Preliminary Detection of Coliform Bacteria and Zink (Zn) Pollution in Blanakan River Estuary, Subang, West Java.

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Abstract. Coliform bacteria are microorganisms that are often used as indicators of the presence of other pathogenic bacteria. Calculation of total coliform bacteria needs to be done to determine the presence of organic waste pollution that occurs in the Blanakan River Estuary, Subang, West Java. The purpose of this study was to determine the total abundance of coliform bacteria (water status based on coliform level) and to study the relationship between total coliform bacteria and Zn heavy metal content in the Blanakan River Estuary. The method used was purposive sampling carried out at three sampling stations. The results obtained showed that the average coliform content in the three stations ranged from $9.2 \times 10^{-2}$ to $3.0 \times 10^{-1}$ (0.092 – 0.3) MPN/ml sample. Zn heavy metal content in the water at three stations ranged from 0.61 mg/l to 2.13 mg/l. The conclusion of this study is the abundance of the average total Coliform bacteria obtained at each station shows that the total content of Coliform bacteria has not exceeded water quality standards according to the Government Regulation no 82 of 2001. The presence of total coliform bacteria has a weak relationship with the presence of metals Zn.

1. Introduction

Blanakan estuary waters affected by various human activities such as settlements, aquaculture, agriculture, tourism, and fishing activities along the Blanakan river towards Blanakan estuary [1]. Disposal of domestic and industrial waste is usually transported to a river closed to residential and industrial sites. River indirectly accepts hazardous waste, and its contents are affecting the primary function of the river. If an industrial activity does not have a proper waste management system, then the industrial wastewater was directly discharged into the river, and the river becomes polluted [2].

Domestic wastewater can be hazardous because of the domestic waste contains pathogenic microorganisms contained in feces and can transmit various diseases when it is infecting the human body [3]. One of the most common microorganisms found in contaminated water bodies is coliform bacteria. Coliform bacteria are microorganisms that can be used as indicators of environmental pollution or poor sanitation related to domestic wastes. Coliform bacteria, including the Enterobacteriaceae, were divided into two groups, namely the fecal and non-fecal. Total coliform is an indicator of bacteria that can be used as a sign of the presence or absence of pathogenic bacteria pollution. This because the presence of total coliform colonies is positively correlated with the presence of pathogenic bacteria.

The presence of heavy metals in the water can come from various sources, including mining, household, agricultural waste, and industrial waste. Waste pollutants will lead to an increase in heavy metals in the river. That will harm the viability of marine ecosystems and also for other living
organisms, especially humans [4]. Heavy metals are divided into two types of heavy metals; they are essential and non-essential. The first type is essential heavy metals needed by every living organism, but some of them (to some degree) are poisonous. This element usually is present in nature in the form of dissolved or suspended (bound to the solid substance) and present as ionic forms. An example of an essential element of metal is Zn. Hence, the purpose of this study is to determine the abundance of total coliform bacteria, status and to study the correlation of total coliform bacteria with heavy metal Zn in estuary waters of the Blanakan river, Subang, West Java.

2. Methods

2.1 Study site
The Blanakan river is located in Subang district, West Java province. The station geocoordinates were from upstream at -6.277444, 107.6599 to downstream (river mouth) -6.240083, 107.667472 (Figure 1). The width of the Blanakan river is varied from 60 m near the river mouth to 11 m in the upstream. In upstream, the river was surrounded by settlements and paddy field, in midstream was transition area from paddy field to fish pond and downstream was dominated by the fish pond and revegetated by mangrove. The coliform samples were collected in the estuary (downstream) of the Blanakan river in Station 1, 2, and 3.

![Figure 1. The locations of 3 stations in the estuary (downstream) of Blanakan river](image)

2.2 Coliform sampling and analysis
The research method is a survey method. The sampling method used is purposive sampling method. [7-8]. Sampling was conducted in April 2019 in the estuary of Blanakan, Subang, West Java. The water samples for the total coliform test were taken using sterile glass bottles then put into the water until it was full. Samples were then stored in a coolbox. Water samples were taken to the BPPT laboratory for testing the coliform. The method used to calculate the total coliform is a method known as Most Probable Number (MPN) [5].

