

Detecting Levels of Environmental Disturbance from Fish Farming Using Multivariate Analysis

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Environmental disturbance of water ecosystem, both natural and anthropogenic, may affect sediment structure, which macrobenthic animals inhabit. This study aims to determine environmental disturbances caused by aquaculture, through linking abiotic parameters and macrobenthos community structures. A coastal area of Menjangan Besar Island, Central Java was researched, between August and November 2018. A systematic, random sampling method was used by collecting three stations from both: Location I (monoculture sites) and Location II (reference sites). Sampling occurred twice, with a sampling interval of two months. Non-metric Multi-Dimensional Scaling (NMDS) of Bray-Curtis similarities and k-dominance curves represented visually the differences between sites over time. To assess the environmental disturbance at each station, EWS-3SWJ software was introduced. Results were acquired using NMDS by location and sampling time. They indicate a tendency of grouping, and hence a degree of difference in macrobenthos community structure response, spatially and temporally. The results of the k-dominance curve at the R3T1 (Reference site; second sampling time) and station M3T1 (Monoculture site; first sampling time) are at the top. This indicates the dominance of a species from *Anodontia* sp. Both locations are classified as moderately disturbed areas, owing to the strong current and organic nitrogen level.

Key words: *Fish Farming, Environment, Water Ecosystem*

Introduction

Aquaculture is usually operated in a limited space, at a high density of farmed biotas, in open or semi-open water ecosystems. For growing the fish, high rate feeding in intensive aquaculture may reduce the quality of the water's ecosystem, due to the unfed pellets produced during the operation that eventually affect farming production capacity. Environmental degradation has an impact on decreasing the quantity of farmed fish production (Putro and Hariyati, 2012), both spatial and temporal (Aufa et al., 2018).

Previous researchers emphasised challenges and problems to be taken into account when dealing with aquaculture practices, including efforts to: increase production and carrying capacity (Mayerle et al., 2020), maintain the quality of feed (Dineshababu et al., 2019; Cadillo-Benalcazar et al., 2020), maintain environmental sustainability and product quality criteria (Luna et al., 2019; Freitas et al., 2019). Also, environmental issues can threaten the sustainability of fish farming itself (Hariyati and Putro, 2018; Yogev et al., 2020). Furthermore, organic materials generated from aquaculture can change aquatic and sedimentary environments, qualitatively and quantitatively. The continuous accumulation of organic matter may reduce oxygen in the boundary layer between water and sediment, which affects changes in the composition of infaunal macrobenthic animals (Hariyati and Putro, 2018; Aufa et al., 2018).

Oxygen depletes mainly because of increased oxygen consumption by bacteria and other organisms, during the degradation of organic matter. If oxygen has been depleted, anaerobic processes occur until ammonia, hydrogen sulphide and methane are produced. This occurs especially in high accumulations of organic matter, due to limited water exchange and turbulence. Furthermore, this condition may reduce the food intake, growth rate, and survival rate of farmed fish. Dissolved oxygen produced from natural processes cannot afford the consumption demand of fish, water and sediment. Thus, together with other abiotic components, dissolved oxygen is essential element influencing productivity of farmed fish (Putro et al., 2018; Li et al., 2019).

Macrobenthic communities inhabiting bottom-of-the-sea waters are sensitive to environmental disturbances (Manoharan et al., 2011; Putro et al., 2017). The analysis of macrobenthic infauna animals has been applied as one of the main criteria, in determining environmental quality for aquaculture management in various countries (Putro and Adhy, 2019a; Aufa et al., 2018). Macrobenthos has an important role as a food network in an aquatic ecosystem and the decomposition of organic material in waters. Polluted waters will affect the survival of macrobenthic organisms, because they are easily affected by the presence of pollutants, both physical and chemical. Because of variability of macrobenthic structure in response to environmental disturbance, further research is needed. The purpose of

