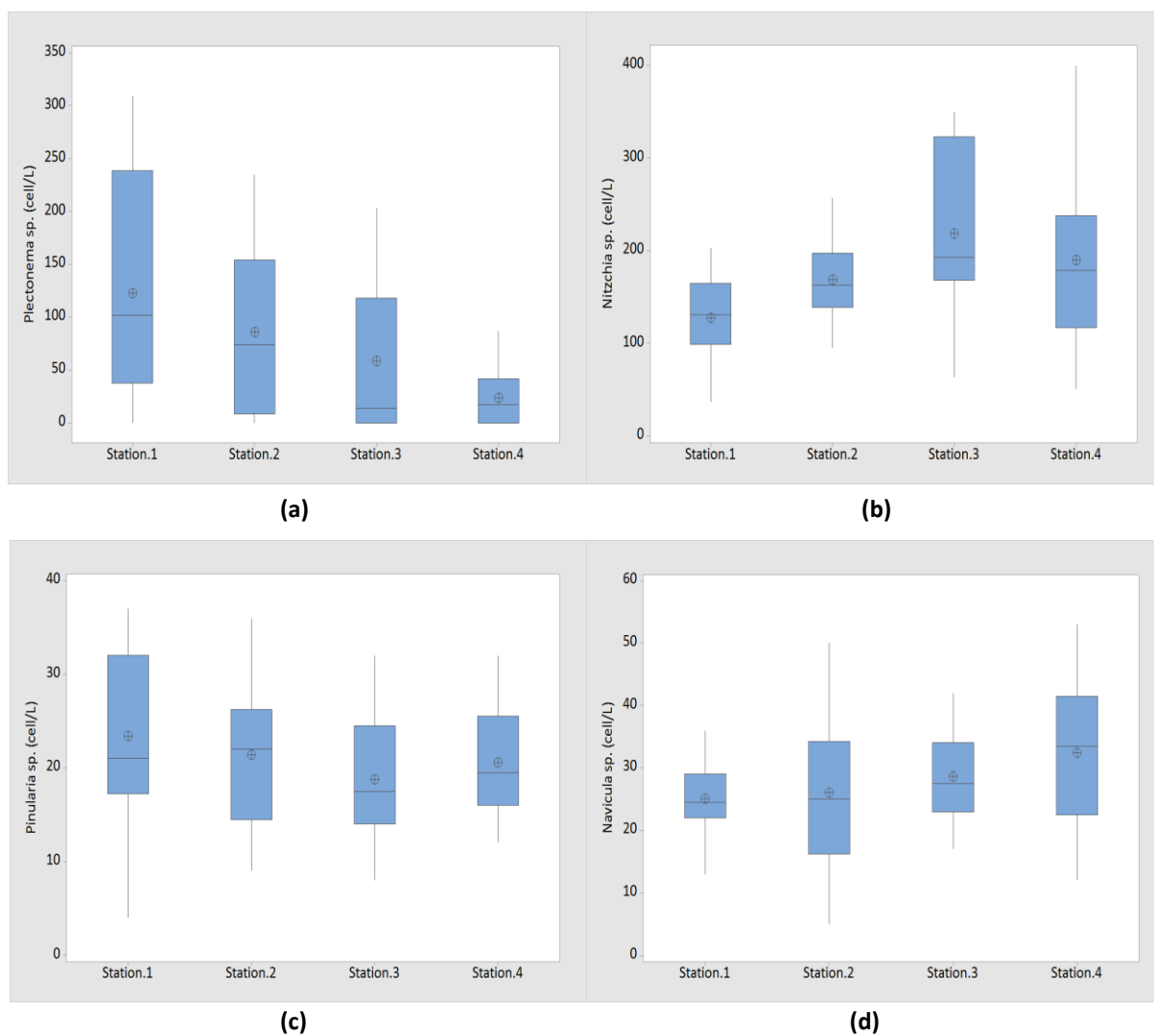


Fig. 8: The average of phytoplankton abundance in the river segment at various station

Fig. 8a to 8d present box plots of their average abundance. Firstly, a comparison among the species at all stations conjectured that *Nitzschia* sp. had a higher abundance, and *Pinularia* sp. was lower than the others. Meanwhile, *Plectonema* sp. had the highest at Station 1 and the lowest at 4. *Nitzschia* sp. had a higher abundance at Station 3 and lower at 1. Moreover, *Navicula* sp. had the highest abundance at Station 4 and the lowest at Station 1. *Pinnularia* sp. gave the highest at Station 1, with the lowest at Station 3. This phytoplankton abundance was strongly influenced by migration, which can occur due to population density and physical environmental conditions, such as changes in temperature and currents (Basu and Mackey, 2018). Secondly, Station 1 was likely to contain a similar abundance in *Plectonema* sp. and *Nitzschia* sp., and the same for *Navicula* sp. and *Pinularia* sp. Stations 2 and 3 showed that the abundance of *Nitzschia* sp. was primarily dominant than others. However, *Navicular* sp. was similar to *Pinularia* sp. Station 4 identified a similar abundance of *Plectonema* sp., *Navicular* sp., and *Pinularia* sp. There is competition in several phytoplankton species that use the same resource lacking in availability, or even regardless of sufficient availability, and competition still occurs when they take advantage of the resource, with one attacking the other or vice versa (Burson *et al.*, 2018).

*The relationship among the bulk parameters, organic fluorescence parameters, and phytoplankton abundance*

The degree correlation between the bulk parameters, fluorescence organic matter, and phytoplankton abundance was examined, as shown in Table 2. Correlation analysis was carried out using TOC and UV<sub>254</sub> concentrations, SUVA value with percentage FRI of AP-like, FA-like, SMPs-like, or HA-like, as well



as the abundance of *Plectonema* sp., *Nitzchia* sp., *Navicula* sp., and *Pinularia* sp. Firstly, based on the correlations of the bulk parameters, TOC concentration was positively higher with Region 1 (AP-like) and Region 2 (FA-like). In addition, UV<sub>254</sub> concentration and SUVA value were significantly correlated with Region 1 (AP-like) and Region 4 (HA-like). The results showed fluorescence spectroscopy, which fractionated AP-like, FA-like, SMPs-like, and HA-like could be used to identify the quantity and quality of organic matter in the source water.

Table 2: The degree correlation among the bulk parameters, fluorescence organic matter, and phytoplankton abundance

Parameters	TOC	UV <sub>254</sub>	SUVA	AP-like	FA-like	SMPs-like	HA-like	Navicula sp.	Plectone ma sp.	Pinnularia sp.
UV <sub>254</sub>	0.085 0.502									
SUVA	-0.044 0.729	0.887 0.000								
AP-like	-0.287 0.022	-0.440 0.000	-0.373 0.002							
FA-like	0.254 0.042	0.105 0.411	0.047 0.710	-0.249 0.048						
SMPs-like	-0.038 0.764	-0.198 0.116	-0.228 0.070	0.638 0.000	-0.085 0.505					
HA-like	-0.035 0.786	0.344 0.005	0.344 0.005	-0.674 0.000	-0.022 0.862	-0.348 0.005				
Navicula sp.	-0.109 0.392	-0.331 0.007	-0.289 0.021	0.193 0.126	-0.102 0.422	0.090 0.480	-0.082 0.521			
Plectonema sp.	0.271 0.030	0.137 0.281	0.131 0.303	-0.346 0.005	0.293 0.019	-0.057 0.652	0.110 0.386	0.166 0.189		
Pinnularia sp.	-0.097 0.448	-0.239 0.058	-0.292 0.470	-0.245 0.051	0.142 0.263	-0.268 0.032	0.220 0.080	0.137 0.279	0.320 0.010	
Nitzchia sp.	-0.243 0.053	-0.283 0.023	-0.203 0.108	0.160 0.205	-0.203 0.107	0.176 0.164	0.070 0.585	0.168 0.184	0.035 0.785	0.174 0.170

Cell Contents description; Pearson correlation (the first row of the number of correlation between parameters); P-value (the second row of the number of correlation between parameters)

This result was expected since TOC measured all organic carbon, including humic and non-humic substances, as presented by AP-like and FA-like. Secondly, a strong positive correlation between UV<sub>254</sub> concentration and SUVA value indicated that higher aromatic conjugated double bond corresponded to higher molecular weight organic, more hydrophobic, and content of humic substances. These results are consistent with the Pearson correlation between bulk parameters of UV<sub>254</sub> correlation, SUVA, and fluorescence organic matters of AP-like and HA-like. Furthermore, it was conjectured that fluorescence spectroscopy could be used to assess the properties of organic matter existing in the source water. Thirdly, the results showed that TOC had a stronger correlation with AP-like than HA-like. This was probably because the humic structure may incorporate protein-like-fluorophores due to weak interactions based on x-x or van der Waals forces between the dissolved organic matter components. Previous studies indicated that proteins and humic supramolecules containing specific structures attained from phenol or aniline might contribute to the fluorescence. Fourth, this study discovered a strong correlation between DOM and phytoplankton abundance. *Plectonema* sp. correlated with TOC, AP-like, and FA-like, while *Navicula* sp. and *Nitzchia* sp. correlated with UV<sub>254</sub>, and *Pinularia* sp. with SMPs-like. The existence of phytoplankton was likely to enhance the quantity and characteristics of DOM in the aquatic environment. The production of marine-like fluorophores accompanied phytoplankton degradation as a significant source of autochthonous DOM (Wada *et al.*, 2007). In addition, higher molecular weight compounds such as protein (tryptophan)-like fluorescence were presented in exudates when phytoplankton grows (Chari *et al.*, 2013). The combination of the bulk parameters (TOC, UV<sub>254</sub>, and SUVA value), fluorescence spectroscopy, and phytoplankton abundance

convinced the quality of organic matter in the surface water. However, it could be eventually used to monitor the water's quality.

