Succession of epiphyte on thallus of *Kappaphycus alvarezii* (Rhodophyta) in horizontal net cage culture

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Abstract. *Elachista flaccida* in one of the epiphytes that attaches on thallus of *Kappaphycus alvarezii* and can reduce the growth of *K. alvarezii* as well. This research aims to identify occurrence and succession of epiphyte that attaches to *K. alvarezii* thallus. Results shows high occurrence of *E. flaccida* on the thallus of *K. alvarezii* during 14 day, 28 days, and 42 days of 6, 7, and 98 ind/m^3/day. *E. flaccida* succession occurs in day 10 of cultivation and decrease in days 20 which is paced by *Gracilaria bursa-pastoris* and *Ceramium* sp. During 30 days, *G. bursa-pastoris* is replaced by *Padina* sp. and *Palisada perforata.* Highest domination of *E. flaccida* has been found in day 10 with the amount of 97.25%. During research periods, water measurement shows that temperature was between 29-31°C, water transparency between 66-81% at 6 m depth, current velocity between 0.0910-0.0950 m/second, salinity between 30-31 ppt, nitrate (NO_3) between 0.0040-0.0070 mg/L, phosphate (PO_4) between 0.0016-0.0020 mg/L, dissolved oxygen (DO) between 6.6-7.2. No significant correlation was found between environment and the occurrence of *E. flaccida* and the growth of *K. alvarezii* during our research.
Key Words: *Eucheuma,* species composition, growth, environmental, red algae.

Introduction. *Kappaphycus alvarezii* is one of the primary commodities on the international markets, and Indonesia is one of the countries that supply raw seaweed to other countries in need (Kasim et al 2016). The increasing demand of *K. alvarezii* raw material is pushed by some of the industrial necessity such as food industry, pharmacy, medical, cosmetics, and paper industry. As a basic material of gelatin producer, alginate and seaweed carrageenan are very saleable in the both local and export market (Chapman & Chapman 1980; Chennubhotla et al 1987; Chennubhotla 1996; Kaladharan et al 1998; Chennubhotla & Kaliaperumal 1999). One of the most cultivated seaweed species is *K. alvarezii* which has economic importance in tropical area and commonly has red color and its wall cell contains high amount of polysaccharide that becomes the most important resource to supply carrageenan in the world (Thirumaran & Anantharaman 2009). In seaweed cultivation, farmers usually found harmful algae that attached to seaweed thallus called epiphyte, which may reduce seaweed productivity. Epiphyte is mostly living as a parasite towards other plantae. Most studies dealt with the colonization and distribution of epiphytes on seaweeds (Al-Handal & Wulff 2008; Worm & Sommer 2000). Moreover, the epiphytic communities were highly heterogeneous, showing a marked patchy distribution on seaweed (Thomas & Jiang 1986). Therefore a research about macroepiphyte succession and type composition on cultivated *K. alvarezii* with horizontal net cage regarding to harmful pest that hamper the seaweed growth is necessary.

The objective of this research is to determine the succession of epiphyte on thallus of *K. alvarezii* cultivated on horizontal net.

Material and Method. The present research was conducted on rainy season from April to June in Tanjung Tiram cultivation areas of South Konawe District, Southeast Sulawesi, Indonesia. Study site was established in two stations. Station 1 was located on 04^0 01'

57.0” LS and 122^0 40’ 30.5” BT and station 2 on 04^0 01’ 57.1” LS and 122^0 46’ 26.5” BT. In order to avoid herbivorous attack on sample, horizontal net cage were used during our research periods (Budiyanto et al 2019; Kasim & Mustafa 2017; Kasim et al 2018). In every station we placed 3 horizontal net cages. Macro epiphytes which were attached on thallus of *K. alvarezii* were collected at each measurement time (every 10 days) from each cage. Collection of microepiphyte were done carefully using pin set and glass sample (Kasim et al 2017). Every sample collection was labeled directly and brought to laboratory for identification. Macroepiphytes were identified up to species level (Setyobudiandi et al 2009; Barbara 2009). Environmental parameters (water quality) analysis was performed at the Analysis Laboratory at Faculty of Fishery and Marine Science, Halu Oleo University, Indonesia.