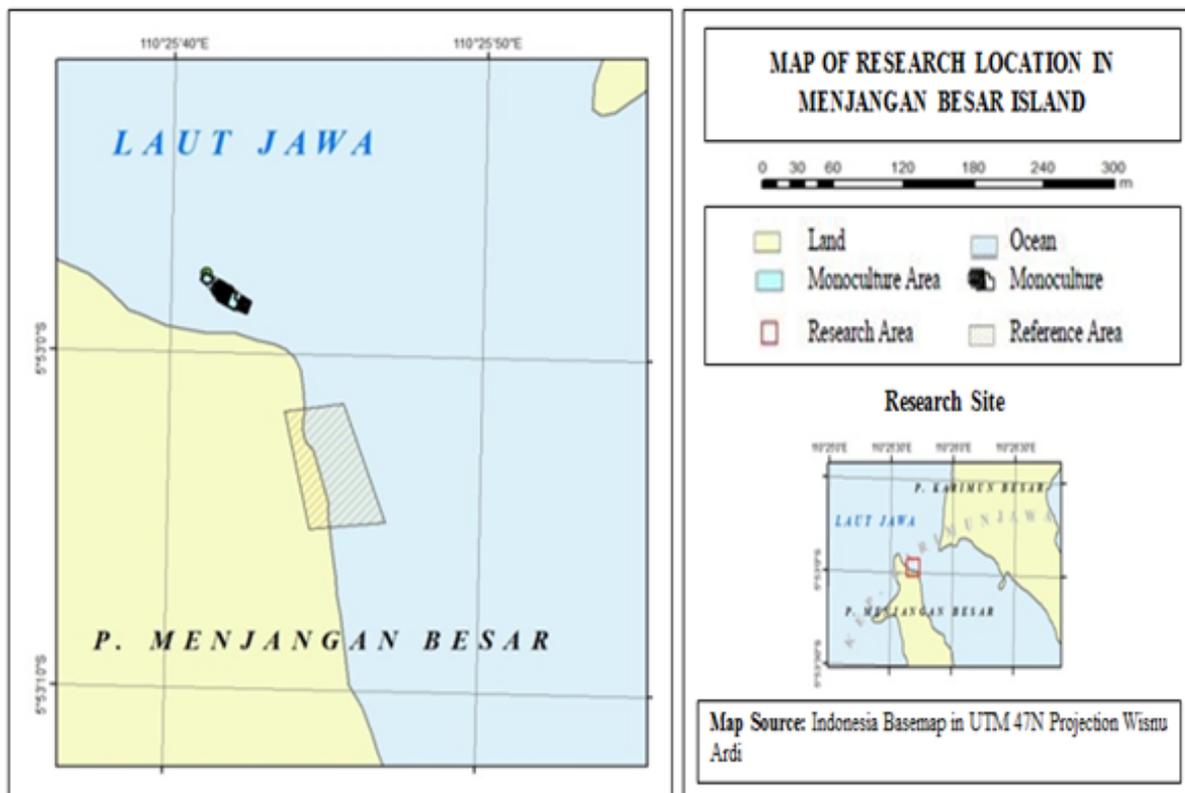
this study was to determine the level of environmental disturbance in the area of Menjangan Besar Island, Karimunjawa, Central Java, by using biotic and abiotic parameters with multivariate approaches.

Research Method

Sampling Sites and Procedures

Sampling occurred at Menjangan Besar Island, Kepulauan Karimunjawa, Central Java, Indonesia, at coordinates between 110°25'40"-110°25'50"E and 5°53'10"-5°53'0"S. Two sampling locations were assessed, with three stations and three replicates for each location, i.e. a floating net cage of monoculture and reference area.

Figure 1. Map of sampling sites at Menjangan Besar Island, Karimunjawa, Central Java, Indonesia as a study site.



