Nutrient Enrichment Impact of Wastewater Shrimp Ponds on Coral Reefs of Nyamplungan Village, Karimunjawa

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Abstract

Increased nutrients can cause a rise prevalence in coral disease. Shrimp pond wastewater can produce nitrate, phosphate, and ammonia from the residual of the feeding and shrimp feces. Nyamplungan village, Karimunjawa was selected as the study site as it represents a location with shrimp pond activity that close to coral reef ecosystem. This study aims to examine the difference of disease prevalence, coral cover, and coral genus diversity at ± 300 m and ± 1000 m from the shrimp pond and to examine the relationships between disease prevalence and nutrients (nitrate, phosphate, ammonia). Coral disease prevalence was measured within a modified 30 x 2 m belt transect at six sampling stations. Station 1, 2, and 3 were 6-8 m in depth (± 1000 m from the shrimp pond), then station 4, 5, and 6 were depth 1-3 m (± 300 m from the shrimp pond). Coral genus diversity was conducted by using the Roving Diver Visual Method. Coral cover was measured within Line Intercept Transect (LIT) 30 m. The results show that station 5 had the highest disease prevalence ($30.80\%\pm2.78\%$). Station 1 had highest coral cover ($90.87\%\pm7.16\%$). Coral genus diversity had a medium category for each station. Disease prevalence was significantly different between station depths of 1-3 m (± 300 m to the shrimp pond) and depths 6-8 m (± 1000 m from the shrimp pond) and depths 6-8 m (± 1000 m from the shrimp pond) and monia (r= 0.975). Disease prevalence had strong relationship with nitrate (r= 0.975), phosphate (r= 0.972) and ammonia (r= 0.958). These results suggest that the continuation of coral monitoring with additional disease incidence and disease progression, temporally.

Keywords: Coral Disease, Shrimp Pond, Nutrient Enrichment, Nyamplungan, Karimunjawa.

Introduction

After Vietnam, Indonesia is one of ASEAN's second-largest shrimp pond-producing countries (FAO, 2020). Shrimp aquaculture is one of Indonesia's fisheries sectors with economic benefits (Bosman et al., 2021; Sanny et al., 2021). Shrimp export value in Indonesia reached US\$ 1.7 billion (38.7%) in 2017 (KKP, 2018). However, the process of shrimp farming activities causes the degradation of mangrove forests and harming the environment (Tarunamulia et al., 2016). The vanname shrimp pond activity in Karimunjawa began in 2016. Shrimp farming activities in Karimunjawa have caused environmental impacts on ecology in the form of pressure on marine biota (Purnomo, 2021). Nyamplungan Village was selected as the study site that represents a location with shrimp pond activity, adjacent to coral reef ecosystems.

Shrimp pond wastewater can produce nitrate, phosphate, and ammonia from the residual of the

feeding and shrimp feces (Gamal et al., 2020; Suhendar et al., 2020). Increased nutrient levels, such as nitrate and phosphate in coastal waters have been suggested as a cause of coral degradation (Rosenberg et al., 2007). The distribution of shrimp pond waste from the estuary to the coast can threaten coral reef ecosystems (Herbeck et al., 2013). Mustafa et al. (2022), reported that harvesting of a culture of shrimp caused an increase in nitrate, ammonia and a decrease in dissolved oxygen concentration in the coastal area. Increased nutrition can increase coral susceptibility to coral disease, because it interferes with the immune system of corals (Higuchi et al., 2015; Dougan et al., 2020). Samlansin et al. (2020) reported that an increase in nitrate of 0.089 mg.L-1 affects 50% of coral mortality for 96 hours. Ammonia of 0.043-0.054 mg.L-1 influenced the percentage of coral mortality exceeding 50% in 48 hours (Chawakitchareon et al., 2018). Karim (2019), reported that there were significant relationships between disease prevalence

and nitrate in coastal waters close from shrimp pond activities.