The coliform analytical methods used were consisted of a presumptive test and completed with a confirmed test. Presumptive tests carried out by using the Lactose Broth (LB) media with a three-tube method, then continue using the confirmative test with Brilliant Green Lactose Broth media (BGLB) and the last was a complete test using endo media [6].
2.3 Zn sampling and analysis.
The water samples for analysis of Zn heavy metal were taken in the same three stations for coliform sampling. Water samples were taken using a polyethylene bottle and stored in a cooler box to be tested in the laboratory. Zn heavy metal analysis was using Atomic Absorption Spectrophotometer (AAS) Shimadzu 6300 variants.

2.4 Data and statistical analysis
The data analysis for calculating the density of coliform bacteria and *Escherichia coli* was based on SNI 2897-2008 [7] with the following formula:

\[
\text{Total Coliform MPN table} = \text{Value} \times \frac{1}{\text{dilution factor}} \quad [8]
\]

Statistical analysis was used to find the relationship between the total coliform with Zn using a correlation test. Correlation test is used to find the relationship and prove the hypothetical relationship between two variables when both variables from the data interval or ratio, and data resources of the two variables are the same. The correlation between the two variables can be determined by testing (r) with the following criteria [5]:
- \( r = 0 \) then no correlation
- \( 0 < r \leq 0.19 \) the correlation is very low
- \( 0.2 < r \leq 0.39 \) then have a low correlation
- \( 0.4 < r \leq 0.69 \) then correlates fairly
- \( 0.7 < r \leq 0.89 \) then have a high correlation
- \( 0.9 < r \leq 1 \) then has a very high and strong correlation
- \( r = 1 \) then has a perfect correlation

3. Result

Table 1. The comparable data of total coliform (MPN/ml) with other locations

| Value/Range | Location                  |
|-------------|---------------------------|
| 9.2 x 10^{-2} - 3.0 x 10^{-1} (0.092 – 0.3) | Blanakan estuary (this study) |
| 4 x 10 - 12.8 x 10 | Morosari estuary, Demak [5] |
| 6.09 x 10^{2} - 6.21 x 10^{2} | Landak estuary, Pontianak [9] |
| 2.4 x 10 - 4.9 x 10 | Siak River, Riau [10] |

In Table 1, we can see the coliform in the study has the lowest value compared to the coliform value from other locations.

Table 2. Total coliform (MPN/ml) in the Blanakan estuary

| Station | Value/Range | Government Regulation No. 82 of 2001 |
|---------|-------------|--------------------------------------|
| 1       | 3 x 10^{-1} (0.3) | 1 x 10^{1} |
| 2       | 3 x 10^{-1} (0.3) | 1 x 10^{1} |
| 3       | 9.2 x 10^{-2} (0.092) | 1 x 10^{1} |

In Table 2, we can see the range of coliform were from 9.2 x 10^{-2} to 3 x 10^{-1} (0.092 – 0.3) MPN/ml. According to Government Regulation No. 82 of 2001 [11].
Table 3. The comparable data of Zn (mg/l) with other locations

| Value/Range  | Location                        |
|-------------|---------------------------------|
| 0.61-2.13   | Blanakan river (this study)     |
| 0.9973-2.7815 | Kamal coast [12]               |
| 0.0053-0.0104 | Membramo river [13]           |
| 0.002-0.012  | Pangkajene river [14]          |

Table 4. Zn (mg/l) in the Blanakan estuary

| Station | Value/Range | Government Regulation No. 82 of 2001 |
|---------|-------------|--------------------------------------|
| 1       | 2.13        | 1 x 10^-2                             |
| 2       | 0.61        | 1 x 10^-2                             |
| 3       | 0.64        | 1 x 10^-2                             |

In Table 4, we can see that Zn obtained from the Blanakan estuary has exceeded a threshold that has been determined by Government Regulation No. 82 of 2001.