## CONCLUSION

This study showed that the quality and quantity of DOM at all stations were significantly different, as classified into two groups with higher bulk parameters at stations 1 and 2 and a lower concentration at 3 and 4. The average TOC concentration for stations 1 and 2 was about a value 10.1-11.7 mg/L, while 3 and 4 were in between 9.8-10.9 mg/L. The average UV<sub>254</sub> concentration for stations 1 and 2 was in the range of 10.1-11.7 mg/L, while 3 and 4 were between 9.8-10.9 mg/L. The average UV<sub>254</sub> concentration for stations 1 and 2 was 0.65-0.8/cm, while 3 and 4 were 0.39-0.65/cm. The average SUVA concentration of stations 1 and 2 was in the range 5.3-6.4 L/mg/m, while 3 and 4 were 4.0-5.3 L/mg/m. In addition, fluorescence spectroscopy with FRI analysis showed stations 1 and 2 were grouped in the high percentage FRI of humic substance-like (FA-like and HA-like) about 74.35%. It was conjectured that stations 1 and 2 were mainly composed of aromatic, hydrophobic, humic substances organic matter, which may be generated from terrestrial systems, while stations 3 and 4 were classified in high percentages non-humic substances-like (AP-like and SMPs-like) about 29.05%. This showed that Station 3 and 4 comprised more autochthonous and sources of organic matter from anthropogenic activities. According to phytoplankton abundance, Station 1 had a high abundance of Plectonema sp. (238.5 cell/L) and Pinularia sp. (32 cell/L), while stations 2 and 3 mainly consisted of Nitzschia sp. (197.5 cell/L and 322.75 cell/L), and Navicula sp. (41.5 cell/L) was dominant at Station 4. The Pearson correlation showed a strong relationship between DOM and phytoplankton abundance. Therefore, Plectonema sp. was in correlation with TOC (0.271), AP-like (-0.346), and FA-like (0.293), while Navicula sp. and Nitzschia sp. correlated with UV<sub>254</sub> (-0.331 and -0.283), and Pinularia sp. correlated with SMPs-like (-0.268). This study conjectured that the bulk parameters of DOM, fluorescence spectroscopy, and phytoplankton abundance could be used to assess the characteristic of DOM, while the combination of these methods could be used to monitor the surface water quality. Future work should be conducted on the laboratory scale for phytoplankton observation in order to identify the characteristic of organic matter that a kind of phytoplankton species has released. Therefore, it could be used to predict the amount of DOM derived by phytoplankton, DOM derived in the aquatic, and DOM from the terrestrial watershed.

## AUTHOR CONTRIBUTIONS

Okik Hendriyanto Cahyonugroho performed the experimental design, analyzed the data, and prepared the manuscript text as well as the literature review. Sucipto Hariyanto and Ganden Supriyanto interpreted the data and helped in manuscript preparation.

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## CONFLICT OF INTEREST

The authors declare no potential conflict of interest regarding the publication of this work. Also, ethical issues, including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and submission, as well as redundancy, have been entirely witnessed by the authors.

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## ABBREVIATIONS

%	Percent
/cm	Per centimeter
ANOVA	Analysis of variance
AP-like	Aromatic proteins-like
C=C	Carbon chain double bonds
cell/L	The number of phytoplankton cell per liter
DOC	Dissolved organic carbon
DOM	Dissolved organic matter
Em	Emission wavelength
Ex	Excitation wavelength
FA-like	Fulvic acid-like
FEEM	Fluorescence spectroscopy using excitation-emission matrices
FRI	Fluorescence regional integration
HA-like	Humic acid-like
L/mg/m	Liter per miligrams per meter
mg/L	Miligrams per liter
mm	Milimeter
μm	Micrometer
<i>Navicula sp.</i>	Navicula species
<i>Nitzchia sp.</i>	Nitzchia species
NOM	Natural organic matter
nm	Nanometer
OCD	Organic carbon detector
P >	Probability value more than
P =	Probability value equal
<i>Pinnularia sp.</i>	Pinnularia species
<i>Plectonema sp.</i>	Plectonema species
P-value	Probability value
SMPs-like	Soluble microbial by products-like
SUVA	Specific ultraviolet absorbance
TOC	Total organic carbon
UV <sub>254</sub>	Ultraviolet at 254 nm wavelength
UVD	Ultraviolet detector
UV/vis	Ultraviolet visible

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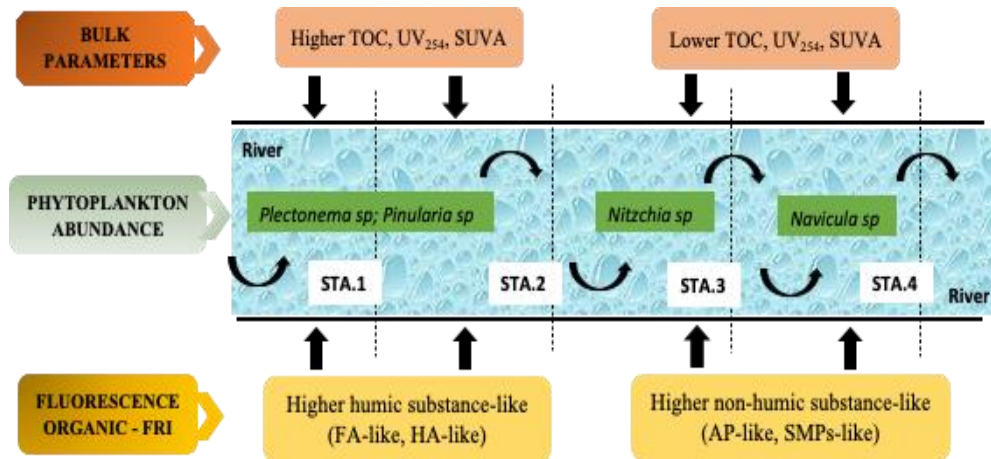
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## GRAPHICAL ABSTRACT



## HIGHLIGHTS

- Characteristic of organic matter and the phytoplankton abundance is different at each segment of surface water;
- There is a substantial correlation among the bulk parameters, fluorescence spectroscopy of DOM and phytoplankton abundance;
- Phytoplankton abundance combined with DOM analysis could be used to evaluate the quantity and quality of organic matter for monitoring surface water quality.

## Dissolved organic matter and its correlation with phytoplankton abundance for monitoring surface water quality

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### ABSTRACT:

**BACKGROUND AND OBJECTIVES:** Dissolved organic matter has a fundamental role in supporting phytoplankton abundance and growth in aquatic environments. However, these organisms produce dissolved organic matter with varied quantities or characteristics depending on the nutrient availability and the species composition. Therefore, this study aims to assess the characteristic of dissolved organic matter on surface water and its correlation with phytoplankton abundance for monitoring water quality.

**METHODS:** The sample was obtained at four Kali Surabaya river stations for further dissolved organic matter analysis and phytoplankton species analysis. The analysis was presented through bulk parameters of total organic, ultraviolet at 254 nm wavelength, specific ultraviolet absorbance value, and fluorescence spectroscopy using excitation-emission matrices with fluorescence regional integration analysis.

**FINDINGS:** The results showed the bulk parameters of dissolved organic matter at all stations were significantly different, as Station 1 and 2 were higher, while 3 and 4 had a lower concentration. Furthermore, the fluorescence spectroscopy identified four components of dissolved organic matter at all stations, namely aromatic proteins-like, humic acid-like, soluble microbial by-products-like, and fulvic acid-like, which is the unit of fluorescence spectra in arbitrary unit. Also, stations 1 and 2 were grouped in the high percentage fluorescence regional integration of humic substance (fulvic acid-like and humic acid-like), while 3 and 4 were classified in the high percentage fluorescence regional integration of non-humic substances (aromatic proteins-like and soluble microbial by-products-like).

**CONCLUSION:** The main phytoplankton species, namely *Plectonema* sp., *Pinularia* sp., *Nitzschia* sp., *Navicula* sp., had the highest abundance at Stations 1, 3, and 4, respectively. A strong correlation between dissolved organic matter analysis and phytoplankton abundance led to the usage of these methods for monitoring surface water quality.