Sediments were taken from the two locations using Eckman Grab. Each sediment sample was put into a 2L plastic jar, containing 10% formalin and 70% ethanol solutions. The physico-chemical water parameters was measured three times, for each location. The parameters measured in this study were pH, temperature (°C), dissolved oxygen (DO), water current, and salinity. Sediment samples from each sampling station were further analysed, for sediment

composition (coarse sand, fine sand, silt, and clay) and organic contents (total carbon and nitrogen organics).

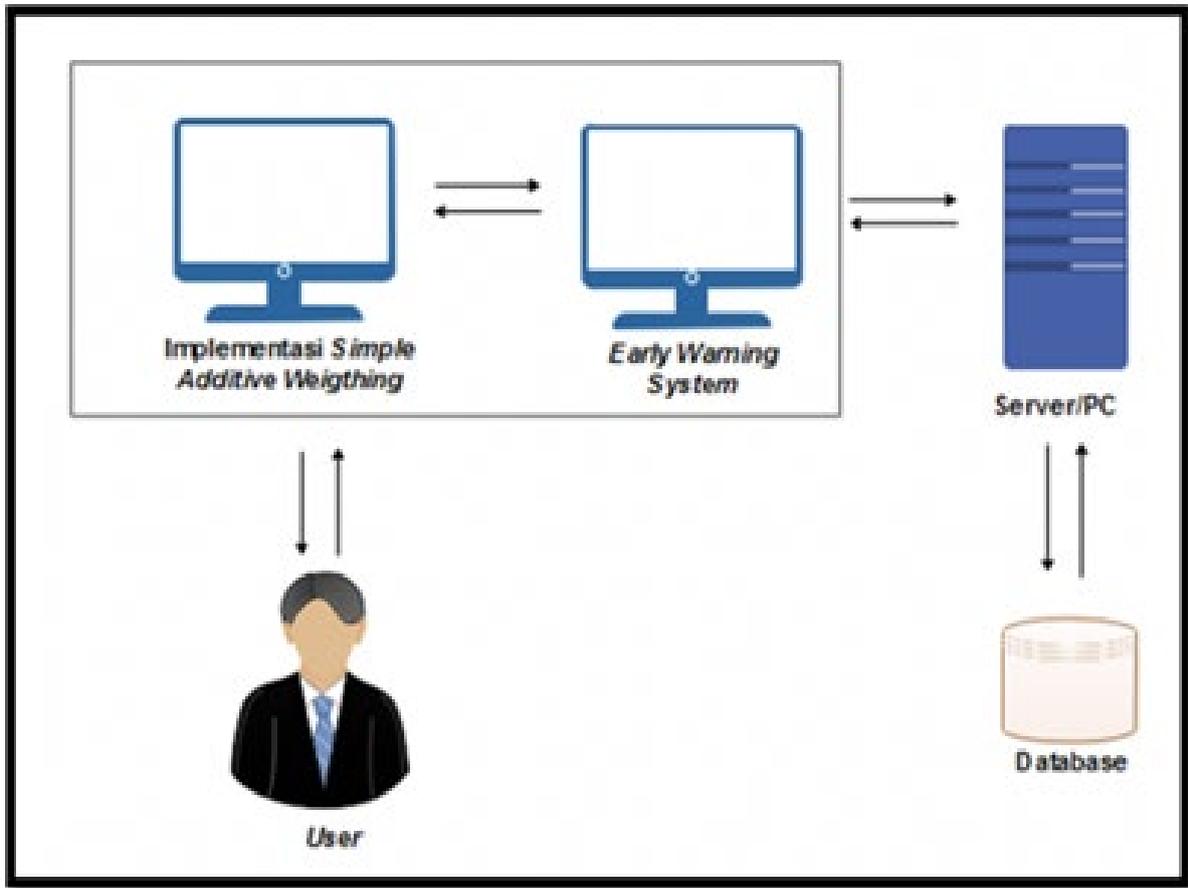
Data Analysis

Data regarding biotic and abiotic parameters were analysed, using multivariate and graphical methodology. The samples were taken at two sampling times, i.e. August and November 2018. The proportion of macrobenthic abundance between the two locations was presented in a pie-diagram, below. Changes in the dominance pattern of macrobenthic assemblages were assessed, using cumulative k-dominance curves for each station plotted on the same graph. The extent of disturbance was evaluated by a comparison of the samples between sites and times based on the curves. The dominance (and intrinsic diversity) of the assemblages was generated by plotting cumulatively ranked abundances against log species/taxa rank (Aufa et al., 2018). Non-metric Multi Dimensional Scaling (NMDS) of Bray-Curtis similarities on log $(X + 1)$ transformed data was used, to provide a visual representation of differences between sites over time on the ordination. The analysis incorporates all sites at all times, to observe any tendency of separation between farmed and reference sites. Correlations between physical-chemical factors, substrate granular composition and the carbon and nitrogen content with the abundance of molluscs, were analysed using BIO-ENV. All multivariate and graphical methodology was analysed using PRIMER software (Aufa et al., 2018).