Table 5. Correlation analysis of total coliform (MPN/ml) with Zn (mg/l) in the Blanakan estuary

| Station | Total coliform | Total Zn |
|---------|----------------|----------|
| 1       | 3 x 10^-1 (0.3)| 2.13     |
| 2       | 3 x 10^-1 (0.3)| 0.61     |
| 3       | 9.2 x 10^-2 (0.092)| 0.64 |

Figure 2. Correlation of total coliform (MPN/ml) with Zn (mg/l) in the Blanakan estuary

4. Discussion

The total coliform obtained at three stations showed different results. Total coliform at the first station was 3 x 10^-1 (0.3) MPN/ml, at the second station was 3 x 10^-1 (0.3) MPN/ml and at third station was 9.2 x 10^-2 (0.092) MPN/ml. The first station and the second station have the highest total coliform while the third station has the lowest total coliform among the three stations. This result occurs because station 1 and station 2 have a location closed to the settlement resulting in many activities that produce domestic waste. Hence it has the highest total coliform. Wastes from residential areas around stations
1 and 2 are one of the largest contributors of coliform bacteria that cause biologically polluted waters. Household waste is a source of high biological contaminants, which can come from the kitchen, bathroom, laundry, a former industrial waste household, and human waste [5]. Station 3 has the lowest total coliform since station 3 was adjacent to the sea so that it has a high salinity level. High and low values of salinity can affect bacterial life. The high salinity will cause a low abundance of coliform bacteria and vice versa, and the low salinity levels will reduce coliform bacteria [5]. When compared to the total coliform in other locations (Table 1), total coliform in the Blanakan estuary was still low. Total coliform bacteria obtained still below the water quality standard according to Government Regulation No. 82 of 2001 [11]. This value indicated that the stream in Blanakan was not biologically poor.

The Zn obtained in the Blanakan estuary showed different results. Zn in station 1 was 2.13 mg/l, in station 2 was 0.61 mg/l and in station 3 was 0.64 mg/l. Station 1 has the high Zn resulting from the use of chemical fertilizers containing Cu and Zn, household wastes containing Zn, corrosion of water pipes, and consumer products (detergent), which were not handled properly [13]. Station 2 is the station with low Zn because station 2 was located near to the mangrove.

When compared with the Zn in the Kamal coast, Zn in the Blanakan estuary was relatively low. However, the Zn was relatively much higher when compared with the Zn in Membramo and Pangkajane rivers. Zn obtained from the Blanakan estuary has exceeded a threshold that has been determined by Government Regulation No. 82 of 2001 [11]. According to Putri et al. (2016), activities around downstream and close to harbor and estuary are contributing to high domestic waste. In Blanakan water, such activities were also found [14].

Significant values obtained from the correlation test in Figure 2 show the $R^2$ value of 0.235, while the $R$-value obtained was 0.484. The results obtained show that the value of $R$ is within the criteria of $0.4 < r \leq 0.69$ [5]. This value indicates that there is a moderate correlation between total coliform with the presence of Zn.

5. Conclusion

Total coliform obtained in the Blanakan estuary ranged from $9.2 \times 10^{-2}$ to $3 \times 10^{-1}$ (0.092 – 0.3) MPN per ml. Total coliform bacteria obtained at each station has not exceeded the quality standard according to Government Regulation No. 82 of 2001. The Zn in the Blanakan estuary was ranged from 0.61 to 2.13 mg/l. The Zn obtained has exceeded the quality standards according to Government Regulation No. 82 of 2001. Based on the correlation test, it shows a considerable correlation between the total coliform with Zn in the Blanakan estuary, Subang, West Java.

Acknowledgment

This research was funded by DRPM UI (Directorate for Research and Community Service of Universitas Indonesia) with contract number NKB-0029/UN2.R3.1/HKP.05.00/2019

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