**KEYWORDS:** *Correlation; Dissolved organic matter; Fluorescence spectroscopy; Phytoplankton.*

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<b>NUMBER OF REFERENCES</b>	<b>NUMBER OF FIGURES</b>	<b>NUMBER OF TABLES</b>
<b>55</b>	<b>8</b>	<b>2</b>

**RUNNING TITLE:** Assessing of characteristic dissolved organic matter and its correlation.

## INTRODUCTION

Human, industrial and agricultural activities have significantly changed aquatic ecosystems due to high organic and inorganic wastewater discharge. This runoff has appeared in the eutrophication of rivers and tributary (Conley *et al.*, 2009; Bhattacharya and Osburn, 2017) causing blooming phytoplankton and consequently, and the environmental issues (Paerl *et al.*, 2008; Heisler *et al.*, 2008; Biggs, 2000). It is eminent that phytoplankton community dynamics (i.e., taxonomic composition, abundance, and biomass) regard the quantity of inorganic phosphorus and nitrogen in the aquatic surrounding (Cao *et al.*, 2016; Cuvin-Aralar *et al.*, 2004). Furthermore, the impact of the organic pollutants contributes to the quantity or quality of dissolved organic matter in surface water. Allochthonous and autochthonous with effluent organic matter are the source of dissolved organic matter (DOM) in the surface water, since allochthonous could be generated from the upstream, midstream and the downstream. The upstream was found to be covered with perennial vegetation; the midstream is used for agriculture and covered with least forest; the downstream was mainly used for residential and utilized for different forms of agriculture (Dumago *et al.*, 2018). In addition, biogeochemical cycles will affect the quality and quantity of DOM from the surrounding environment. Also, DOM has an essential role in supporting phytoplankton abundance and growth in aquatic surroundings (Kissman *et al.*, 2017; Burpee *et al.*, 2016) due to its usage as an organic nutrient source. It can be used by these micro-organisms as a source of nitrogen, phosphorus, and carbon when inorganic phosphorus and nitrogen are unavailable (Burpee *et al.*, 2016). The primary producers were proposed as an important source that influences its composition in surface water (Biddanda and Benner, 1997). Conversely, DOM can be produced by phytoplankton (Thornton, 2014), with varied characteristics and quantity which are mostly dependent on nutrient availability (Myklestad, 1995), composition of phytoplankton type (Biddanda and Benner, 1997), and bacterial interaction (Ramanan *et al.*, 2016). According to previous studies, various types of DOM have been found and released by different taxonomic groups of phytoplankton (Fukuzaki *et al.*, 2014; Romera-Castillo *et al.*, 2010). Phytoplankton production, microbial metabolism, residue from microbial degradation after their death and other processes, release protein-like materials as one of DOM components (Liu *et al.*, 2019; Mangal *et al.*, 2016). The fluorescence spectroscopy fingerprints, identified the signals of protein-like and humic-like materials released from extracellular *Microcystis aeruginosa* (Ziegmann *et al.*, 2010). In addition, the DOM which is closely related to the phytoplankton community dynamics, mainly consist of humic-like and protein-like materials (Suksomjit *et al.*, 2009; Zhang *et al.*, 2014) and exhibits their blooming (Altman and Paerl, 2012; Hounshell *et al.*, 2017). The qualitative and quantitative methods for characterizing organic matter analysis have been implemented to clarify the types of DOM transformation through the treatment process or in source water and their following removal. For example, using the bulk parameters of dissolved organic carbon (DOC) concentration, UV/vis at 254 nm wavelength to measure the aromaticity degree of organic matter and specific ultraviolet absorbance (SUVA) (Edzwald *et al.*, 1985; Lai *et al.*, 2015; Hidayah *et al.*, 2017), high-performance size exclusion chromatography (HPSEC) with ultraviolet detector (UVD) or an on-line organic carbon detector (OCD) (Jiao *et al.*, 2014; Lai *et al.*, 2015), fluorescence spectroscopy as well as fluorescence excitation-emission matrices (FEEM) (Hidayah *et al.*, 2017; Ho *et al.*, 2019). These procedures have been previously applied in observing the contribution of phytoplankton degradation to DOM as chromophoric by using fluorescent spectroscopy (Zhang *et al.*, 2009), to characterize DOM excreted by phytoplankton (Chari *et al.*, 2013), and to reveal its relationship with the community (Liu *et al.*, 2021). The use of bulk parameters and fluorescent spectroscopy methods, simultaneously for

characterizing organic matter considering the phytoplankton abundance, have been rarely observed. Therefore, resulting in poor implementation of optimal water quality control measures. Furthermore, using these techniques to characterize organic matter and its correlation with phytoplankton abundance for monitoring surface water quality seems to urgently need implementation. Hence, this study aims to assess the characteristic of dissolved organic matter on surface water, as well as its correlation with phytoplankton abundance using the bulk parameters and fluorescence spectroscopy to monitor surface water quality. This study was conducted in the Kali Surabaya River, Surabaya, Indonesia, in 2021.

## MATERIALS AND METHODS

### *Data collection*

This study used water from the Kali Surabaya River in Surabaya city, a surface water source for public supply. The position of station 1 to station 4 is as shown in [Table 1](#) and [Fig. 1](#). The sample was collected twice per week from January to March 2021, and the DOM analysis, as well as phytoplankton abundance was measured through the bulk parameters and fluorescence spectroscopy. The parameters include TOC,  $UV_{254}$ , SUVA value, while fluorescence spectroscopy identified aromatic proteins-like (AP-like), humic acid-like (HA-like), soluble microbial products-like (SMPs-like), and fulvic acid-like (FA-like). As this study targeted on dissolved organic matter in source water, 0.45 m filter paper was used to filter the collected source water (Millipore Corporation, USA) to eliminate suspended particles before analysis the parameters. Furthermore, the ultraviolet absorbance at 254 nm ( $UV_{254}$ ) and total organic carbon (TOC) concentration of the water was measured for common physicochemical characteristics based on Standard Methods procedures ([APHA et al., 2012](#)).

Table 1: The study sampling location characteristics

No.	Sampling station	Coordinate	Climate	Environmental condition
1	Rolag Telu dam	7°26'40" S 112°27'25" E	- Tropical - Sunny weather - Temperature 29°C	- Downstream of the Brantas river - Stagnant water - No residential
2	Wringin Anom district	7°24'21" S 112°30'27" E	- Tropical - Sunny weather - Temperature 29°C	- Agricultural land - There are residential - There are domestic activities (bathing, washing, latrine)
3	Cangkir district	7°22'04" S 112°37'47" E	- Tropical - Sunny weather - Temperature 29°C	- Industrial area - Densely populated - Temporary dump site
4	Karang Pilang drinking water company inlet	7°20'54" S 112°40'51" E	- Tropical - Sunny weather - Temperature 29°C	- Industrial area - There are residential - There are domestic activities (bathing, washing, latrine)



Fig. 1: Geographic location of the study area in the Kali Surabaya River, Indonesia

TOC was quantified using TOC-500 Model (Shimadzu, Japan), while  $UV_{254}$  was detected by UV/vis spectrophotometer Model U-2001 (Hitachi, Japan). The SUVA value showed the dissolved organics were contained in hydrophilic fraction as calculated from measurements of  $UV_{254}$  and DOC samples. Perkin Elmer LS-55 spectrometer with excitation-emission wavelength pair was used to measure the fluorescence in the source water. Moreover, the excitation-emission matrix (EEM) were resulted for each sample by skimming overexcitation (Ex) wavelengths between 230 and 400 nm at an interval of 10 nm with emission (Em) wavelengths between 300 and 547.5 nm at 0.5 nm interval (Murphy *et al.*, 2013; Hidayah *et al.*, 2017). Counting of fluorescence regional integration (FRI) analysis was used to provide the cumulative fluorescence reaction of organic matter with identical characteristic in selected regions by integration beneath EEMs (Chen *et al.*, 2003). The phytoplankton sampling was conducted using a plankton net mesh size 60 mm as much as 100 liters. Meanwhile, its identification was carried out in the laboratory using a binocular microscope with 10 x 10 magnification (AmScope B100B-MS). Also, the abundance was calculated using Sedgewick-Rafter Counting Chamber for three replications (Marienfeld GmbH).

#### *Analytical framework*

The Kolmogorov-Smirnov, one-way Analysis of Variance (ANOVA), and Pearson correlations were applied utilizing SPSS Statistics 17.0 software (IBM, Armonk, NY, USA). The Kolmogorov-Smirnov test opposed the empirical cumulative distribution function of bulk parameters data and the results of FRI analysis with the distribution expected when the data were standard. When the observed difference is adequately significant, the test will reject the hypothesis of bulk parameters data, the results of FRI analysis data, and phytoplankton abundance normality. However, when the p-value of this test is less than 5%, it can be concluded that the bulk parameters data, the results of FRI analysis data, and phytoplankton abundance are non-normal. The one-way ANOVA was applied to determine whether any statistically significant differences between the means of bulk parameters and the results of FRI data. It was also used to determine at least two groups of the parameters data as the results of FRI analysis were different. In addition, The Pearson correlation coefficients measured the strength of the

linear relationship variables among TOC, UV<sub>254</sub>, SUVA value, AP-like, FA-like, SMPs-like, HA-like, and phytoplankton abundance.