Karimunjawa National Park has 27 islands and a variety of coral reefs (Wijayanto et al., 2021). However, there is a lot of damage and degradation of coral reef ecosystems on Karimunjawa Island caused by anthropogenic factors, mostly caused by human activities such as tourism activities (Januardi et al., 2016; Farid et al., 2018; Munasik et al., 2020). Coral disease had been observed in 102 coral species in 54 different countries, with 27 diseases reported from the Caribbean and 13 from the Indo-Pacific and Red Sea In 1990 (Green and Bruckner, 2000). Coral disease outbreaks can cause significant coral mortality (Maynard et al., 2015; Walton et al., 2018). The factor decreasing water quality environment plays a very important role in the emergence of microorganisms agents that carry pathogens increasing the susceptibility of the corals to disease (Peters, 1984; Sabdono et al., 2012; D'Angelo and Wiedenmann, 2014; Messyasz et al., 2021). Environment factors such as increasing temperature, light intensity, water discharge from the mainland and increasing nutrient content may cause corals to be easily infected with diseases (Kaczmarsky et al., 2005; Kaczmarsky, 2006; Heron et al., 2010; Johan et al., 2020). Several studies have reported on coral disease prevalence in Karimunjawa. Sabdono et al. (2019) reported that the location with high nutrient level of the floating net cage in Genting Island, Karimunjawa had coral disease prevalence of 41.61± 3.93%. Nursalim et al. (2022), reported that coral disease prevalence in floating cage at Menjangan Island, Karimunjawa was (39.85%, 43.61% and 33.14%). The rapid assessment in the present study is aimed to examine the difference of disease prevalence, coral cover and coral genus diversity at depth 1-3 m (±300 m from shrimp pond) and depth 6-8 m (±1000 m from the shrimp pond) and to examine the relationships between disease prevalence and nutrient (nitrate, phosphate and ammonia) in Nyamplungan Village, Karimunjawa.

Materials and Methods

Study area and environmental parameters

The study sites were located in Nyamplungan Village, Karimunjawa Island, Jepara Regency, Central Java Province (Figure 1.). This research was conducted in 1 April 2021. Prior to investigation, reefs were selected based on ± 300 m from the shrimp ponds and ± 1000 m from the shrimp ponds. Coral disease assessment was conducted at six stations. Stations 1, station 2 and station 3 were in the depth

of 6-8 m (±1000 m from the shrimp ponds). Station 4, station 5 and station 6 were in the depth of 1-3 m (±300 m from the shrimp ponds). The coordinates and depth of the study location were shown in Figure 1. Nitrate, phosphate and ammonia concentrations in the water column were measured at the seven stations. Six stations were in coastal area and one site in waste discharge of the shrimp pond. Data was collected during high to low tide to evaluate the potential site-specific effects of sewage discharge and excess feed from waste discharge from shrimp pond. Water samples were collected from a water column by using a dark-glass bottle. Total Suspended Solid (TSS) was tested gravimetrically with the standard SNI 6869.3:2019. The total nitrate, phosphate and ammonia concentrations in the water column were measured to evaluate the potential sitespecific effects of wastewater discharge and excess feed from coastal and shrimp pond using a spectrophotometer- UV. The nitrate test was reacted with hydrochloric acid, then the intensity of the color that occurred was measured at a wavelength of 220 nm and 275 nm with a maximum of 11 ppm. Phosphate level test was measured by Vanadomolybdophosphoric Acid-Spectrophotometry. Ammonia was tested by spectrophotometer with phenate based on SNI 06-6989.30-2005.

Disease and coral genus identification

Identification of coral disease refers to Raymundo et al. (2008). All corals were identified to the genus level in situ, according to Kelley (2009). Underwater camera of Olympus TG-3 was used to take pictures for disease identification and coral genus.

Coral disease prevalence

The belt transect method was used to describe the condition of coral disease prevalence, coral cover and coral genus diversity. Modification of the length of transect line measuring of 30 meters with 2 meters width on the left and right sides, with a modified plot of 10 x 2 m (Raymundo *et al.*, 2008). Data collected directly in the field by scuba diving at each site from 1 April 2021. Coral disease prevalence was calculated according to Raymundo *et al.* (2008).

