Similarity microalgal epiphyte composition on seagrass of *Enhalus acoroides* and *Thalasia hemprichii* from different waters

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Similarity microalgal epiphyte composition on seagrass of *Enhalus acoroides* and *Thalassia hemprichii* from different waters

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Abstract. The epiphytes are all autotrophic organisms that are permanently attached to rhizomes, roots, and leaves of seagrasses. The epiphyte is an important primary producer for the seagrass ecosystem and contributes significantly to the food chain. This study aims to identify the composition of microepiphyte algae on *Enhalus acoroides* and *Thalassia hemprichii* and their similarity levels of both compositions. The 20 leaves samples of *E. acoroides* and *T. hemprichii* were observed. The epiphytic microalgae which found on the surface of the seagrass leaves were scrapped, collected in a bottle sample, and fixed with 70% alcohol and identified into genera. The relation of epiphytic microalgal genera to the location and species of seagrass was analyzed using similarity analysis. The Chrysophyta, Cyanophyta, and Chlorophyta epiphytic microalgae were found. There were similarity variations of the microalgal epiphyte in seagrass of *E. acoroides* and *T. hemprichii* and seagrass habitat sites. Morphology and seagrass life affects the abundance and diversity of the epiphytic microalgal attached to the seagrass and it may be associated with the epiphytic lifetime in the seagrass.

1. Introduction

Seagrasses are a major functioning element resulting high productivity of tropical marine regions. They occur in the form of multispecific beds (constructed by more than one species of seagrass) or monospecific (constructed by only one species of seagrass) [1]. The presence of seagrasses provide habitat and consequently, increase the abundance and diversity of marine organisms [2, 3], such as epiphytes that live on leaves and stems of seagrasses. Selection by organisms to seagrass habitat may be influenced by several factors including habitat structures which leads to differences in habitat suitability, food availability and protection provided [4].

Epiphytes are the foremost component of majority seagrass ecosystems. They are also the primary food resource for grazers. Microalgal epiphyte was the most abundant and diverse found in seagrass, especially seagrass leaves [5]. The epiphyte abundance and species composition are strongly influenced by the seagrass species which is related to the variation of leaf turnover of different species of seagrass [6]. Faster leaf turnovers limit the time for colonization of epiphytes compared to the slower ones. The size of seagrasses and the rate of seagrass turnover are important in determining loads of epiphytic organisms attached to them [5]. Therefore, the difference of seagrass bed concerning the number of species (mono or multispecific) will influence the diversity and biomass of
epiphyte attached to seagrass leaves in such meadows. The studies on ephypite of the seagrass mostly were macroalgal [7, 8], unspecified epiphytic algae [9] or microalgal ephypite on certain species of seagrass, such as Halodule wrightii [10], Cymodoaceae serrulata [11]. Comparing ephypitic microalgae on two different species has only conducted in Posidonia oceanica and Cymodocea nodosa [12]. Therefore this study aims to identify the composition of microepiphyte algal live on the seagrass of Enhalus acoroides and Thalassia hemprichii and their similarity levels of both compositions.

2. Materials and Methods
The study was performed in June 2016 at Bandengan (06°33.905'S, 110°39.315'E, and Ujungpiring (06°30'74,0"S, 110°40'18,4" E) Waters of Jepara Regency. E. acoroides and T. hemprichii are the most dominated seagrass species in those study area [13, 14, 15].

Ephypite microalgal was scraped from the seagrass leaves which came from two study sites. A total of twenty leaves of E. acoroides and T. hemprichii were cut at the base of the leaves, stored in a sample bag sealed filled with seawater and placed into cool-box and taken to the laboratory for observation. The leaves were cut and categorized (tip, middle, and base of the leaves). The epiphytic microalgae found on the surface of the leaves portion was carefully scrapped, collected in a bottle sample, fixed with 70% alcohol and identified into species according to [16]. The relation of epiphytic microalgal species to the location and species of seagrass was analyzed using Sørensen similarity analysis [17]. The similarity is high when the value more than 50% and vice versa.

\[
Sørensen\ similarity\ (S) = \frac{2a}{2a+b+c}
\]

a = the number of species shared by both samples (fraction a)
b and c = numbers of species occurring only in the first and only in the second sample, respectively

3. Results and Discussion
The seagrass species observed in present work are E. acoroides and T. hemprichii which live on the intertidal coast with sandy to coarse rubble substrates. A total of 32 genera of ephypite microalgal found on the leaves of two species of seagrasses, i.e., 25 and 21 genera found on the leaves of E. acoroides and T. hemprichii respectively from Bandengan waters and while the leaves of E. acoroides and T. hemprichii from Ujungpiring waters provide habitat for 26 and 21 genera of microalga ephypite (table 1, 2). Among the genera found in both location, the highest genera number belong to Chrysophyta Division, followed by Chlorophyta and Cyanophyta.