Level of Disturbance Assessment

The status of the aquatic environment was determined using the Early Warning System-EWS-3SWJ software (Putro and Adhy, 2019a). It is an ecological assessment for water ecosystems using macrobenthic assemblage. The EWS-3SWJ application measured the level of water quality in aquaculture environments, by taking into account the biotic and abiotic components as well as diversity, evenness, and dominance indices. Each component has a weight level that affects the results of determining the level of environmental disturbance. The weight of each component results from the extraction of a team experienced in this field of research at the Center of Marine Ecology and Biomonitoring for Sustainable Aquaculture (Ce-MEBSA: ce-mebsa.fsm.undip.ac.id). The main output of the EWS-3SWJ application is to show the level of disturbance at the observed location. It is complemented by conclusions and recommendations. Other capabilities of this application including its feature of user settings, along with password management, counting water quality by adding taxa indicators. Further, each calculation process can be managed together as a data bank for further analysis related to water quality in various places/regions.

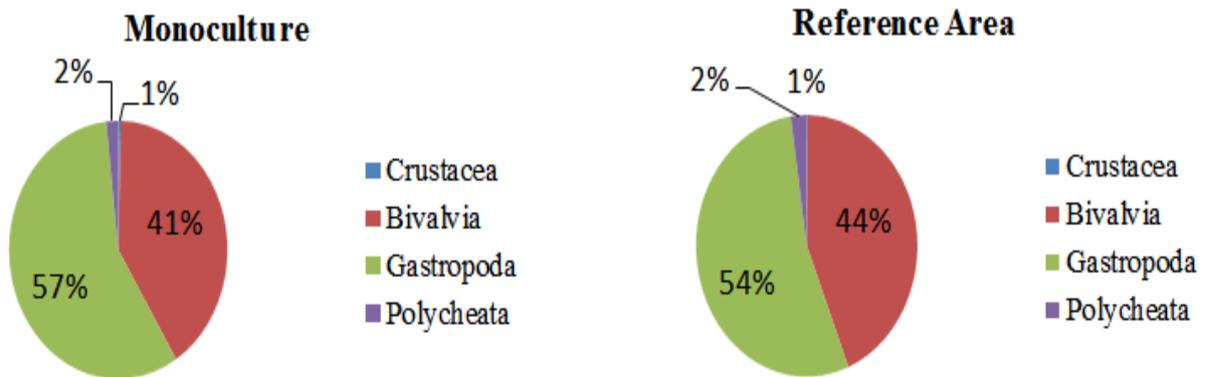
Figure 2. The schematic flow chart of operational procedure of the EWS-3SWJ software.



Macrozoobenthic Structure: Abundance and Proportion

This study found 89 species with a total abundance of 1,135 individuals, consisting of 52 families and 4 macrozoobenthic classes; i.e the Gastropoda class comprised 52 species from 31 families, the Bivalvia class comprised 26 species from 12 families, the Polychaeta class comprised eight species from seven families, and the Crustacea class comprised two species from two families.