Coral genus diversity

The Roving Diver visual survey method was used to record data on species composition and abundance of all coral genus. The roving diver survey technique refers to Sabdono *et al.* (2021). The roving diver survey method involved 3 divers to observe the organisms for about 40 min to record all coral genus



Figure 1. Map of study area, Nyamplungan waters Karimunjawa.

in the maximum 8 m depth. Relative abundance coral genus and species richness were analyzed according to Sabdono *et al.* (2021). The diversity, evenness, and dominance index were analyzed with the Shannon – Wiener Index used by (Odum, 1971). Category: $H' \le 1$ = low biodiversity; $1 \le H' \le 3$ = medium biodiversity; $H' \ge 3$ = high biodiversity. Ludwig dan Reynolds (1988), stated that evenness index 0–0.5 is depressed community, 0.5–0.75 is unstable community, 0.75–1.00 is stable community.

Coral cover

The line intercept transect (LIT) method was used to determine the percentage of live coral cover, by placing the line transect in the center of the belt transect. This sampling protocol has previously been used to assess coral reef health conditions in different study areas (Sabdono *et al.*, 2014; Wijayanti *et al.*, 2014; Sabdono *et al.*, 2019). Coral coverage data was determined according to English *et al.* (1997).

Statistical analysis

One-way ANOVA analysis was used to compare coral disease prevalence and coral cover followed by Tukey's test to determine the significance of variation between the mean of each station and water depths of 1-3 m (\pm 1000 m from the shrimp ponds) by 6-8 m (\pm 300 m from the shrimp pond). Data normality and homogeneity of variance were checked before one-way ANOVA analysis. If the data was not normal, arcsine was used to transform the

data. One-way ANOVA was performed with a significance level of 5% (P<0.05). Statistical analysis was performed using SPSS version 16 computer software. Statistical analysis of correlation regression was used to gauge the relationship between coral disease prevalence and coral cover with nutrient concentrations (nitrate, phosphate and ammonia).

Results and Discussion

Environmental parameters

Environmental parameter data was presented in Table 1. Temperature data for each station ranges from 28-30°C. Station 1 had the highest temperature value (30°C), and the lowest in Station 4. The mean prevalence White Band Disease (WBD) in stations 1, 2 and 3 and stations 4, 5 and 6 were (0.70%±0.13%) and (2.60%±0.22%). Gignoux-Wolfsohn et al. (2020), reported that White Band Disease (WBD) outbreak began in July 2014 in Miami Beach, Florida (USA) when the sea surface temperatures reached 29°C. The results of the water turbidity data showed that station 1, 2 and 3 (6.2-7.4 m) was higher than station 4-6 (1.6-2.3) (Table 1.). TSS value showed that stations 4-6 (74-149 mg.L⁻¹) was higher than stations 1, 2, and 3 (32-52 mg.L⁻¹). Fonnegra et al. (2021) reported that higher turbidity and high temperature stress in the rainy season were associated with the White Plague (WP) disease prevalence. WP prevalence was only found at station 4, 5 and 6 (±300 m from the shrimp pond) (Figure 4.).

The current and wind velocity at station 1, 2, and 3 were 0.63-0.65 m.s⁻¹ and 5.5-8.3 m.s⁻¹ are higher than the current and wind velocity at station 4-6 0.12-0.23 m.s⁻¹ and 0.43-0.58 m.s⁻¹, respectively (Table 1.). The negative impact of currents was that they can deliver sediment deposits and organic and inorganic materials such as nitrates, phosphates and ammonia into the corals. According Johan *et al.* (2020) the current factor can serve as an introduction to the spread of coral disease.

The results of salinity measurements show that Stations 1, 2, 3, 4 and 6 were not much different (Table 1), while station 5 (27 ppt) had the lowest salinity value compared to other stations. This happens because station 5 was close to the estuary. Therefore, the effect of the run-off of the estuary waters causes the salinity at station 5 to be the lowest. According Dedi et al. (2016), decreased and increased salinity of 22-25 ppt can cause stress on sensitive corals. The pH value showed there was not much difference at each station from 7, 6 to 8, 0. This showed that pH value can still be tolerated for the survival of coral life. The DO value showed that stations 1, 2 and 3 (5.31-6.27 mg.L-1) had a higher value than Stations 4, 5 and 6 (2.38-3.85 mg.L⁻¹) (Table 1). Fonnegra et al. (2021), reported that lower dissolved oxygen caused higher WP prevalence.