It is shown in table 3, that almost all comparison have high similarity index (SI > 50 %) with the value in the range of 51,43~80,95 %. There were only 4 value with high dissimilarities, such as TBU (Basal of T. hemprichii leaves of Ujungpiring Waters)-TMU (Middle of T. hemprichii leaves of Ujungpiring Waters) (50 %), TBB (Basal of T. hemprichii leaves of Bandengan Waters) -EMB (Middle of E. acoroides leaves of Bandengan Waters) (48,48 %), TTU (Tip of T. hemprichii leaves of Ujungpiring Waters)-EMB (Middle of E. acoroides leaves of Ujungpiring Waters) (47,37 %) and TBU (Basal of T. hemprichii leaves of Ujungpiring Waters)-EMU (Middle of E. acoroides leaves of Ujungpiring Waters) (45,16 %). The water quality of both study sites was presented in figure 1 and table 4.
Table 1. “Presence (+)-absence(-)” genera list of epiphytic microalgae identified on the leaves of *E. acoroides* and of *T. hemprichii* in Bandengan waters of Jepara.

| No.  | Genera               | *Enhalus acoroides* |          |              | *Thalassia hemprichii* |          |              |
|------|----------------------|---------------------|----------|--------------|------------------------|----------|--------------|
|      |                      | Tip of the leaves   | Middle of the leaves | Base of the leaves | Tip of the leaves   | Middle of the leaves | Base of the leaves |
| 1    | Achananthes         | +                    | +         | -            | +                      | +        | +            |
| 2    | Amphora             | +                    | +         | -            | +                      | +        | +            |
| 3    | Ardissonea          | +                    | +         | -            | -                      | +        | +            |
| 4    | berkeleya           | +                    | +         | +            | +                      | +        | +            |
| 5    | Cocconeis           | +                    | +         | +            | +                      | +        | +            |
| 6    | Coscinodiscus       | +                    | +         | +            | +                      | +        | +            |
| 7    | Diatomeae           | +                    | +         | +            | -                      | -        | -            |
| 8    | Diplonesi           | +                    | +         | +            | -                      | +        | -            |
| 9    | Gomphonema          | -                    | +         | -            | -                      | +        | -            |
| 10   | rammatiophora       | +                    | +         | -            | -                      | -        | -            |
| 11   | Gyrosigma           | +                    | +         | +            | +                      | +        | -            |
| 12   | Hyalodiscus         | +                    | +         | +            | +                      | +        | -            |
| 13   | Hyalosira           | +                    | +         | +            | -                      | -        | -            |
| 14   | Leptocylindrus      | -                    | -         | +            | +                      | +        | +            |
| 15   | Licomorpha          | +                    | +         | +            | -                      | -        | -            |
| 16   | Melosira            | +                    | +         | +            | +                      | +        | +            |
| 17   | Navicula            | +                    | -         | +            | +                      | +        | +            |
| 18   | Neosynedra          | -                    | +         | -            | -                      | -        | -            |
| 19   | Nitzschia           | -                    | -         | -            | +                      | +        | +            |
| 20   | Pleurosigma         | -                    | +         | +            | -                      | -        | -            |
| 21   | Rhizosolenia        | +                    | +         | -            | -                      | -        | -            |
| 22   | Synedra             | -                    | -         | -            | -                      | -        | -            |
| 23   | Thalassionema       | -                    | -         | -            | -                      | -        | +            |
| 24   | Thalassiothrix      | -                    | -         | +            | -                      | -        | -            |
|      | **Division Chlorophyta** |                          |          |              |                          |          |              |
| 1    | Atractomorpha       | +                    | +         | +            | +                      | +        | +            |
| 2    | Characium           | +                    | -         | -            | -                      | -        | -            |
| 3    | Stigeoclonium       | +                    | -         | +            | -                      | +        | -            |
|      | **Division Cyanophyta** |                          |          |              |                          |          |              |
| 1    | Anabaenopsis        | -                    | +         | -            | -                      | -        | -            |
| 2    | Merismopedia        | +                    | +         | -            | -                      | +        | -            |
| 3    | Microcystis         | +                    | -         | -            | +                      | +        | -            |
| 4    | Planktolyngbia      | -                    | -         | -            | +                      | +        | -            |
| 5    | Trichodesmium       | -                    | -         | -            | -                      | -        | -            |
Table 2. “Presence (+)-absence (-)” genera list of epiphytic microalgae identified on the leaves of *E. acoroides* and of *T. hemprichii* in Ujungpiring waters of Jepara.