## RESULTS AND DISCUSSION

### *The bulk parameters of dissolved organic matter in the river segment.*

The distribution data for the bulk parameters of dissolved organic matter in the river segment as tested by Kolmogorov–Smirnov showed the TOC concentration ( $P > 0.15$ ), UV<sub>254</sub> concentration ( $P > 0.15$ ), and SUVA value ( $P > 0.15$ ) was normal. Furthermore, the normal distribution data was performed using ANOVA testing to know the differences in mean concentrations of TOC and UV<sub>254</sub>, as well as SUVA value. ANOVA with the Tukey 95% confidence interval also determined whether there were statistically significant or non-significance differences. The results indicated statistically significant differences in the mean concentration of the bulk parameters among the river segment with a p-value of 0.011, 0.001, and 0.004 in TOC, UV<sub>254</sub>, and SUVA values, respectively. Moreover, enough evidence was provided, which concluded that the average of the bulk organic matter parameters at all stations was significantly different. The Tukey analysis classified the bulk parameters concentration at each station into two main groups. Station 1 and 2 were grouped in the high concentration, while 3 and 4 were classified in the bulk parameters' low concentration, which means the former had averages significantly different from the latter. The average TOC concentration for stations 1 and 2 was about a value 10.1-11.7 mg/L, while 3 and 4 were between 9.8-10.9 mg/L. The average UV<sub>254</sub> concentration for stations 1 and 2 was in the range of 10.1-11.7 mg/L, while 3 and 4 were in between 9.8-10.9 mg/L. The average UV<sub>254</sub> concentration for stations 1 and 2 was in the range of 0.65-0.8/cm, while 3 and 4 were 0.39-0.65/cm. The average SUVA concentration of stations 1 and 2 was 5.3-6.4 L/mg/m, while 3 and 4 were 4.0-5.3 L/mg/m. Furthermore, statistical box plot analysis presented the pattern for the bulk parameters of dissolved organic matter in the surface water. Figs. 2, 3, and 4 show a box plot of the average concentration of TOC, UV<sub>254</sub>, and average SUVA value respectively. Fig. 2 shows that the highest average TOC concentration occurred at Station 2 with a varying range. In comparison, the lowest average TOC concentration with a low range occurred at station 4. In addition, the results showed the average concentration from the highest to the lowest was found at stations 2, 1, 3, and 4. The surface water used in this study contained 7.36 – 15.50 mg/L TOC concentration, which was typically associated with the DOC range. River water has a typical concentration about 2 to 10 mg/L of dissolved organic carbon, which was much higher than groundwater and seawater. Variation in average concentrations of TOC indicated various physical or ecological drivers, chemical processes, spatial changes, which can significantly affect on organic matters dynamics (Maie et al., 2006). The organic matter compositional changes could be induced by biophysical controls, such as changes in composition, which likely result in bioavailability, photoreactivity, nutrient cycling, or chelating capacity and can affect carbon fluxes consequentially ecological drivers not accounted for (Jaffe, 2008). In addition, the hydrology dynamics of surface runoff contributed to the surface water stream (Hood et al., 2006).

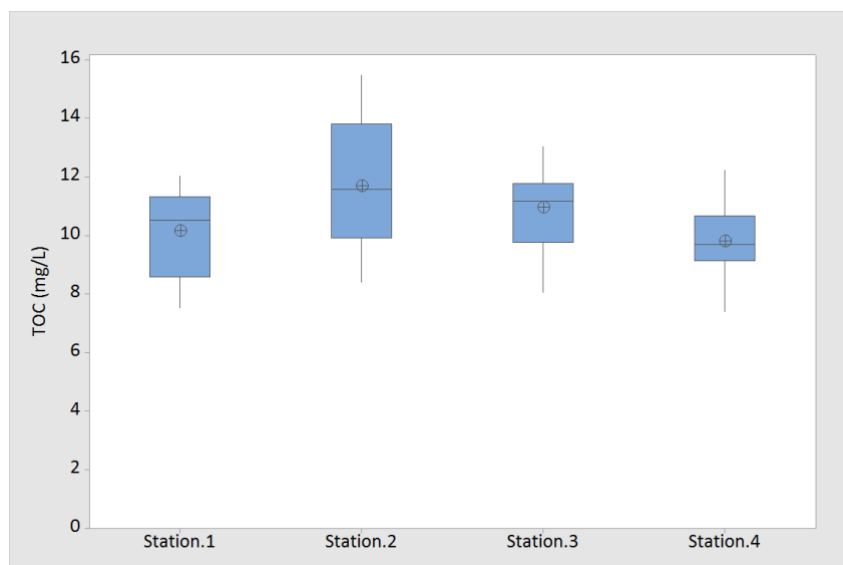


Fig. 2: The average TOC concentration in the river segment at various stations.

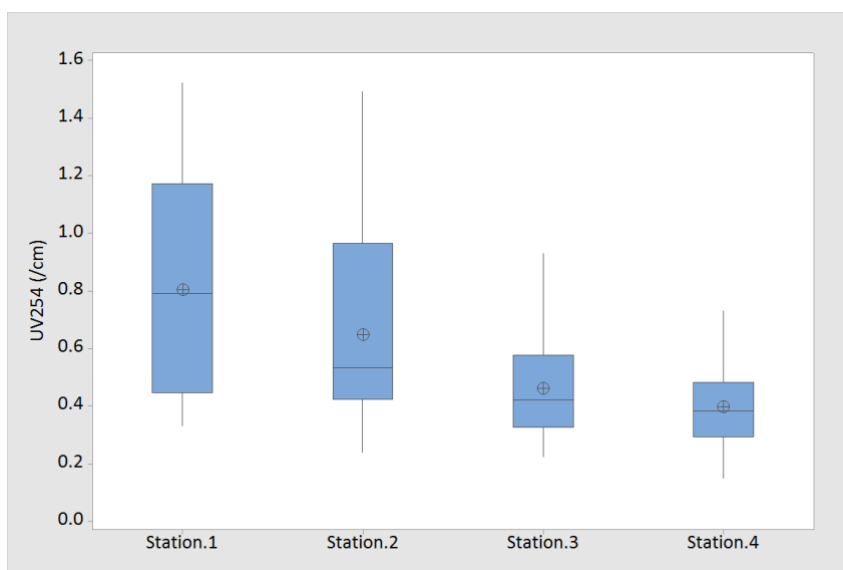


Fig. 3: The average UV<sub>254</sub> concentration in the river segment at various station

Fig. 3 describes the concentration of UV<sub>254</sub>, which corresponded to the organic compounds with an aromatic structure, double bonds of C=C (Matilainen *et al.*, 2011). In this study, the concentration of UV<sub>254</sub> for surface water was 0.148 – 1.524/cm, which was within the typical range of river (0.085 – 0.4/cm) (Edzwald *et al.*, 1985). The results showed that the average highest aromatic compound was detected at Station 1, while Station 4 had the average lowest concentration. Therefore, Station 1 contained higher humic matter with conjugated C=C double structural bonds than the others. Meanwhile, Station 4 contained lower humic matter than the others. As well known, organic compounds of humic matter contain unsaturated carbon bonds (double or triple) or aromatic rings in their molecular structure. Hence, it absorbs an amount of UV light through the water sample (Her *et al.*, 2002).