Water physico-chemical parameters measurement was done 3 times within 30 days with 10 days’ interval starting from day 10 (first pickup). Measured parameters was temperature using thermometer, water transparency using Secchi disk tool, current speed using current meter, salinity using handrefraktometer, nitrate, phosphate, and dissolved oxygen (DO) sample in the field was preserved using a solvent. Nitrate samples were preserved first using H_2SO_4 solvent until pH level reached 2, phosphate measurement was performed at the field by filtering phosphate liquid sample using filter paper and DO sample management was also handled in the field using 2 mL of NaCl solvent and $MnSO_4$. The samples were placed into a cool box before until further analyze in the laboratory. To clarify the correlation between macroephiphyte succession and the growth of *K. alvarezii*, we performed Correlation analysis using SPSS program, version 26.

Results and Discussion. Percentage of macroepiphyte species composition which has found on thallus of *K. alvarezii* in floating cage during research period was dominated by *Elachista flaccida.* This species has been found in day 10 until day 30 with the an occurence between 94.35-97.25% respectively. *Ceramium* sp. has been found in day 20 around 0.76-0.77% and *Padina* sp. has been found only 4.27% in day 30 (Figure 1).

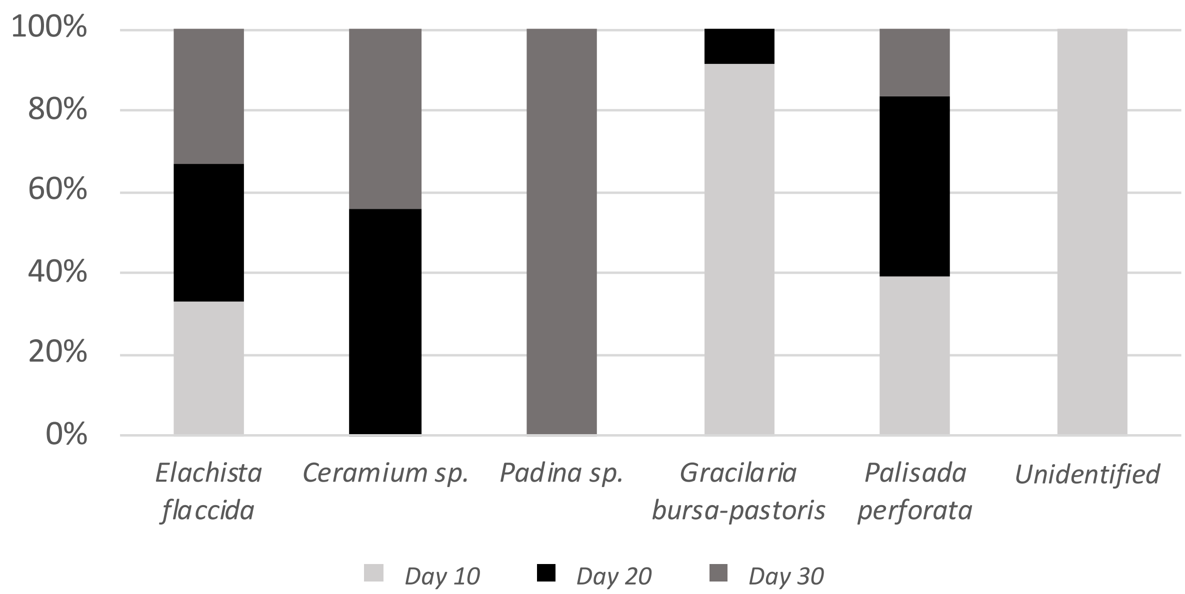

Figure 1. Percentage of macroepiphyte species composition on *Kappaphycus alvarezii* thallus.

E. flaccida was also found and its domination occurred on floating cages in all stations. Abundance of *E. flaccida* found on horizontal net cage after 10 days was 2,230 ind/cm^2 and 2,490 ind/cm^2 in Station 1 and Station 2 respectively. Abundance of *E. flaccida* decreased after 30 days and become 245 ind/cm^2 and 373 ind/cm^2 in Station 1 and Station 2, respectively (Figure 2).