Coral cover and disease

The observation during the present study found 5 types of coral disease Black Band Disease (BBD), White Syndrome (WS), White Band Disease (WBD), Yellow Band Disease (YBD) and Atramentous Necrosis (AtN) and 1 coral health compromiser (Pigmented Response (PR)) found at Station 1, 2, and 3. There were 7 types of coral disease (BBD, WS, WBD, Ulcerative White Spot (UWS), YBD, AtN and WP) and 2 coral health compromisers (PR and Growth Anomaly (GA) found at Station 4, 5, and 6 (Figure 2 and 3.). This showed that the type of coral disease at Station 4. 5. and 6 was higher than Station 1. 2. and 3. Previous study revealed that there were 7 types of coral diseases found on Menjangan Besar Island Karimunjawa, namely BBD, Brown Band Disease, UWS, WS, YBD, WP dan WBD (Nursalim et al., 2022).

Prevalence of coral disease and coral health compromiser types were shown in Figure 4. WS had the highest prevalence at depth 6-8 m (2.31%). Palupi *et al.* (2018), reported that WS was the main disease prevalence in Kessilampe Waters, Kendari, South East Sulawesi. WS was also found on Karimunjawa, Kemujan, Menjangan Besar, Tengah and Seruni Island (Sabdono *et al.*, 2019). BBD had the highest prevalence at depth 1-3 m (8.63%). Sabdono *et al.* (2019) also reported that the presence of BBD coral disease was mostly found in waters close to floating net cage activities that produce nutrient waste. Accordance to the statement Delpopi *et al.* (2015), BBD contributed to the decrease in coral cover of scleractinia.

The different values of coral disease prevalence are shown at (Figure 5.). Station 5 had highest disease prevalence ($30.80\%\pm2.78$). Figure 5 also showed the value of coral cover. Station 4 had lowest coral cover ($44.57\%\pm2.27$). These results of these data indicated that the lower the coral disease prevalence, the higher the coral cover value. Miller et *al.* (2015) stated that the lower coral covers make corals more susceptible to diseases. Station 5 had a highest value of disease prevalence. However, this prevalence was lower than previous studies conducted under the marine floating cages at Genting and Menjangan Besar Island, Karimunjawa (Sabdono *et al.*, 2019; Nursalim *et al.*, 2022).

One-way ANOVA showed that there was a significant difference (P<0.05) in coral disease prevalence and coral cover between depth 1-3 m (±300 m from the shrimp ponds) and 6-8 m (±1000 m from the shrimp ponds). Based on Figure 7, there was a strong relationship between the coral disease prevalence and nitrate (r= 0.975), phosphate (r= 0.946) and ammonia (r= 0.9172) concentration. Karim (2019) reported that there was a significant relationship between the value of nitrate on the type of coral disease UWS and WS prevalence in coastal waters close to shrimp pond activities. High nutrient values can trigger corals to become more susceptible to disease because it interferes with the coral's immune system (Higuchi et al., 2015; Dougan et al., 2020). Johan et al. (2020) stated that high level nutrient was a potential factor supporting the onset of coral disease. Nutrient wastewater from shrimp ponds can cause significant growth of Cyanobacteria, Alphaproteobacteria and Vibrio sp (Becker et al., 2017; Riandi et al., 2021). Pathogenic microorganisms Cyanobacteria and Vibro sp were identified in corals infected BBD and WS (Muller et al., 2017; Agung et al., 2020; Khodzori et al., 2021). Therefore, BBD and WS disease types had a high prevalence at Stations 4, 5 and 6 (Figure 4.). Sabdono et al. (2019), reported that there was a difference in coral disease prevalence between marine floating cage locations and the control locations.