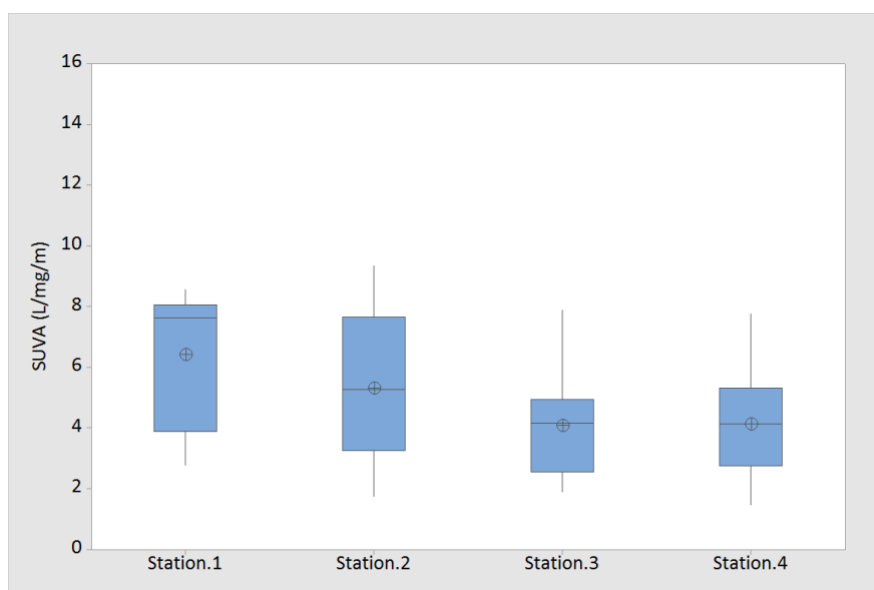


Fig. 4: The average SUVA value in the river segment at various station

Fig. 4. Shows the hydrophobicity of organic matter characteristic or specific UV-absorbance (SUVA) value. The results revealed a value between 1.45 – 9.36 L/mg/m. However, it was mostly higher than 4 along the river segment, which means that the organic matter is mainly consists of humic, hydrophobic, and high molar mass organic material. According to Edzwald and Tobiason (2011), SUVA is a parameter of the organic matter composition in water. Source water with SUVA values  $\geq 4$  indicated that natural organic matter composed mainly of humic or hydrophobic matter, while those  $< 2$  contained mainly non-humic or hydrophilic natural organic matter. The results were consistent with the high concentration of  $UV_{254}$  (0.148 – 1.524/cm). The values typically ranged from 1.0 to 6.0 L/mg/m for surface water. However, values greater than 6.0 were revealed for interstitial waters dominated by a solid terrestrial signature (Jaffe *et al.*, 2008). According to previous studies, these higher values can be as a result of the absorption at 254 nm from colloids, iron, or other components in the sample (Weishaar *et al.*, 2003; Hudson *et al.*, 2007). Combining the bulk parameters of TOC,  $UV_{254}$ , and SUVA value led to characterize the organic matter in the river. Station 2 was mainly composed of the highest TOC with lower aromatic and hydrophobic than 1, and vice versa. Also, station 4 was mainly composed of lower bulk parameters than 3. Therefore, 2 contained more aliphatic organic matter that does not absorb at 254 nm than the others. The lower SUVA value among all stations indicated the mixtures of aquatic humics, hydrophobic and hydrophilic, and molecular weights of organic matter.

*Characteristic of fluorescence dissolved organic matter in the river segment through volumetric fluorescence distribution.*

Fig. 5. Illustrates the fluorescence excitation-emission matrices (FEEMs) for dissolved organic matter in the river segment at a different station, taken on the first week sampling time. Dissolved organic carbon was classified into four regions based on its excitation/emission wavelengths (Ex/Em), namely Region 1 indicated the aromatic proteins-like (AP-like), such as tyrosine and tryptophan, at Ex/Em  $< 250$  nm/ $< 350$  nm. Region 2 identified the fulvic acid-like (FA-like) substances at Ex/Em  $< 250$  nm/ $> 380$  nm, Region 3 was corresponded to the soluble microbial by products-like (SMPs-like) substances at Ex/Em 250-280 nm / $< 380$  nm, while Region 4 was identified as the humic acid-like (HA-like) substances with Ex/Em  $> 280$  nm/ $> 380$  nm (Chen *et al.*, 2003).

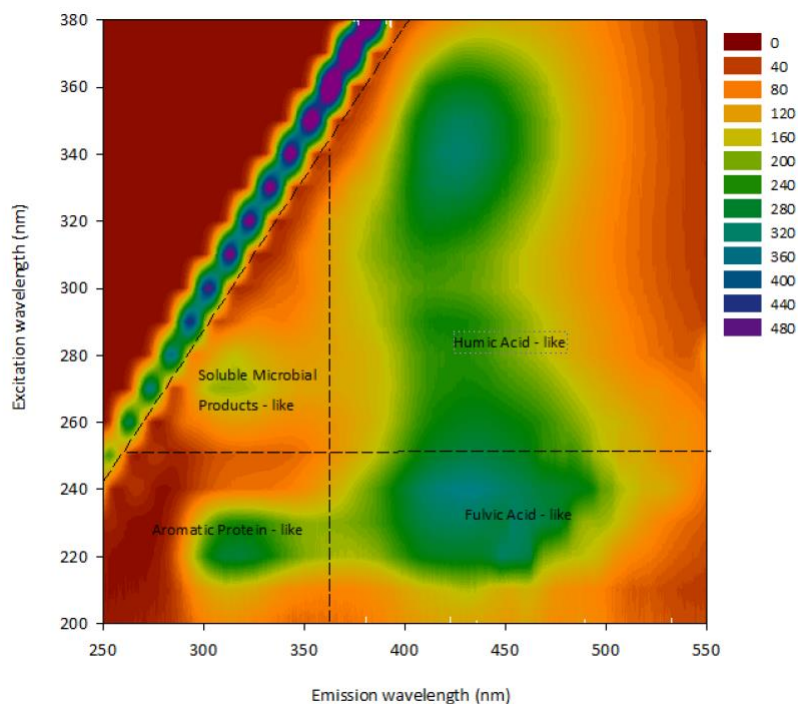




Fig. 5: Spectrum of fluorescence spectrometer analysis in the river segment

This study shows that the fluorescence component from FEEM analysis has consistent results with previous studies (Her *et al.*, 2003; Yao *et al.*, 2016; Moradi *et al.*, 2018; Hidayah *et al.*, 2020). Generally, HA-like and FA-like correlated with aromatic compounds. They mainly exist as carboxylic and phenolic functional groups in natural dissolved organic matter. These fluorescence structures are mostly present as a significant percentage of humic substances, which typically represent over 50% of natural organic matter (Shon *et al.*, 2012). In addition, source water may contain protein-like materials which microbial activities can generate. The amount, characteristics, and properties of dissolved organic matter in the aquatic system depend on their origin and environmental biochemical cycles. Sources of organic matter are classified as allochthonous (generated from a terrestrial watershed) and autochthonous (produced by organism activities, such as phytoplankton activities) (Chari *et al.*, 2013; Zhang *et al.*, 2009; Haraguchi *et al.*, 2019). Terrestrial watershed is mainly composed of humic substances such as fulvic and humic acids as well as humin, which are primarily hydrophobic and rich in aromatic carbon. The autochthonous source material is microbially derived organic, such as algal-derived and effluent organic matter (Kelso and Baker, 2020). Fig. 6 showed the percentage fluorescence response, which was calculated by Fluorescence Regional Integration (FRI) method. The percentage of fluorescence distribution indicated the four fractions quantity of fluorescence organic matter. This study classified the fraction into humic and non-humic substances-like. The first was represented by Region 2 (FA-like) and 4 (HA-like), while the second one by Region 1 (AP-like) and 3 (SMPs-like). Firstly, the results showed the highest total percentage of FRI in Region 2 and 4 was at Station 1 (76.6%), and the lowest total percentage for humic substances-like was at Station 4 (69.2%). Both components are classified as humic substances and are mainly composed of aromatic compounds with high to medium molecular weight (Watson *et al.*, 2018; Hua *et al.*, 2020). Their total percentage FRI showed a consistent UV<sub>254</sub> concentration and SUVA value. Furthermore, Station 1 had the highest bulk parameters, while 4 had the lowest. Secondly, the highest total percentage FRI of Region 1 and 3 (30.8%) was identified at Station 4, with the lowest at 1 (23.4%). This indicated that Station 4 contained abundant proteins substances and microbial-like fluorescence than the others and followed the lowest SUVA value of Station 4 with the highest for Station 1. Region 1 and 3 correlated with high molecular weight protein-like, which had chemical properties related to aromatic amino acids, tryptophan or tyrosine-like (Yamashita *et al.*, 2008; Hua *et al.*, 2020) and low molecular weight microbial humic-like as well as less conjugated double bond organic matter (Nguyen *et al.*, 2013; Hua *et al.*, 2020).

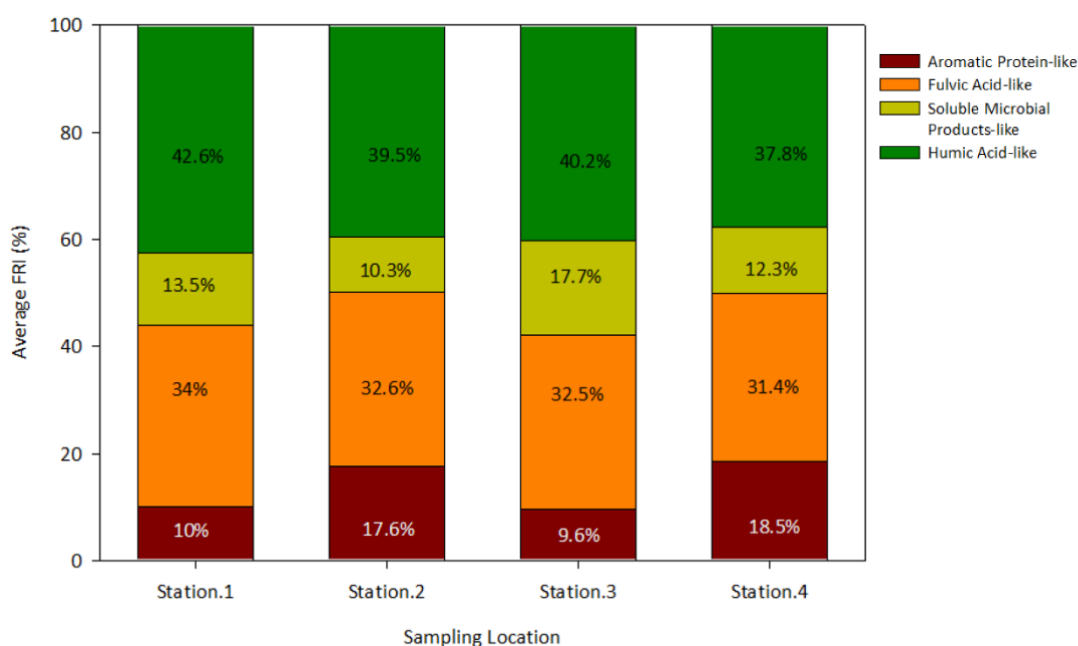


Fig. 6: FRI distribution of fractionated organic matter from the various river segment