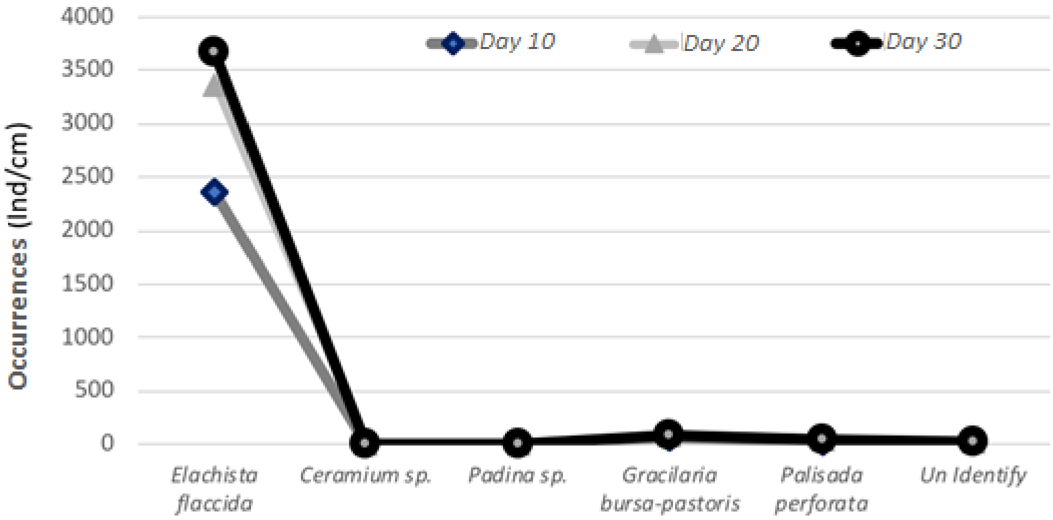

Figure 2. Occurrence of macroepiphyte in floating cages.

The results concerning *K. alvarezii* seaweed weight measurements, cultivated within Station 1 and Station 2 with initial weight of 50 g are presented in Figure 3.

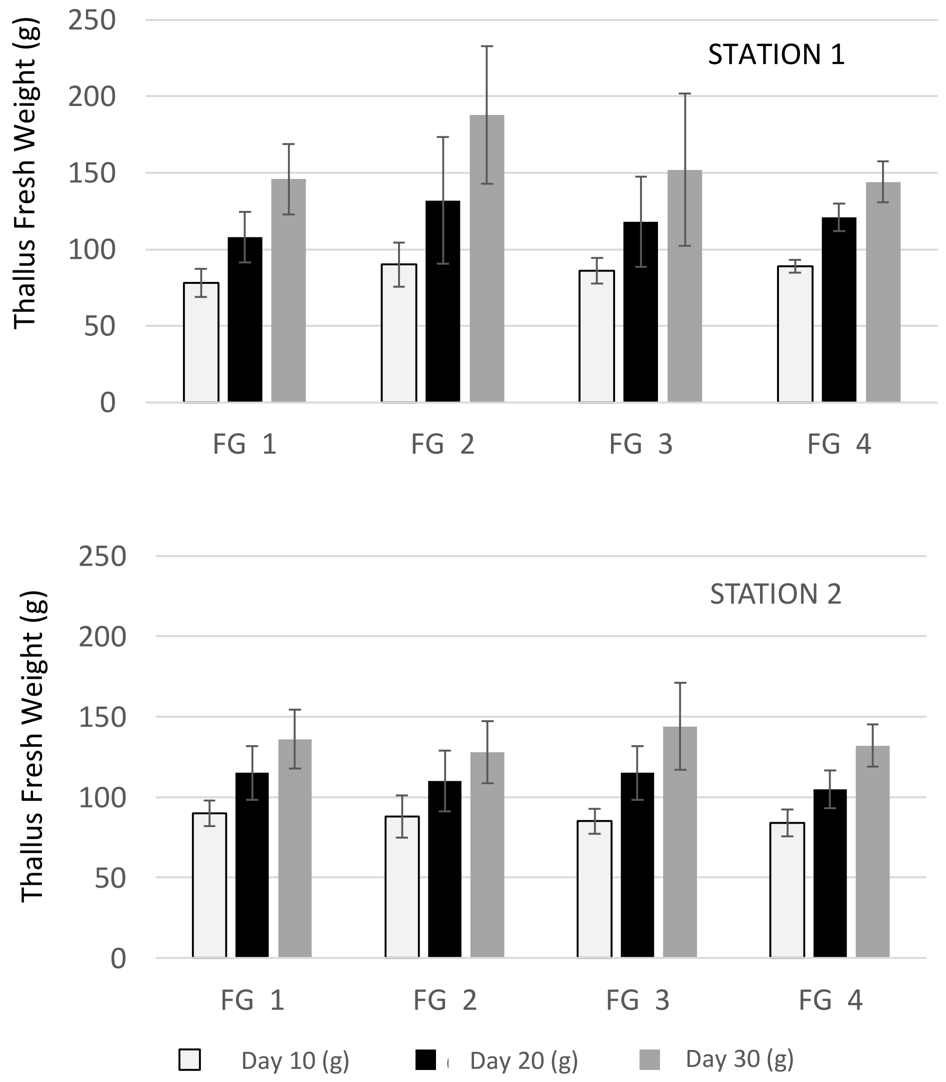

Figure 3. Results of *Kappaphycus alvarezii* thallus weight measurement.