Figure 3. The proportion of macrobenthos abundance on floating net cage and reference area



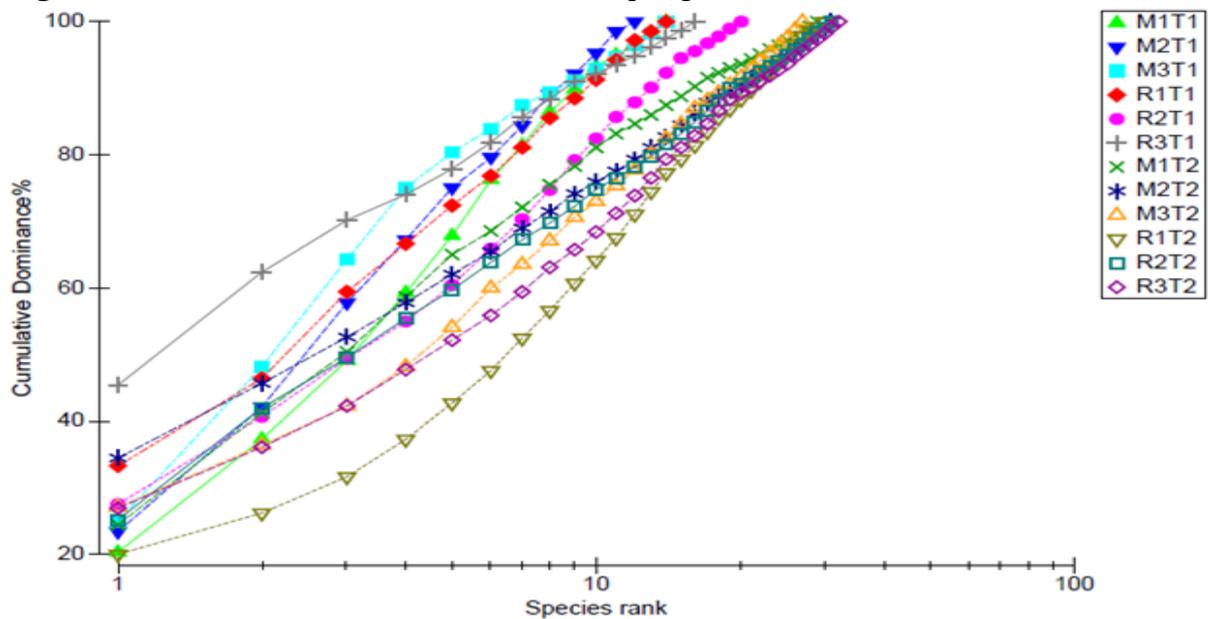
The result of identification is a family of Gastropoda and Bivalva class, with a fairly high distribution found in the two sampling locations. This is because Gastropoda and Bivalvia class can live and thrive on a wide variety of sedimentary structures, and can adapt to waters dominated by silt and sand substrate types (Putro and Muhammad, 2019b).

The k-Dominance Curves

Based on the k-dominance curves, Station R1T2 (reference area site; second sampling time) configures on the bottom of the curves, indicating that the station has the most diverse area compared to other stations (Figure 4). This is evidenced by the calculation of the Shannon-Winner index value (H') that the station R1T2 diversity index value reached 3.33. Furthermore, not only do many species make up the community, but there is also a high level of species evenness (Glover and Taylor, 2007).

Meanwhile, station R3T1 (Reference site; second sampling time) and station M3T1 (Monoculture site; first sampling time) configure on the top of curves, indicating that the station has a less diverse area, compared to other station are at the top. This indicates the dominance of a species from Anodontia sp (Bivalvia class).

Figure 4. The k-dominance curves for each sampling site.