The distribution data for the fluorescence of dissolved organic matter in the river segment was tested by Kolmogorov–Smirnov and the bulk parameters. The results showed distribution data for percentage FRI of Region 1 (AP-like), 2 (FA-like), 3 (SMPs-like), and Region 4 (HA-like) with  $P > 0.000$ ,  $0.007$ ,  $0.000$  and  $0.013$  respectively were normal. Furthermore, Analysis of Variance (ANOVA) testing was carried out to determine the differences in mean percentage FRI for each region. The statistical analysis ANOVA One-Way with the Tukey 95% confidence interval also determined whether statistically significant or non-significance differences in percentage FRI of AP-like, FA-like, SMPs-like, and HA-like among all stations. The results showed statistically significant differences in the mean percentage FRI of all fluorescence organic fractions at all stations with  $p = 0.000$ ,  $0.007$ ,  $0.000$ , and  $0.013$  in AP-like, FA-like, SMPs-like, HA-like, respectively. The results provided enough evidence to conclude that the mean percentage FRI of all fluorescence organic fractions at all stations was significantly different. Moreover, the Tukey analysis classified their percentage FRI at each station into two main groups. Station 1 and 2 were grouped in the high percentage FRI of humic substance-like (FA-like and HA-like), while 3 and 4 were classified in the low percentage. This means the former had an average percentage FRI of FA-like and HA-like, which were significantly different from the latter. In addition, stations 3 and 4 were grouped in the high percentage FRI of non-humic substance-like (AP-like and SMPs-like), while stations 3 and 4 were grouped in the low percentage. This showed both had average percentage FRI of AP-like and SMPs-like, which were significantly different from stations 1 and 2. Moreover, statistical box plot analysis presented the pattern of the fluorescence organic matter in the river segments. Fig. 7a to 7d presented box plot with average percentage FRI of the organic matter. Firstly, a comparison among all fluoresces organic compounds showed the average FRI of HA-like was much higher and much lower for SMPs-like than the others. However, HA-like, located at Region 4 of the fluorescence spectra, had the most extensive range of excitation and emission wavelengths. Therefore, the humic acid substances-like region had the most extensive volume distribution of FRI when compared to others (Chen *et al.*, 2003). Meanwhile, SMPs-like or Region 3 comprised a dominant percentage of the fluorescence in wastewater treatment plant effluent (Chen *et al.*, 2003) and was closely related to the phytoplankton activities (Liu *et al.*, 2021; Hua *et al.*, 2020). Second, the average percentage FRI of the organic matter indicated different quantities and quality at each station. The non-humic substances-like fluorescence as presented by AP-like, SMPs-like, with statistical analysis, had a higher percentage FRI at stations 3 and 4 than the others. It was likely that Station 3 and 4 had a higher percentage of extracellular biological organic matter fraction than the other river segments. The fraction was supposed to contain soluble microbial products of amino acids and carbohydrates. Tryptophan and tyrosine which are aromatic amino acids, were confirmed as biological activity products in natural systems and exhibited a distribution of fluorescence response similar to AP-like and SMPs-like of this study (Coble 2007; Determann *et al.*, 1998). The humic substances-like fluorescence as presented by FA-like and HA-like had a higher percentage FRI at stations 1 and 2 than others and were tested by ANOVA One-Way. Combining the bulk parameters of TOC,  $UV_{254}$ , SUVA value, and fluorescence spectroscopy convinced the characteristic of organic matter in the river. Station 1 and 2 had high  $UV_{254}$  concentration, SUVA value, high percentage FRI of FA-like and HA-like substances. It was conjectured that stations 1 and 2 were mainly composed of aromatic, hydrophobic, humic substances organic matter, which may be generated from terrestrial systems.

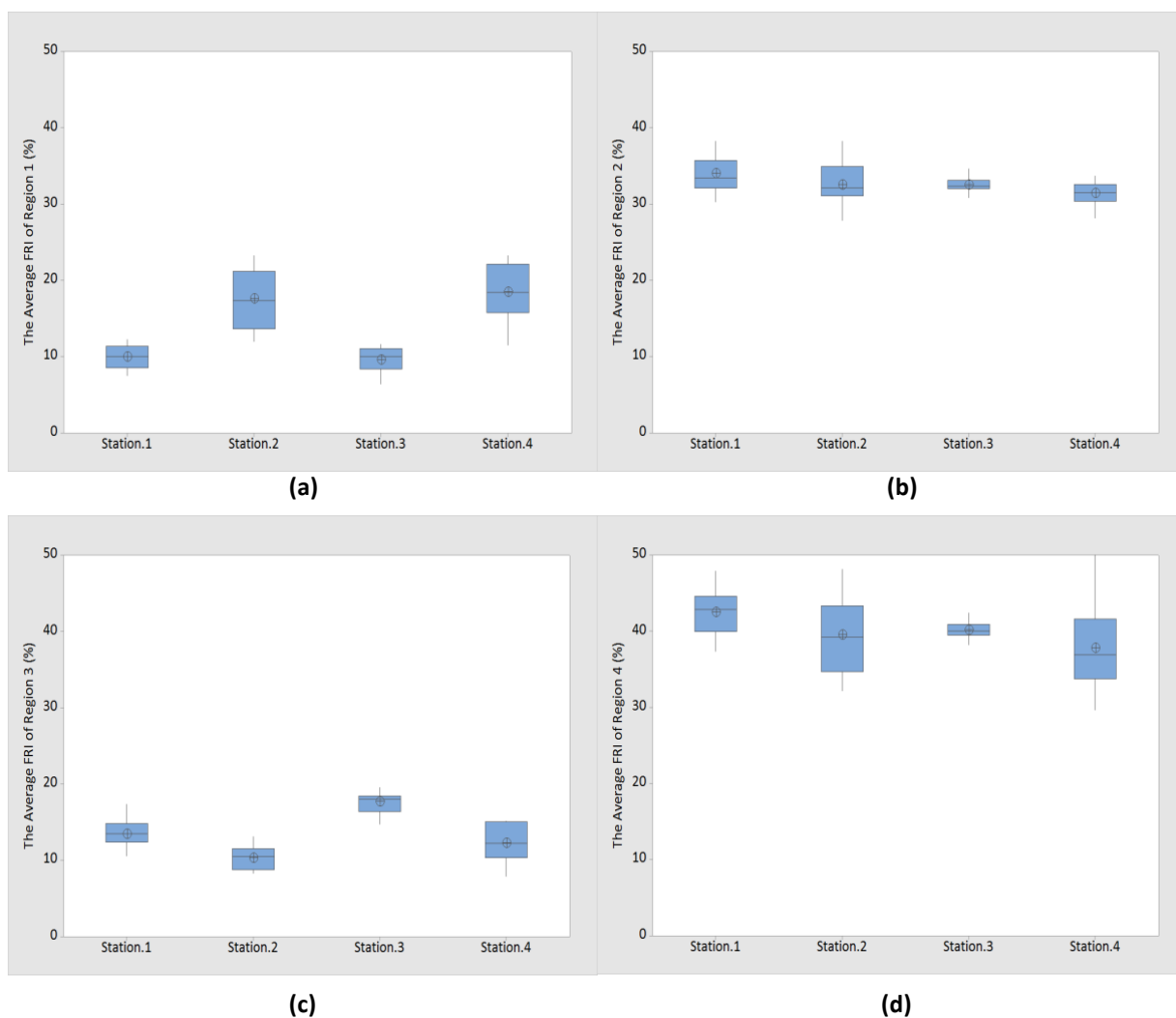


Fig. 7: The average percentage FRI of fluorescence organic matter in the river segment at various stations

Station 3 and 4 had lower UV concentration and SUVA values, with a high percentage FRI of AP-like and SMPs-like than the others. There was a lower SUVA value among all stations indicates in the mixtures of aquatic humics, hydrophobic and hydrophilic, as well as molecular weights of organic matter. This showed that Station 3 and 4 comprised more autochthonous and sources of organic matter from anthropogenic activities. The river ecosystem, which source is terrestrial, autochthonous, and anthropogenic, provided hotspots for storing, transporting, and transforming organic matter. The sources proportions were primarily and terrestrially derived with increased autochthonous inputs from macrophytes. In addition, the sources of dissolved organic matter are a mixture of terrestrial, autochthonous, or primarily from wastewater effluent (Kelso and Baker, 2020).