Coral genus abundance and diversity

The present work revealed that there were 30 coral genus found in Nyamplungan Waters, Karimunjawa (Table 2). The highest and lowest number of species richness was at Station 3 (20 genus) and Station 5 (12 genus) (Table 2). Acropora had the highest relative abundance at Station 1 (21.55%) and Station 2 (13.15%). Barus *et al.* (2018) stated that Acropora was one of the coral genera that

St	Temperature (°C)	Turbidity (m)	Cui (m V	rrent I.S ⁻¹) D	V (n V	Vind n.s ⁻¹) D	TSS (mg.L ⁻¹)	Salinity (ppt)	рН	D0 (mg.L ⁻¹)
1	30	7.4	0.65	East	8.3	East	32	31	7.8	6.27
2	30	6.2	0.60	East	5.5	East	52	30	7.9	5.7
3	30	6.6	0.63	East	6.4	East	48	31	7.9	5.31
4	29	1.8	0.23	West	5.2	West	74	29	8.0	3.84
5	30	1.6	0.24	North	4.3	North	149	27	7.6	2.38
6	28	2.3	0.12	West	5.8	West	78	31	7.8	3.34

Table 1. Environmental parameters along the sampling sites station.

Note: V= Velocity, D= Direction, DO= Dissolved oxygen



Figure 2. Coral disease states present (A and B= Black Band Disease (BBD), C and D = White Syndrome (WS); E= Yellow Band Disease (YBD); F= White Plague (WP); G= Ulcerative White Spot (UWS); H= White Band Disease (WBD); I= Atramentous Necrosis (AtN).



Figure 3. Coral health compromiser states present (A = Growth Anomaly (GA), B = Pigmented Response (PR); C= Fish Bites; D= Macroalgae Competition).



Figure 4. Coral disease and health compromiser prevalence at the depth of 6-8 m (±1000 m from shrimp pond) and depth 1-3 m (±300 m from shrimp pond).









was often found in waters with moderate to low nutrients level. *Porites* genus had highest relative abundance values at Station 3 (14.29%), Station 4 (21.15%), Station 5 (21.97%) and Station 6 (25.58%). *Porites* can survive, even in high sedimentation conditions (Aldyza and Afkar, 2015). Solihuddin *et al.* (2019) reported that *Porites* had a high coral cover value on Karimunjawa Island. Station 4, Station 5 and Station 6 had a TSS value above the quality standard for coral reef growth of 20 mg.L⁻¹ (Table 1.). Ali *et al.* (2020) also reported that the genus *Porites* is able to survive in waters with high nutrient values.

Coral genus diversity, evenness and dominance data were shown in Table 2. The coral genus diversity index at all stations were in the medium category. This is because of the high relative abundance of certain coral genus at each station. Coral genus diversity index is influenced by the coral genus relative abundance that occupies each station (Ekel *et al.*, 2021). The evenness index category showed that station 1, 2, 3, 5, and 6 were classified as stable community. However, station 4 had lowest category evenness index (0.45 ± 0.09). This is showed that corals of station 4 was classified as depressed community.

Nutrient and coral prevalence and coral cover correlation

The wastewater shrimp pond nutrients value in Nyamplungan Village were nitrate (5.144 mg.L-1), phosphate (0.256 mg.L-1) and ammonia (5.971 mg.L-1). This values exceeded the quality standard for marine biota (KEPMEN LH, 2004). It can be impact to corals because the results of this nutrients shrimp pond wastewater can flow to coastal area.

Table 2. Relative abundance of coral genus at Nyamplungan waters.