Notes:

- aM1T1: Station of the first monoculture for the first sampling time; M2T1: Station of the second monoculture for the first sampling time;
- bM3T1: Station of the third monoculture for the first sampling time; R1T1: Station of the first reference area for the first sampling time;
- cR2T1: Station of the second reference area for the first sampling time; R3T1: Station of the third reference area for the first sampling time;
- dM1T2: Station of the first reference area for the second sampling time; M2T2: Station of the second reference area for the second sampling time;
- eM3T2: Station of the third reference area for the second sampling time; R1T2: Station of the first reference area for the second sampling time;
- fR2T2 : Station of the second reference area for the second sampling time; R3T2: Station of the third reference area for the second sampling time.

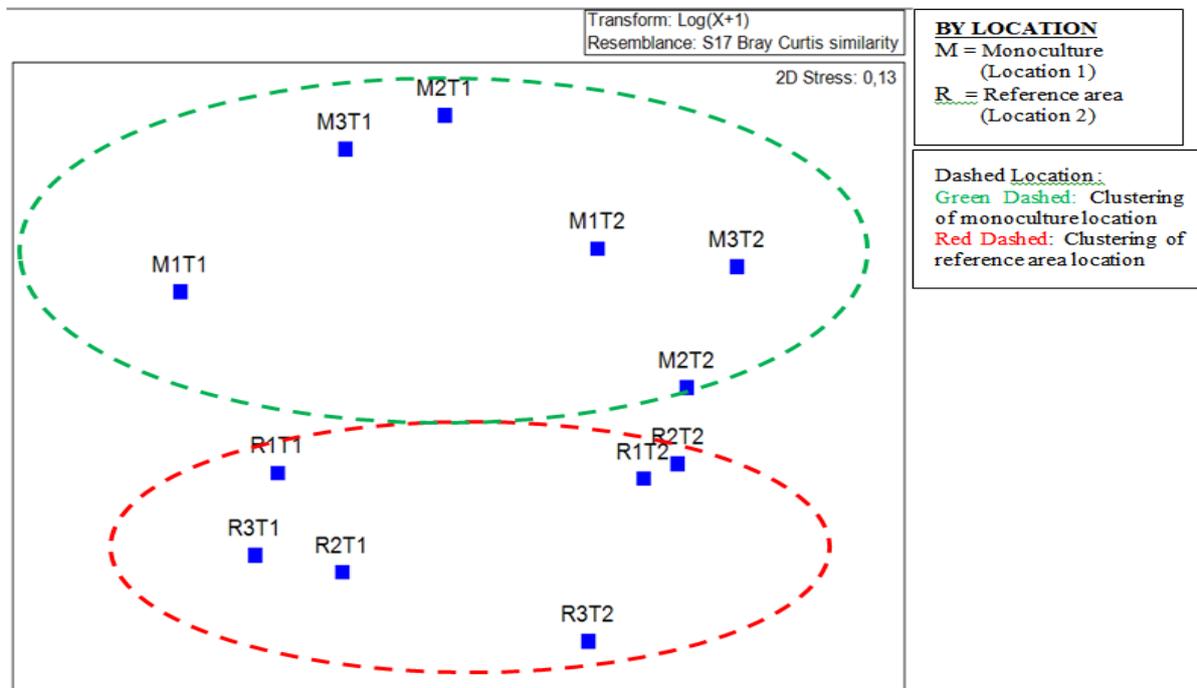
The analysis of the average composition of macrobenthic abundance showed that in the Bivalvia class, *Anodontia* sp. had a high average number at each sampling station. There was a high frequency of *Anodontia* sp. presence, and the amount distributed evenly in each sampling station, because bivalves have a wide distribution and are able to live in various types of substrate and extreme conditions. This statement is supported by Lebata et al. (2001), that bivalves, especially the Lucinidae family, have high diversity with geographic distribution from the Western Indo-Pacific to the West Atlantic waters. Substrate types are extensively distributed at each station in the research location, as sand and mud, so that they match the characteristics of their typical life.