*Contribution of phytoplankton abundance to fluorescence dissolved organic matter in the river segment.*

This study discovered four main phytoplankton species with various abundance in the river segments, namely *Plectonema* sp., *Nitzschia* sp., *Navicula* sp., and *Pinularia* sp. The distribution data of the phytoplankton abundance in this segment was tested by Kolmogorov–Smirnov. The results showed a usual distribution data for *Plectonema* sp., *Nitzschia* sp., *Navicular* sp., and *Pinularia* sp. as abundance  $P > 0.000$ ,  $0.007$ ,  $0.000$ , and  $0.013$ , respectively. Furthermore, ANOVA testing was carried out to determine the differences in the mean phytoplankton abundance of the river segments. The statistical analysis ANOVA One-Way with the Tukey 95% confidence interval determined whether there were

statistically significant or non-significance differences in the abundance of the species among all stations. According to the results, there were statistically significant differences in the mean abundance of phytoplankton at all stations with p-value = 0.006 and 0.01 in *Plectonema* sp. and *Nitzschia* sp. abundance, respectively. Meanwhile, the analysis generated p-value = 0.156 and 0.412 for *Navicula* sp. and *Pinularia* sp. abundance, respectively, therefore, classified as only one group of phytoplankton abundance. This showed that there were non-significantly differences in both species abundance among all stations. The Tukey analysis classified *Plectonema* sp. and *Nitzschia* sp. abundance at each station into two main groups. Station 1 and 2 were grouped in the high *Plectonema* sp. and low *Nitzschia* sp. abundance, while Station 3 and 4 were classified in the low *Plectonema* sp. and high *Nitzschia* sp. abundance. Furthermore, the statistical box plot analysis presented the pattern of the phytoplankton abundance in the river segments.

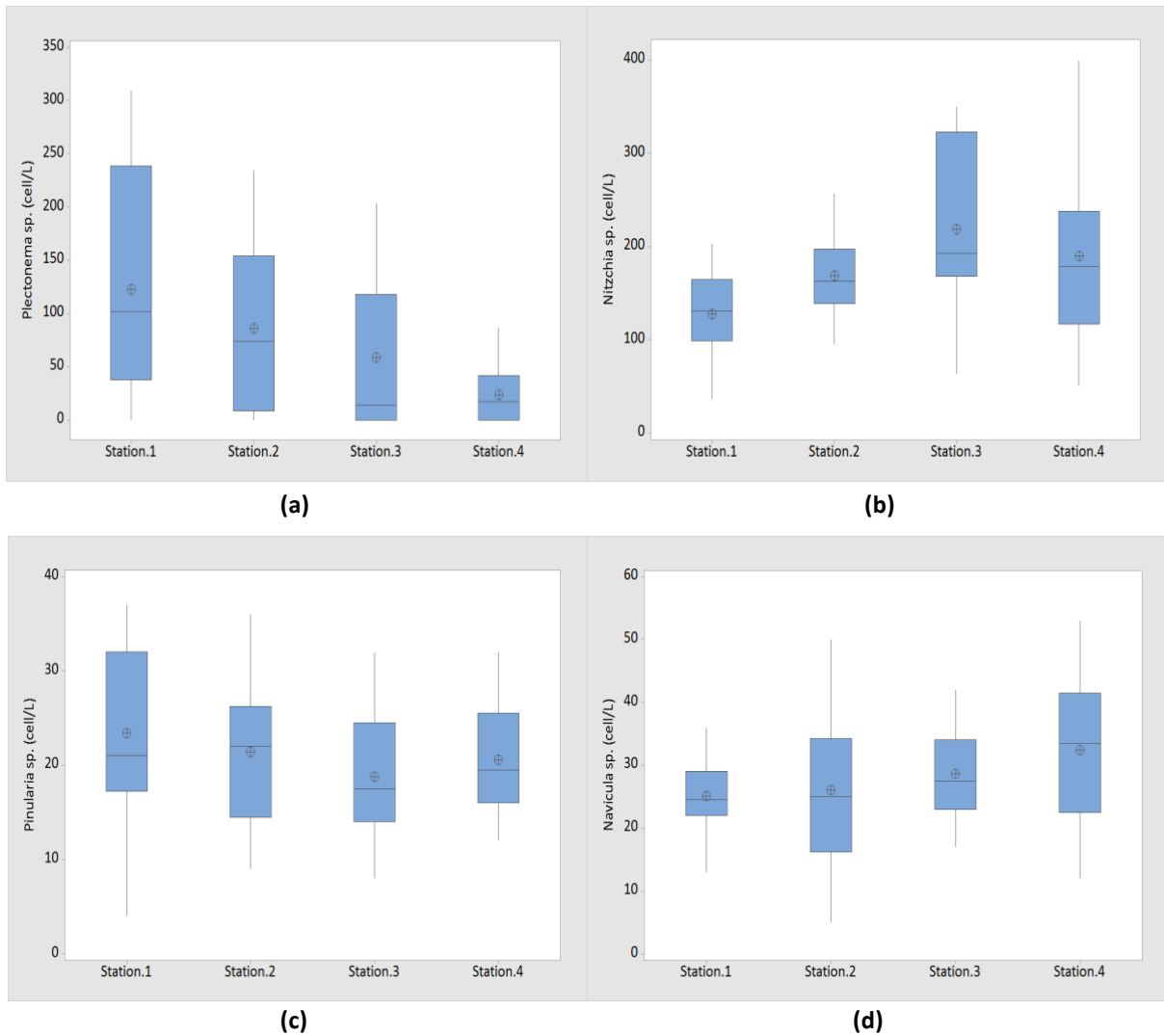


Fig. 8: The average of phytoplankton abundance in the river segment at various station

Fig. 8a to 8d present box plots of their average abundance. Firstly, a comparison among the species at all stations conjectured that *Nitzchia* sp. had a higher abundance, and *Pinularia* sp. was lower than the others. Meanwhile, *Plectonema* sp. had the highest at Station 1 and the lowest at 4. *Nitzchia* sp. had a higher abundance at Station 3 and lower at 1. Moreover, *Navicula* sp. had the highest abundance at Station 4 and the lowest at Station 1. *Pinnularia* sp. gave the highest at Station 1, with the lowest at Station 3. This phytoplankton abundance was strongly influenced by migration, which can occur due to population density and physical environmental conditions, such as changes in temperature and currents (Basu and Mackey, 2018). Secondly, Station 1 was likely to contain a similar abundance in *Plectonema* sp. and *Nitzchia* sp., and the same for *Navicula* sp. and *Pinularia* sp. Stations 2 and 3 showed that the abundance of *Nitzchia* sp. was primarily dominant than others. However, *Navicular* sp. was similar to *Pinularia* sp. Station 4 identified a similar abundance of *Plectonema* sp., *Navicular* sp., and *Pinularia* sp. There is competition in several phytoplankton species that use the same resource lacking in availability, or even regardless of sufficient availability, and competition still occurs when they take advantage of the resource, with one attacking the other or vice versa (Burson et al., 2018).

*The relationship among the bulk parameters, organic fluorescence parameters, and phytoplankton abundance*

The degree correlation between the bulk parameters, fluorescence organic matter, and phytoplankton abundance was examined, as shown in Table 2. Correlation analysis was carried out using TOC and

UV<sub>254</sub> concentrations, SUVA value with percentage FRI of AP-like, FA-like, SMPs-like, or HA-like, as well as the abundance of *Plectonema* sp., *Nitzschia* sp., *Navicula* sp., and *Pinularia* sp. Firstly, based on the correlations of the bulk parameters, TOC concentration was positively higher with Region 1 (AP-like) and Region 2 (FA-like). In addition, UV<sub>254</sub> concentration and SUVA value were significantly correlated with Region 1 (AP-like) and Region 4 (HA-like). The results showed fluorescence spectroscopy, which fractionated AP-like, FA-like, SMPs-like, and HA-like could be used to identify the quantity and quality of organic matter in the source water.