| No. | Genera              | *Enhalus acoroides* | *Thalassia hemprichii* |
|-----|---------------------|--------------------|------------------------|
|     |                     | Tip of the leaves  | Middle of the leaves   | Base of the leaves | Tip of the leaves | Middle of the leaves | Base of the leaves |
| 1   | Achnanthes          | +                  | +                      | +                    | +                   | +                    | +                    |
| 2   | Amphora             | -                  | -                      | -                    | +                    | +                    | +                    |
| 3   | Ardissonea          | +                  | +                      | +                    | +                    | +                    | +                    |
| 4   | Berkeleya           | +                  | +                      | -                    | +                    | +                    | +                    |
| 5   | Cocconeis           | +                  | +                      | +                    | +                    | +                    | +                    |
| 6   | Coscinodiscus       | +                  | +                      | +                    | +                    | +                    | +                    |
| 7   | Diatomae            | +                  | +                      | +                    | +                    | +                    | -                    |
| 8   | Diplonesi           | +                  | +                      | +                    | +                    | +                    | -                    |
| 9   | Gomphonema          | -                  | +                      | -                    | -                    | -                    | -                    |
| 10  | Grammatiophora      | +                  | +                      | -                    | -                    | -                    | -                    |
| 11  | Gyrosigma           | +                  | +                      | +                    | +                    | +                    | +                    |
| 12  | Hyalodiscus         | -                  | +                      | +                    | +                    | +                    | -                    |
| 13  | Hyalosira           | +                  | +                      | +                    | +                    | +                    | -                    |
| 14  | Leptocylindrus      | +                  | +                      | +                    | +                    | +                    | +                    |
| 15  | Licomorpha          | +                  | +                      | +                    | +                    | +                    | -                    |
| 16  | Melosira            | +                  | -                      | +                    | +                    | -                    | +                    |
| 17  | Navicula            | +                  | -                      | +                    | +                    | -                    | -                    |
| 18  | Neosynedra          | -                  | -                      | -                    | -                    | -                    | -                    |
| 19  | Nitzschia           | +                  | -                      | +                    | +                    | +                    | -                    |
| 20  | Pleurosigma         | +                  | +                      | +                    | +                    | +                    | -                    |
| 21  | Rhizosolenia        | +                  | -                      | +                    | +                    | -                    | +                    |
| 22  | Synedra             | -                  | +                      | -                    | -                    | -                    | -                    |
| 23  | Thalassionema       | +                  | +                      | -                    | -                    | -                    | -                    |
| 24  | Thalassiothrix      | +                  | +                      | +                    | +                    | -                    | -                    |

Division Chrysophyta

1. Atractomorpha
2. Characium
3. Stigeoclonium

Division Chlorophyta

1. Anabaenopsis
2. Merismopedia
3. Microcystis
4. Planktolyngbia
5. Trichodesmium
Table 3. The result of Sørensen similarity index of ephypite microalga genera among the seagrass species (E: E. acoroides; T: T. hemprichii), the portion of leaves (T: tip, M: middle, B: basal) middle and location (B: Bandengan waters, U: Ujungpiring Waters).

|     | ETB | EMB | EBB | TTB | TMB | TBB | ETU | EMU | EBU | TTU | TMU | TBU |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| ETB | 0,00| 80,95| 70,27| 70,27| 73,68| 54,55| 73,91| 66,67| 71,79| 63,16| 51,43| 58,06|
| EMB | 80,95| 0,00| 66,67| 59,46| 63,16| 59,46| 65,22| 66,67| 71,79| 63,16| 62,86| 58,06|
| EBB | 70,27| 66,67| 0,00| 71,43| 60,61| 71,43| 68,29| 64,86| 76,47| 66,67| 53,33| 53,85|
| TTB | 70,27| 59,46| 71,43| 0,00| 72,73| 71,43| 71,43| 54,05| 64,71| 72,73| 73,33| 61,54|
| TMB | 73,68| 63,16| 60,61| 66,67| 0,00| 66,67| 72,73| 52,63| 74,29| 64,71| 58,06| 66,67|
| TBB | 54,55| 48,48| 68,75| 71,43| 72,73| 0,00| 63,41| 54,05| 64,71| 72,73| 73,33| 61,54|
| ETU | 73,91| 65,22| 68,29| 63,41| 61,90| 63,41| 0,00| 69,57| 79,07| 71,43| 56,41| 51,43|
| EMU | 66,67| 66,67| 64,86| 54,05| 52,63| 54,05| 69,57| 0,00| 56,41| 47,37| 51,43| 45,16|
| EBU | 71,79| 71,79| 76,47| 64,71| 74,29| 64,71| 79,07| 56,41| 0,00| 80,00| 62,50| 64,29|
| TTU | 63,16| 63,16| 66,67| 72,73| 64,71| 72,73| 71,43| 47,37| 80,00| 0,00| 77,42| 59,26|
| TMU | 51,43| 62,86| 53,33| 73,33| 58,06| 73,33| 56,41| 51,43| 62,50| 77,42| 0,00| 50,00|
| TBU | 58,06| 58,06| 53,85| 61,54| 66,67| 61,54| 51,43| 45,16| 64,29| 59,26| 50,00| 0,00|

Figure 1. The Nutrient concentration in the water of the study sites.

