Organism Associated with *Cymodocea Serulata* in Different Habitats near Urban Coastal Area

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Abstract. The development of tourism in Bali is very rapid, where most area are in direct contact with residential areas. Seagrass that grows in coastal area is under pressure due to tourism activities. This study aims to determine the state of urban coastal areas with an indicator of organisms associated with seagrass *Cymodocea serulata*. The samples of *C. serulata* leaves were collected from Sanur Beach, Denpasar and Samuh Beach, Nusa Dua, Badung, Bali. These two areas are tourism areas with high tourist visits, and are located near the residential areas. Sanur Beach is different from Samuh Beach, where the seagrass habitats at Sanur Beach are also used as a mooring place for motorized boats, crossing Penida strait from Sanur to Nusa Penida and Nusa Lembongan. The results showed that periphyton that lived in association with *C. serulata* were higher at Samuh beach (12 species) compared to Sanur beach (3 species). However, the total bacteria associated with seagrass leaves was higher at Sanur beach (58.0 x 10⁴ CFU / mL) compared to that of Samuh beach (51.3 x 10⁴ CFU / mL). Three species of periphyton with the high number of individuals found on *C. serulata* leaves at Samuh beach were *Ardissonia fulgens*, *Synedra* sp, and *Thalassionema nitzschioides*, while 3 species were found at the Sanur beach were *Thalassiothrix* sp, *Oscillatoria* sp., and *Paracalanus palvus*. Four potential fungal species were found associated with seagrass leaves, 3 of them identified as *Aspergillus eff. niger*, *A. clavatus* and *Penicillium purpurogenum*, while 1 isolate has not been identified. The results of this study found that habitat differences and water designation affected the diversity of organisms associated with seagrass beds of *C. serulata*.

Keywords: urban area, tourism, *Cymodocea serulata*, periphyton

1. Introduction

The abundance and diversity of organisms depends on environmental condition. The sensitivity of the communities to the alteration of the environment and their tolerance to environmental pressure has been considered as a measure of ecosystem health [1,2]. The quality degradation of the environment can
be caused by natural and anthropogenic activities. Human activities such tourism industries have important contribution to the quality of environment. Increasing tourism industries along the coastal area are, direct or indirectly contributed to the quality to the surrounding environment, exposed the fragile ecosystems to environmental degradation. Uncontrolled tourism development in the coastal brings negative impact to the area due to overcrowding, poor sewage disposal, boat-generated waste, beach erosion and destruction of wildlife habitats [3].

Sanur beach at Sanur and Samuh beach at Nusa Dua are famous as tourist destination at Bali Island. Sanur beach is differ to the Samuh beach, which Sanur beach, besides as tourist destination, also the area was functioned as harbour of boats powered by large machines to cross Penida Strait from Sanur to Nusa Penida island. Samuh beach, on the other hand, only functioning as tourist destination and harboured by few traditional fishing boat. Oil spill from boats is inevitable affecting the abundance and diversity of macro- and micro invertebrate living in those two areas. It has been found that oil spills contain polycyclic aromatic hydrocarbons (PAHs) are widely spread in marine ecosystems, correspond to acute pollution and other human activities [4]. Sanur and Samuh beaches are the habitat of seagrasses, and 7 species of seagrasses were identified at Sanut beach, one of them was Cymodocea serulata [5]. This species was also found at Samuh beach.

Seagrasses are habitats for coastal living organisms such as fish and other organisms associated with them. The leaves of the seagrasses is a unique habitat for microorganism, with specific association among diverse aquatic microorganisms are capable of colonizing surfaces of various kinds, leading to the formation of biofilms [6]. This unique association provides microorganisms a better access to nutritional resources, enhanced organism interactions, and stability. Periphyton is the aquatic biota that develops on submerged substrates when nutrients and light are available, formed assemblages, attached on any wet substrate [7]. Alteration of coastal environment due to anthropogenic activities on the coastal area and its affect to the diversity of organism attached to seagrasses in tropical area has not much studied. The limitation of information about the diversity of organism that play roles to the alteration of environment, particularly in tropical with the impact of human activities was the aim of this study.