Genus	Relative abundance (%)									
	Station 1	Station 2	Station 3	Station 4	Station 5	Station 6				
Acropora	21.55	13.15	11.37	15.11	8	11.92				
Porites	14.06	11.15	14.29	21.15	21.97	25.58				
Leptoria	0	0	3.06	0	0	0				
Helliopora	7.1	5.01	4.81	0	0	0				
Turbinaria	3.61	10.01	4.96	4.23	7.32	3.2				
Montastrea	4.52	0	4.08	6.34	7.32	4.94				
Leptastrea	0	0	0.87	0	0	2.91				
Echinopora	4.52	4.55	2.48	3.32	10.99	2.33				
Pachyseris	4.26	6.48	6.56	6.95	9.58	0				
Favites	3.87	0	2.62	0	0	3.2				
Millepora	0	6.94	5.69	9.67	0	3.78				
Montipora	5.03	7.74	9.91	9.37	9.86	13.95				
Oxypora	0	7.62	5.54	0	0	0				
Pocillopora	2.46	2.73	5.1	2.11	2.54	6.1				
Leptoseris	0	0	1.17	0	0	0				
Fungia	2.97	3.53	2.33	5.44	3.66	3.49				
Favia	2.58	0	3.79	0	0	3.78				
Psammocora	0	2.39	4.52	0	0	0				
Pavona	13.03	10.47	5.54	0	10.7	4.65				
Euphyllia	2.97	0	1.31	3.32	0	0				
Diploastrea	5.29	1.25	0	0	0	0				
Plerogyra	2.19	1.86	0	8.46	2.25	0				
Astreopora	0	2.96	0	0	0	0				
Stylophora	0	1.14	0	1.21	0	0				
Gorgonia	0	1.02	0	0	0	0				
Caulastrea	0	0	0	3.32	0	0				
Goniastrea	0	0	0	0	5.63	0				
Pseudosiderastrea	0	0	0	0	0	3.2				
Siderastrea	0	0	0	0	0	3.49				
Platygyra	0	0	0	0	0	3.49				
Species richness	16 genus	18 genus	20 genus	14 genus	12 genus	16 genus				
Diversity index	2.17	2.52	2.58	1.11	2.12	2.31				
Evenness	0.78	0.92	0.92	0.45	0.94	0.89				
Dominance index	0.19	0.09	0.09	0.01	0.14	0.13				



Figure 7. The relationship between disease prevalence and nitrate, phosphate and ammonia concentration.



Figure 8. The relationship between coral cover and nitrate, phosphate and ammonia concentration.

Mustafa et al. (2022) reported that nutrients (nitrate, phosphate and ammonia) level at coastal area that close to shrimp pond activity were higher than control area. The correlation between coral disease prevalence with nitrate, phosphate and ammonia was shown in Figure 7. The correlation regression analysis between coral disease prevalence and nitrate, phosphate and ammonia concentration showed r value for that nitrate, phosphate, and ammonia of 0.975; 0.9772; 0.958, respectively. This showed that there were strong-positive relationships between the coral disease prevalence and nitrate, phosphate and ammonia. Kaczmarsky and Richardson (2011) states that an increase in nitrate and phosphate concentrations will cause higher coral disease. Siladharma and Karim (2017), reported that coral

disease prevalence had strong relationship with TSS and nitrate. This was confirmed by *Dougan et al.* (2020) that an increase in nutrients can lead to growth of coral disease. However, Sabdono *et al.* (2019) reported that there was no positive correlation between nitrate and phosphate concentrations and the prevalence of coral disease under floating net cages.

The correlation between coral cover with nitrate, phosphate and ammonia were shown in Figure 8. It showed a strong-positive effect of coral cover to nitrate, phosphate, and ammonia with r-values of 0.9860; 0.987 and 0.980, respectively. (Figure 6). Guo *et al.* (2019), reported that an increase in nutrients will cause the high value of

macroalgae cover, but lower the coral cover. There was macroalgae competition on corals at Station 5 (Figure 3). However, it was found outside the belt transect. Vaughan *et al.* (2021) reported that high nutrient values will lead to a total loss of coral cover and an increase in macroalgae. However, the mean of coral cover had moderate category at station with depth 1-3 m (\pm 300 m from shrimp pond).

Conclusion

Coral disease prevalence in the station with depth of 1-3 m (±300 m from the shrimp pond) was higher than the station with the depth of 6-8 m (±1000 m from the shrimp ponds), but in the contrary with coral cover. There was significant differences disease prevalence and coral cover within depth of 6-8 m (±1000 m from the shrimp ponds) and depth of 1-3 m (±300 m from the shrimp ponds). There was no different coral genus diversity index (H') category of all station. The results showed there is a strong between nitrate. relationship phosphate and with ammonia concentrations coral disease prevalence and coral cover.

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