The abundant presence of *Anodonta* sp., dominating sampling stations, can be used as a bioindicator. Bivalves can be used as a biofilter, one that filters water from which it also absorbs nutrients, including soluble pollutants in water (Lebata et al., 2001). Absorbed pollutants will accumulate in the mollusc body and may be used for bioaccumulation and bio-mineralisation assessment. The presence and abundance of certain bivalves, thus, may indicate the level of water pollution. For instance, *Anodonta* sp. stores sulfur-oxidising bacteria in gills that may be used as an indicator of environmental disturbance (Meyer et al., 2008).

Non-Metric Multidimensional Scaling (NMDS)

In Figure 5, results using Non-Metric Multidimensional Scaling (NMDS) by location indicate a tendency of grouping of stations, for each location. This implies a degree of difference in macrobenthic structure, between the location of the farmed and the reference area. This accords with (Clarke et al., 2014), who stated that macrobenthic assemblages respond spatially, because of difference characteristics of sediments and food availability that may differ between the two sampling locations.

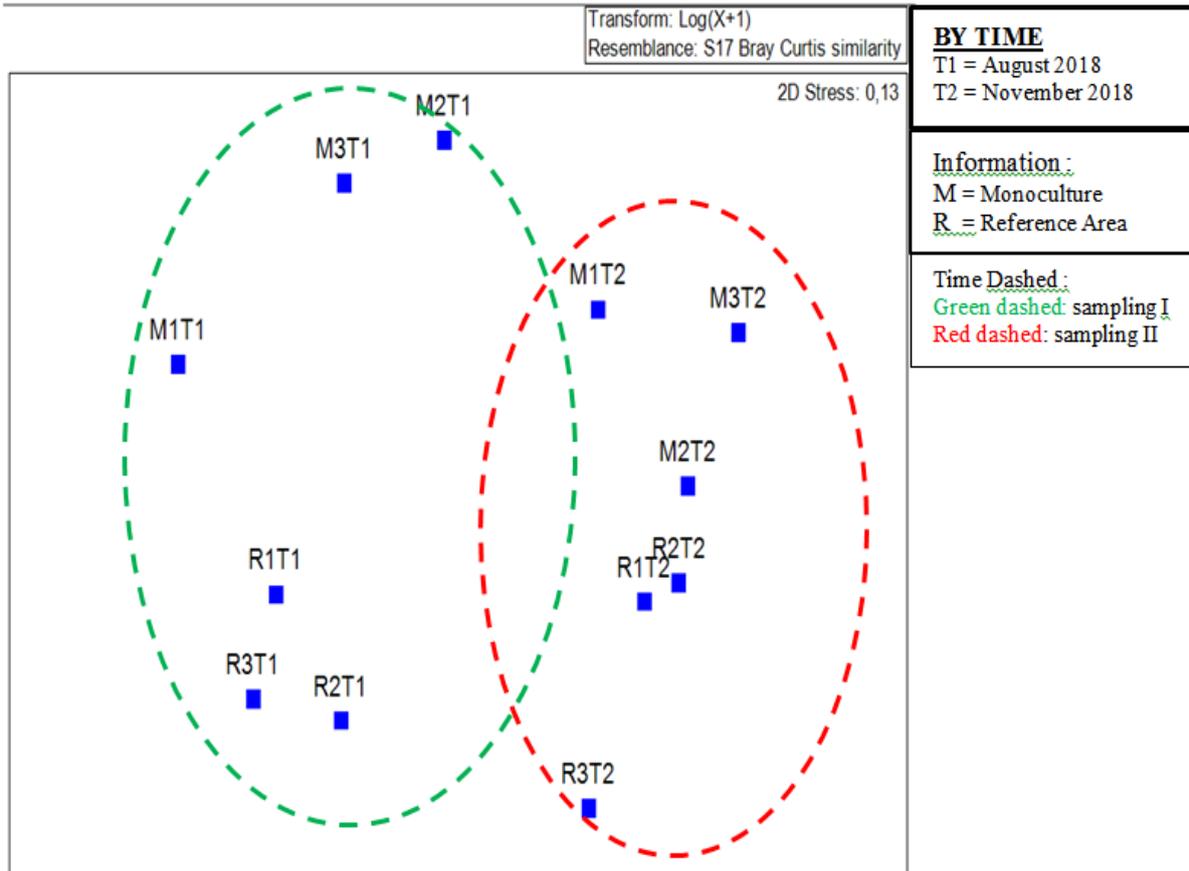
Figure 5. Ordinates of the grouping of NMDS macrobenthos by location on Menjangan Besar Island.