Table 2: The degree correlation among the bulk parameters, fluorescence organic matter, and phytoplankton abundance\*

Parameters	TOC	UV <sub>254</sub>	SUVA	AP-like	FA-like	SMPs-like	HA-like	Navicula sp.	Plectone ma sp.	Pinnularia sp.
UV <sub>254</sub>	0.085 0.502									
SUVA	-0.044 0.729	0.887 0.000								
AP-like	-0.287 0.022	-0.440 0.000	-0.373 0.002							
FA-like	0.254 0.042	0.105 0.411	0.047 0.710	-0.249 0.048						
SMPs-like	-0.038 0.764	-0.198 0.116	-0.228 0.070	0.638 0.000	-0.085 0.505					
HA-like	-0.035 0.786	0.344 0.005	0.344 0.005	-0.674 0.000	-0.022 0.862	-0.348 0.005				
Navicula sp.	-0.109 0.392	-0.331 0.007	-0.289 0.021	0.193 0.126	-0.102 0.422	0.090 0.480	-0.082 0.521			
Plectonema sp.	0.271 0.030	0.137 0.281	0.131 0.303	-0.346 0.005	0.293 0.019	-0.057 0.652	0.110 0.386	0.166 0.189		
Pinnularia sp.	-0.097 0.448	-0.239 0.058	-0.292 0.470	-0.245 0.051	0.142 0.263	-0.268 0.032	0.220 0.080	0.137 0.279	0.320 0.010	
Nitzschia sp.	-0.243 0.053	-0.283 0.023	-0.203 0.108	0.160 0.205	-0.203 0.107	0.176 0.164	0.070 0.585	0.168 0.184	0.035 0.785	0.174 0.170

\*Cell Contents description; Pearson correlation (the first row of the number of correlation between parameters); P-value (the second row of the number of correlation between parameters)

This result was expected since TOC measured all organic carbon, including humic and non-humic substances, as presented by AP-like and FA-like. Secondly, a strong positive correlation between UV<sub>254</sub> concentration and SUVA value indicated that higher aromatic conjugated double bond corresponded to higher molecular weight organic, more hydrophobic, and content of humic substances. These results are consistent with the Pearson correlation between bulk parameters of UV<sub>254</sub> correlation, SUVA, and fluorescence organic matters of AP-like and HA-like. Furthermore, it was conjectured that fluorescence spectroscopy could be used to assess the properties of organic matter existing in the source water. Thirdly, the results showed that TOC had a stronger correlation with AP-like than HA-like. This was probably because the humic structure may incorporate protein-like-fluorophores due to weak interactions based on x-x or van der Waals forces between the dissolved organic matter components. Previous studies indicated that proteins and humic supramolecules containing specific structures attained from phenol or aniline might contribute to the fluorescence. Fourth, this study discovered a strong correlation between DOM and phytoplankton abundance. *Plectonema* sp. correlated with TOC, AP-like, and FA-like, while *Navicula* sp. and *Nitzschia* sp. correlated with UV<sub>254</sub>, and *Pinularia* sp. with SMPs-like. The existence of phytoplankton was likely to enhance the quantity and characteristics of DOM in the aquatic environment. The production of marine-like fluorophores accompanied phytoplankton degradation as a significant source of autochthonous DOM (Wada *et al.*, 2007). In addition, higher molecular weight compounds such as protein (tryptophan)-like fluorescence were presented in exudates when phytoplankton grows (Chari *et al.*, 2013). The combination of the bulk parameters (TOC, UV<sub>254</sub>, and SUVA value), fluorescence spectroscopy, and phytoplankton abundance

convinced the quality of organic matter in the surface water. However, it could be eventually used to monitor the water's quality.

## **CONCLUSION**

This study showed that the quality and quantity of DOM at all stations were significantly different, as classified into two groups with higher bulk parameters at stations 1 and 2 and a lower concentration at 3 and 4. The average TOC concentration for stations 1 and 2 was about a value 10.1-11.7 mg/L, while 3 and 4 were in between 9.8-10.9 mg/L. The average UV<sub>254</sub> concentration for stations 1 and 2 was in the range of 10.1-11.7 mg/L, while 3 and 4 were between 9.8-10.9 mg/L. The average UV<sub>254</sub> concentration for stations 1 and 2 was 0.65-0.8/cm, while 3 and 4 were 0.39-0.65/cm. The average SUVA concentration of stations 1 and 2 was in the range 5.3-6.4 L/mg/m, while 3 and 4 were 4.0-5.3 L/mg/m. In addition, fluorescence spectroscopy with FRI analysis showed stations 1 and 2 were grouped in the high percentage FRI of humic substance-like (FA-like and HA-like) about 74.35%. It was conjectured that stations 1 and 2 were mainly composed of aromatic, hydrophobic, humic substances organic matter, which may be generated from terrestrial systems, while stations 3 and 4 were classified in high percentages non-humic substances-like (AP-like and SMPs-like) about 29.05%. This showed that Station 3 and 4 comprised more autochthonous and sources of organic matter from anthropogenic activities. According to phytoplankton abundance, Station 1 had a high abundance of *Plectonema* sp. (238.5 cell/L) and *Pinularia* sp. (32 cell/L), while stations 2 and 3 mainly consisted of *Nitzschia* sp. (197.5 cell/L and 322.75 cell/L), and *Navicula* sp. (41.5 cell/L) was dominant at Station 4. The Pearson correlation showed a strong relationship between DOM and phytoplankton abundance. Therefore, *Plectonema* sp. was in correlation with TOC (0.271), AP-like (-0.346), and FA-like (0.293), while *Navicula* sp. and *Nitzschia* sp. correlated with UV<sub>254</sub> (-0.331 and -0.283), and *Pinularia* sp. correlated with SMPs-like (-0.268). This study conjectured that the bulk parameters of DOM, fluorescence spectroscopy, and phytoplankton abundance could be used to assess the characteristic of DOM, while the combination of these methods could be used to monitor the surface water quality. Future work should be conducted on the laboratory scale for phytoplankton observation in order to identify the characteristic of organic matter that a kind of phytoplankton species has released. Therefore, it could be used to predict the amount of DOM derived by phytoplankton, DOM derived in the aquatic, and DOM from the terrestrial watershed.

## **AUTHOR CONTRIBUTIONS**

O.H. Cahyonugroho performed the experimental design, analyzed the data, and prepared the manuscript text as well as the literature review. S. Hariyanto and G. Supriyanto interpreted the data and helped in manuscript preparation.

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## **CONFLICT OF INTEREST**

The authors declare no potential conflict of interest regarding the publication of this work. Also, ethical issues, including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and submission, as well as redundancy, have been entirely witnessed by the authors.

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## ABBREVIATIONS

%	Percent
/cm	Per centimeter
ANOVA	Analysis of variance
AP-like	Aromatic proteins-like
C=C	Carbon chain double bonds
cell/L	The number of phytoplankton cell per liter
DOC	Dissolved organic carbon
DOM	Dissolved organic matter
Em	Emission wavelength
Ex	Excitation wavelength
FA-like	Fulvic acid-like
FEEM	Fluorescence spectroscopy using excitation-emission matrices
FRI	Fluorescence regional integration
HA-like	Humic acid-like
L/mg/m	Liter per miligrams per meter
mg/L	Miligrams per liter
mm	Milimeter
$\mu\text{m}$	Micrometer
<i>Navicula sp.</i>	Navicula species
<i>Nitzchia sp.</i>	Nitzchia species
NOM	Natural organic matter
nm	Nanometer
OCD	Organic carbon detector
$P >$	Probability value more than
$P =$	Probability value equal
<i>Pinnularia sp.</i>	Pinnularia species
<i>Plectonema sp.</i>	Plectonema species
<i>P-value</i>	Probability value
SMPs-like	Soluble microbial by products-like
SUVA	Specific ultraviolet absorbance
TOC	Total organic carbon
UV <sub>254</sub>	Ultraviolet at 254 nm wavelength
UVD	Ultraviolet detector
UV/vis	Ultraviolet visible



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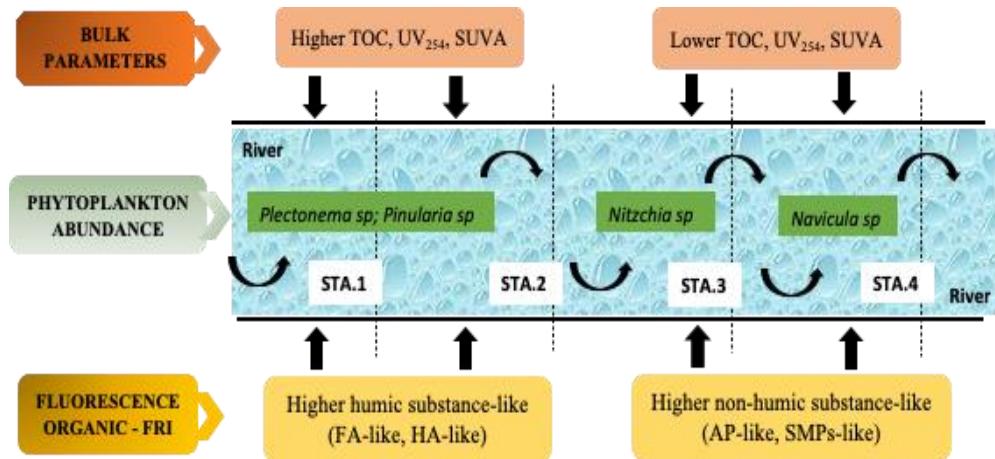
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## GRAPHICAL ABSTRACT



## HIGHLIGHTS

- Characteristic of organic matter and the phytoplankton abundance is different at each segment of surface water;
- There is a substantial correlation among the bulk parameters, fluorescence spectroscopy of DOM and phytoplankton abundance;
- Phytoplankton abundance combined with DOM analysis could be used to evaluate the quantity and quality of organic matter for monitoring surface water quality.