Organic pollution detection based on protozoa saprobity index at Bojonegara waters of Banten Bay

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Abstract. One of the problems of the aquatic environment is organic pollution which can disturb the balance of the ecosystem. The study was conducted to analyze organic pollution based on protozoa saprobity index and to analyze some environmental parameters that affect the protozoan population in the waters of Bojonegara Banten Bay. Descriptive method with survey techniques was used in this study, consisting of 3 observation stations determined based on the presence of different industrial activities. Data were analysed through calculation of the saprobic index, diversity index and abundance, while the analysis of the relationship of several environmental parameters was conducted using the PCA (Principal Component Analysis) program. The results showed that 12 genera of Protozoa were identified with a total of 102 individuals. The waters of Bojonegoro, Banten Bay are already polluted organically with moderate pollution criteria (Beta-meso saprobic) with saprobic index values ranging from 2.30 - 2.47, while the environmental parameters that have the highest influence on organic pollution are brightness, temperature, pH, turbidity, total dissolved solid (TDS), dissolved oxygen, and biological oxygen demand (BOD).

1. Introduction
Bojonegara waters are part of the Banten Bay that is located in the coastal area of Cilegon. This area is an open bay bounded by Tanjung Platu and Tanjung Pontang on the right and left, as well as a place for river flow [1]. Cilegon city as a large-scale industrial city gives influence not only on economic aspects but also on the surrounding environments [2]. Large industrial activities, fisheries, and other human activities cause a lot of waste to be discharged into the sea, which causes pollution [1]. One way to measure water quality is to know the saprobic index. This system studies the effect of pollution originating from organic waste discharges on aquatic biota [3,4]. The study was conducted to determine the quality of waters based on the level of organic pollution through saprobity index analysis using protozoa as an indicator, as well as knowing some environmental parameters that affects the abundance of protozoans in Bojonegoro waters of Banten Bay, as basic information for environmental management and control for anthropogenic negative impacts in ecosystem and fisheries sector.

2. Materials and methods
The study was conducted in Bojonegara Banten Bay from November to December of 2019. Sampling was conducted at 3 observation stations which were different locations near factories, station 1 was located near the shipyard factory, station 2 near the sugar refinery factory, and station 3 near the lubricant factory. Research using water analysis methods is commonly used in hydrobiological observations [5,6]. Protozoan samples were taken by filtering as much as 50 liters of water in a composite manner using...
Van-dorn, then filtered with plankton net to obtain 20 ml sample volume. The filtered water sample was put into a bottle, preserved with 4% formaldehyde for further observation, and identified under a microscope with 100x to 400x magnifications. Observations were made 3 times in laboratories using the identification manual books.

Environmental parameters, which include physical and chemical parameters, that were measured comprised of: temperature, salinity, brightness, base substrate, depth, pH, DO, BOD, nitrate, nitrite, and orthophosphate. Data were analyzed descriptively through the calculation of abundance index and protozoan saprobic index [3]. The effect of environmental parameters on the saprobic index was analyzed using multivariate statistical analysis based on the Principal Component Analysis program [7].

3. Results and discussion

3.1. Abundance of protozoa

The total number of protozoa recorded were as much as 102, consisting of 14 species belonging to 12 genera. The highest abundance was Euglena spirogyra (0.504 ind/l) and the lowest was Amphisiela sp (0.036 ind/l). The total abundance based on stations showed that the highest abundance was at station 2 (1.530 ind/l) followed by station 1 (1.116 ind/l) and station 3 (1.008 ind/l) (Table 1).

| No | Protozoa               | Observation Station | Total abundance |
|----|-----------------------|---------------------|-----------------|
| 1  | Euglena mutabilis     | 0.036               | 0.216           | 0.036 | 0.324 |
| 2  | Euglena spirogyra     | 0.144               | 0.216           | 0.144 | 0.504 |
| 3  | Euglena anabaena      | 0.144               | 0.216           | 0.036 | 0.432 |
| 4  | Vorticella sp         | 0.036               | 0.108           | 0.072 | 0.216 |
| 5  | Didinium nasutum      | 0.180               | 0.108           | 0.036 | 0.324 |
| 6  | Paramaecium caudatum  | 0.072               | 0.144           | 0.072 | 0.224 |
| 7  | Stentor               | 0.036               | 0.072           | 0.036 | 0.216 |
| 8  | Gymnodinium aeruginosum| 0.144              | 0.072           | 0.036 | 0.252 |
| 9  | Stylonica mytilus     | 0.036               | 0.072           | 0.036 | 0.144 |
| 10 | Cyclidium glaucoma    | 0.072               | 0.144           | 0.108 | 0.324 |
| 11 | Amphisiela sp         | 0.036               | 0.036           | 0.036 | 0.036 |
| 12 | Euplotes sp           | 0.036               | 0.108           | 0.072 | 0.216 |
| 13 | Nasula ornata         | 0.072               | 0.108           | 0.036 | 0.216 |
| 14 | Amoeba proteus        | 0.180               | 0.072           | 0.072 | 0.252 |
| Total |                      | 1.116               | 1.530           | 1.008 |

The highest composition based on the genus of protozoa was represented by Euglena (35%) and the lowest was represented by Amphisiella (1%) (Figure 1).