Non-Metric Multidimensional Scaling (NMDS) analysis indicates a difference between sampling time, which is indicated by the grouping of ordinates based on time (Figure 6).

Clustering of macrobenthic composition between stations at sampling time I (indicated by a green dashed line) and sampling II (indicated by a red dashed line) shows that macrobenthic abundance and composition at different sampling times are factors in grouping stations. This may be influenced by different physical and chemical sediment properties, at both sampling times (Clarke, 1990).

Figure 6. Ordinates of the grouping of NMDS by time on Menjangan Besar Island



One of the macrobenthos families with similarities between the location of monoculture, and the reference area, is the class Lucinidae of the species *Anodontia* sp. According to (Ambariyanto, 2011), close ordinate positions between stations illustrate that samples have similarities in species composition. Further, if the station data points in the plot are far apart, there are differences in species composition in the group. The difference in evenness of macrobenthos structure, between the two sampling locations, indicates potential environmental disturbance due to aquaculture activities (Putro, 2010).

Environmental Status of the Waters of Menjangan Besar Island

Quality in the aquatic environment can be identified by monitoring the ecosystem, including assessment in physics, chemistry, and biology (Ambariyanto. 2011). The EWS-3SWJ software calculates and weighs abiotic and biotic values in their analysis. Biotic factors are macrobenthic abundance, competition, biotic indices and abiotic factors; the results of measurements of physical and chemical factors of water and sediment. Based on analysis using the EWS-3SWJ software, the environmental status of the waters of Menjangan Besar Island was included in Moderately Disturbed Areas. Both locations were classified as moderately disturbed areas, owing to the strong current and organic nitrogen level.

Table 1: Environmental status based on analysis using EWS-3SWJ software

Station and Time to Take Research Samples	Environmental Status
M1T1	<i>Moderately Disturbed Area</i>
R2T1	<i>Moderately Disturbed Area</i>
M1T2	<i>Moderately Disturbed Area</i>
R2T2	<i>Moderately Disturbed Area</i>

aM1T1: first monoculture station in the first sampling;

bR2T1: second station reference area for the first sampling;

cM1T2: first monoculture station in the second sampling;

dR2T2: second station of the reference area on the second sampling.

Organic matter found in sediment inhabiting macrobenthic community is essential as a food source. The availability of food can affect the number of species and the abundance of animals. Benthic animals are closely related to the availability of organic matter contained in the substrate, because organic matter is a source of nutrients for biota which generally live on or in the substrate (Sharani et al., 2018). Organic matter in a substrate in a certain level is a food for macrobenthos, but if excessive, it can be toxic to benthic organisms. According to (Putro, 2007), in certain concentrations substrate organic matter is needed as a feed source for certain organisms.

Conclusion

Macrobenthic assemblages exhibited slightly differently in their abundance and proportion for each taxa, between fish farming and reference locations. Results using NMDS by location and sampling time indicated a tendency of grouping. This indicates a degree of difference in macrobenthic community structure response, spatially and temporally. The results of the k-dominance curve at the R3T1 (Reference site; second sampling time) and station M3T1 (Monoculture site; first sampling time) are at the top of the curves, which indicates the



dominance of a species from *Anodontia* sp. Both locations were classified as moderately disturbed areas, due to the strong current and moderate content in the organic nitrogen level.

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