2. Methods

2.1. Sample collection

Samples of leaves of Cymodocea serulata contained organism were collected from 2 sites: Sanur Beach and Samuh Beach. Samur beach located at Sanur, one of tourist destination and boat harbour to Nusa Penida Island, a small island located at the east of Sanur. Samuh beach is another tourist destination, located at Nusa Dua, but this site was not used as a bat harbour. Samples were preserved in 70% alcohol until processing.

2.2. Sample processing and analysis

The leaves samples of C. serulata that have been collected were brought to the Laboratory of Aquatic Resource Management, Faculty of Marine and Fishery, Udayana University. Leaves were grazed using a scalpel to collect the organism attached to the leaves. Then, they observed and identified under light microscope. Identifications of organism were based on “Encyclopedia of Marine Gastropods by Alain Robin, 2008”. Bacteria and fungi identification were conducted at Laboratory on Microbiology, Faculty of Mathematics and Science, Udayana University. Bacteria isolation was done using plating methods following Nester et al. [8]. Preparation of samples for fungi identification was following Pfaffer [9]. Segments from leaves of C. serulata were plated on potato dextrose agar medium amended with an antibiotic (chloramphenicol 150 mg l-1) in Petri dishes and incubated in a light chamber for four weeks at 26°C; the light regimen was 12 h of light followed by 12 h of darkness. The Petri dishes were observed periodically, and fungi that grew out from the tissues were isolated and identified based on spore morphology.
3. Result and Discussion

3.1. Results

3.1.1. Periphyton diversity.

The result shows that the diversity of periphyton at Sanur beach were much lower than that of Samuh beach. There were only 3 species periphyton found at Sanur beach with the average number ranged from 0.33 to 1.33 individual per collection (Figure 1).

![Figure 1. The average number of periphyton species collected from the leaves of *C. serulata* at Sanur beach.](image1)

3.1.2. Fungi and bacteria diversity.

At Samuh beach, on the other hand, 13 species were identified (Figure 2). The most common species found at Samuh beach was Ardissonia fulgens, with the average number of 48.33. Two other species that were commonly found at Samuh beach were Synedra sp with the average number of 29.67 and Thalassionema nitzschioides with the average number of 29.33 (Figure 2a). The other species have the average number range from 0.33 to 2.33 (Figure 2b).

![Figure 2. The average of three most commonly species (2a) and 10 other species of periphyton collected from the eaves of *C. serulata* at Samuh beach.](image2)

The species of fungi isolated from the leaves of *C. serulata* were slightly differ between sites (Sanur and Samuh beaches). Five species of fungi were isolated from Samuh beach, while 4 from Sanur beach. *Apergillus niger* was isolated from both sites at all collections. *Penicillium purpurogenum* and *Aspergillus clavatus* were also collected from both sites, but only *P. Purpurogenum* was collected from all collection from Samuh beach. *Aspergillus* sp 1 and *Aspergillus* sp 2 were not found on the leaves of *C. serulata* collected from Sanur beach, but present from the samples from Samuh beach. On the other hand, the Isolate 1 was only present at the sample from Sanur beach (Table 1).
Diverse aquatic microorganisms are capable of colonizing surfaces of various kinds, leading to the formation of biofilms and to the development of specialized processes within these structures. The organisms in biofilms create their own microhabitats. Such as microalgae, bacteria, fungi, protozoa, metazoa and epiphytes form periphyton biofilms. The contribution of heterotrophic, autotrophic and detritus within the organic pools is reflection of the function of microbial assembles to the biomass, particularly the C/N ratios [12]. Prokaryotic assembles was found as indicator of the ecosystem health by measuring its diversity and activity related to ecological quality [2,10]. The abundance and diversity of organisms and sensitivity of community to the alteration of the environment and their tolerance to environmental pressure has been considered as a measure of ecosystem health [1,2].