Table 4. The Water quality in Bandengan and Ujungpiring Waters.

| Parameter               | Bandengan Waters | Ujungpiring Waters |
|-------------------------|------------------|--------------------|
| Temperature (°C)        | 27-33            | 30-32              |
| Salinity (ppt)          | 26-31            | 27-32              |
| pH                      | 8                | 6                  |
| DO (ppm)                | 3,54-6           | 2,5-6              |
| Light intensity (cm)    | 98-110           | 85-110             |
| Depth (cm)              | 98-110           | 85-110             |

It is known that there were 5 and 3 species of seagrass inhabit in Bandengan and Ujungpiring Waters [14, 15]. E. acoroides and T. hemprichii, belong to Family of Hydrocharitaceae, were found growing in mixed species meadow. E. acoroides is a very distinctive seagrass with very long and ribbon-like (30 – 150 cm long, approx. 1.25 – 1.75 cm wide), with many parallel veins, generally dark green in color, thick and tough leaves. The 'toughness' apparent were caused by the raised margins.
along the sides of the leaves, which generated by inrolling and thickening of the lateral edges. *T. hemprichii*, exhibits some variation in leaf width and length. However, the basic structure remains the same. The rhizome is thick (up to 5 mm thick), and distinctive, since the nodes, where the old shoots joined the leaf-bearing branch, are plainly visible with a prominent scale at each. Their pale basal leaf sheath is 3–7 cm long and well developed. Leaves are 10–40 cm long, ribbon-like and often slightly curved laterally. Leaf width is generally in the range of 0.4–1.0 cm. There are 10–17 longitudinal leaf veins. The leaves have numerous large tannin cells grouped in short black bars running parallel to the long axis of the leaf. These 'bars' are visible to the naked eye and are one of the diagnostic features of this species. The leaf tip is rounded and sometimes slightly serrated [18].

Compared to other research, there were 27 genera of microalgal ephypite in Teluk Awur [15] and 28 in Karimunjawa Waters [19]. The rapid growth of epiphytes, such as diatoms belongs to Chrysophyta Division which was considered the most important structural elements of the epiphyte on seagrass [19], which usually the result of the high nutrients.

Some epiphytic algae were known as specific and obligate epibionts on certain hosts [20], although most of them were essentially facultative and were not specifically associated with a host species [21]. Algal epiphytes play an important role in coastal benthic communities, as they provide a potential mutualistic interspecific association [22], as food and habitats for animals as well as considered as primary producers in the food chain [23, 24]. The epiphytic algae associated with seagrass are contributed 62, 50 and 44 % of primary production for *Syringodium filiforme*, *T. testudinum* and *Halodule wrightii*, respectively [25], 19–37 % for *T. hemprichii* [26] and 2–9 % for *E. acoroides* [27]. This contribution might be affected by the seagrass species which compose one bed. Other study done by [12] appeared that there were different epiphytic microalgal species composition on the leaves of *P. oceanica* and *C. nodosa*. It may vary due to local environmental conditions (hydrodynamics, light penetration), host characteristics (meadow type, shape forms of leaves, lifespan, and growth rate), and grazing effect which seem to be responsible for those dissimilarities in epiphytic microalgae communities. In present work, the similarity of epiphyte microalga seems due to the similar morphology of seagrass leaves, i.e., ribonlike of *E. acoroides* and *T. hemprichii*.

Previous studies by [28, 29, 30] showed that epiphytic community structure was influenced by abiotic factors such as light, temperature, nutrients, and water motion, as well as by biotic factors such as leafage, seasonal cycle of the host, and grazing pressure by herbivores [31, 32, 33, 34]. The similar of water quality of both study sites (figure 1; table 4) also affect the similarity epiphyte microalga inhabitant ion seagrasses leaves. The density of the seagrass canopy and shoot size can significantly modify epiphytic biomass, presumably due to effects of light penetration [35, 36], whereas shoot morphology, leaf, and stem ages can influence epiphyte distribution and abundance due to differences in the surface area that is available for epiphyte settlement [37, 38].

4. Conclusion

Most of the epiphytic microalgae comparison among the portion of the leaves, species of seagrasses and study sites have high similarity index (SI > 50 %) with the value in the range of 51.43–80.95 %.

Only four dissimilarities happened. It showed the diversity of ephypitic microalgae on the seagrass leaves.

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