![Figure 1. Composition of protozoa.](image-url)
3.2. Saprobic index

Saprobic index values from 3 observation stations range from 2.30 -2.47 with the highest to lowest values represented by stations 3, 1, and 2 respectively. (Table 2).

| Station | Saprobic index value | Pollution Level Criteria |
|---------|----------------------|--------------------------|
| 1       | 2.380                | Beta-Mesosaprobic (moderately polluted) |
| 2       | 2.305                | Beta-Mesosaprobic (moderately polluted) |
| 3       | 2.471                | Beta-Mesosaprobic (moderately polluted) |

3.3. Environmental parameters

The results of measurements of environmental parameters at each station and the time of observation, showed varying results, but most of the environmental parameters were still within the normal range for the life of aquatic biota. PCA analysis results showed several environmental parameters which are the main characteristics that affects the value of the saprobic index. Those parameters are: brightness, temperature, pH, turbidity, TDS, dissolved oxygen (DO) and biological oxygen demand (BOD).

Referring to table 1, the identified protozoas are noted as bioindicators of organic pollution. The protozoas are supported by environmental parameters that are still in the normal range, including pH in the range of 6-8. The genus of Euglena has the highest abundance spread over three observation stations compared to another genus. *Euglena spirogyra* dominated the waters followed by *Euglena anabaena* and *Euglena mutabilis*. This is because Euglena favor sandy substrates and clay bottom habitats. It also is a tolerant species in moderately polluted environments such as environments with a large amount of organic matter [8]. The abundance of *Cyclidium* is quite high due to the appropriate environmental parameter like pH, because the normal pH range for protozoa is 7-8. *Cyclidium* is also an organism that tolerates the acidic conditions of many fouling materials. Most of the protozoa found belongs to the Ciliophora subclass, which has a cosmopolitan nature and has a high tolerance to extreme conditions [6].

Based on the saprobity index value in table 2, the waters of Bojonegoro of Banten Bay was indicated by organic parameter as polluted with moderate criteria. This shows that in the environment, there has been a change in the scale of space and time and as an indication of anthropogenic environmental pressures [9].

In accordance with the criteria of the saprobic index: values in the range of 1.00-1.50 includes Oligosaprobic, which means that the waters are not polluted or lightly polluted, values in the range of 1.51-2.50 includes Beta mesosaprobic, which means that it is moderately polluted, values in the range of 2.51-3.50 includes Alfa-Mesosaprobic, which means that it is highly polluted and values in the range 3.51-4.00 includes Polysaprobic, which means that the waters are extremely polluted [3]. The results of previous studies show that in the waters of the Cilegon coast, pollutants indicators of industrial waste discharges such as Nitrite and Ammonia are free at a set threshold [2].

Referring to figure 1, the number and composition of aquatic organisms correlate with the level of water pollution. Some microscopic organisms such as diatoms and dinoflagellate zooplanktons can be used as estimators of pollutants and water conditions [10]. Protozoas are part of the planktonic group and can be used as bioindicators because they have a high sensitivity to the presence of contaminants [11]. Protozoan species variations found at each observation station were almost uniform. Some species inhabit the beta-mesosaprobic waters zone due to their sensitivity to environmental conditions such as the presence of nitrate and nitrite. *Paramecium* was found abundantly, as well as *Didinium* which is known as a predator of *paramecium* [12]. *Vorticella*, which is an indicator of beta-mesosaprobic waters, was found at each observation station. This species has a wide tolerance in the polisabrobic and beta-mesosaprobic zone, which contains a lot of organic matter [13]. Polisabrobic means that the waters are extremely polluted [3].
The results of the PCA analysis is that the parameters that affect the protozoan index values are brightness, temperature, pH, turbidity, TDS, DO, and BOD. Brightness directly affects turbidity, the lower the turbidity, the higher the brightness will be, but the higher the TDS, the higher the turbidity and it will have an impact on the life of the organisms. Brightness is greatly influenced by weather, time of measurement, and suspended solids [14]. Turbidity of the waters tends to be high, ranging between 68-105 NTU. This could be caused by many factory waste discharges that contains suspended material. This condition affects the lives of protozoa and macrobenthos [15,16].

The temperature range is still within normal limits with a range of 29.7 – 31.0 oC. Organic discharges are wastes that are easy to destroy or are degraded by microorganisms, so that if they are discharged into water it will increase the population of microorganisms and will increase BOD levels. Dissolved oxygen levels are relatively low in the range of 2.4 – 4.4 mg/l. This is inversely proportional to the relatively high BOD value of 4.01 - 5.86 mg/l. The high value of BOD indicates organic pollution [8].

4. Conclusion
The water of Bojonegoro, Banten Bay is indicated to have been organic polluted with medium category (beta-meso saprobic), characterized by high abundance of several protozoa acting as bioindicators of organic pollution, which includes Euglena, paramecium, Didinium, Vorticella and Cyclidium. Some parameters that indicates the extent of pollution are brightness, temperature, TDS, pH, turbidity, dissolved oxygen, and biological oxygen demand (BOD). This condition is an early warning to properly manage the ecosystem in an effort to support the sustainable development program.

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