Periphyton that were found in less disturbed area at Samuh beach was higher compared to Sanur beach, indicating the pressure of environmental affecting the abundance of periphyton. Sanur beach that received more disturbance support much less periphyton population than Samuh beach. Oscillatoria sp. was the only species of periphyton found at both sites, indicating this species are adapted to a wide range of environmental condition. Complex communities of autotrophic and heterotrophic microorganisms such as microalgae, bacteria, fungi, protozoa, metazoa and epiphytes form periphyton biofilms [11]. The organisms in biofilms create their own microhabitats.

Microbial parameters and processes have been identified to have abilities to detect changes in environmental quality [2]. The structure and function of natural marine ecosystem is a concern for managing the impacts of human activities. Marine microorganism are believed to contribute to present of particulate organic matter in the ocean and drive global biogeochemical cycles [2,10]. The abundance and diversity of organisms and sensitivity of community to the alteration of the environment and their tolerance to environmental pressure has been considered as a measure of ecosystem health [1,2].

There is an increased awareness that marine environments are important roles in global climate changes. The structure and function of natural marine ecosystem is a concern for managing the impacts of human activities. Marine microorganism are believed to contribute to present of particulate organic matter in the ocean and drive global biogeochemical cycles [2,10]. The abundance and diversity of organisms and sensitivity of community to the alteration of the environment and their tolerance to environmental pressure has been considered as a measure of ecosystem health [1,2].

Discusion

Two species of bacteria were isolated from the leaves of C. serulata, namely Sterptococcus sp and Bacillus sp. The volume of both species of bacteria were much higher at Sanur beach (58 x 104 CFU/mL) compared to Samuh beach (51.3 x 104 CFU/mL).

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Table 1. A Species of fungi identified from the leaves of Cymodoce serulata collected from Sanur and Samuh beaches.

| No. | Species/collection number | Sanur Beach | Samuh Beach |
|-----|----------------------------|-------------|-------------|
| 1   | Aspergillus niger          | +           | +           | +           |
| 2   | Aspergillus sp. 1          | -           | -           | +           |
| 3   | Aspergillus sp. 2          | -           | -           | +           |
| 4   | Penicillium purpurogenum   | +           | -           | +           |
| 5   | Aspergillus clavatus       | +           | -           | +           |
| 6   | Isolate 1                  | +           | +           | -           |

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Marine environment is highly affected by the impact of anthropogenic activities. Increasing in development of tourism industries alongside the beaches have a huge impact to the marine environment. Boats powered by machines are increasingly used to transport people from one island to the other. Sanur beach, Bali, as one famous tourist destination is also a place to harbour boats powered by large machines to cross the Penida strait to Penida and Lembongan Island. Oil spills from the boat machines is inevitable. Oil spills contain polycyclic aromatic hydrocarbons (PAHs) are widely spread in marine ecosystems, correspond to acute pollution and other human activities [4]. Human activities have increase the level of PAHs in the environment, particularly in marine ecosystem raising serious environmental issues and human health concerns Some PAHs and their metabolites are considered highly toxic mutagens and carcinogens [4].

4. Conclusion
The diversity of periphyton and fungi were different between Sanur and Samuh beaches. It has been found that the diversity of periphyton and fungi were found higher at Samuh beach, where the beach was not harboured by the boat. The diversity of bacteria was not differ between sites. Oscillatoria sp. was the only species of periphyton found at both sites.

5. Acknowledgment
This article was presented at the 2nd International Conference on Smart City Innovation (ICSCI) 2019, jointly held by Universitas Indonesia and Universitas Diponegoro. ICSCI conferences have been supported by the United States Agency for International Development (USAID) through the Sustainable Higher Education Research Alliance (SHERA) Project for Universitas Indonesia’s Scientific Modelling, Application, Research, and Training for City-centered Innovation and Technology (SMART CITY) Center for Collaborative Research, administered through Grant #AID-497-A-1600004, Sub Grant #IIE-00000078-UI-1

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