Based on the observation towards macroepiphyte species composition on *K. alvarezii* thallus in horizontal net cages, it has been found 6 species of Rhodophyta class with 2 species (*Gracilaria bursa-pastoris,* and *Palisada perforata*) and Phaeophyta class with 3 species (*Ceramium* sp., *E. flaccida*, and *Padina* sp.) and also unidentified species (Marlia et al 2016; Marlia 2016). The most dominated macroepiphyte species was *E. flaccida* whereas the excessive occurrence found in Station 1 and 2 was 94.35-97.25%. The decrease of *E. flaccida* was started in day 20 and higher decrease was observed in day 30. This phenomenon may be caused by change in environmental condition (water), particularly due to temperature and current velocity which provokes some epiphytes detach from seaweed thallus (Marlia 2016). Some epiphyte will be influenced by the speed of the current. The current can release epiphyte from its attachment so that the structure of the epiphyte community can also change. Tolerance of current speed for a biofoulling organism is around 0.10 m/second Raikin (2004). However, because *E. flaccida* epiphyte is attached firmly on a thallus, determines some particular epiphytes survival on thallus. Some epiphyte is buried on the surface of the thallus and classified as a strong epiphyte on its host surface which causes the thallus surface becomes harsh as it has bulges (Rahman & Magdalena 2015). Macroepiphyte with the lowest species composition found was *Ceramium* sp. and *P. perforata.* The low macroepiphyte species composition that attaches to the thallus is caused by unstable weather condition during the cultivation process. Based on research time in study site, climate season was rainy while *P. perforata* macroepiphyte blossom in dry season (summer). Qualitative observation that has been conducted in 1998 showed that two species of Rhodophyta *P. perforata* and *Gracilaria coronopifolia* was excessive in the area of Hengchun Peninsula. The two species mentioned before prefer different season for thriving, *P. perforata* has a high growth in summer and *G. coronopifolia* has excessive growth throughout the year except April-May (Tsai et al 2005).

Growth of *K. alvarezii* during sampling periods showed that fresh weight increased form its initial weight, it is assumed that environmental factors still supported the growth of *K. alvarezii*. Sufficient nutrient elements supported by ocean current flow, causes agitation process so that nutrient absorbance by *K. alvarezii* is well indicated and the growth tends to increase. Besides, other factors such as temperature, salinity, current speed, water transparency, dissolved oxygen, total nitrogen and orthophosphate also affect the growth and development of *K. alvarezii.* Bulboa & Paula (2005) reported that hydrodynamic factor in the ocean that is affected by climate change is extremely impactful towards the growth of *K. alvarezii.*

Until day 10, *E. flaccida* and *G. bursa-pastoris* has been found in high density attached to *K. alvarezii* thallus*.* In day 20 they has been found starting to reduce drastically and to be replaced by the new species of *Ceramium* sp. In day 30, *E. flaccida* and *Ceramium* sp. were still attached event hood drastically reduce in number on thallus *K. alvarezii*, and *G. bursa-pastoris* was no longer attached and replaced with a new species of *Padina* sp. and *P. perforata*. Meanwhile in Station 2, in day 10, has been found *E. flaccida*, *G. bursa-pastoris*, and *P. perforata*. In day 20, those species has been found but the individual amount was reduced drastically and replaced by *Ceramium* sp. In day 30, macroepiphyte *E. flaccida* still was found but with a very reduced amount and started to be replaced by *P. perforata* and *Ceramium* sp. and after days 30, replaced by *Padina* sp. Disappearance of macroepiphyte which initially occurs and attaches to the thallus or macroepiphyte that sustain a succession is caused by the lack energy of its parasitism on the cultivated *K. alvarezii* thallus because there is no holdfast as a gripper causing a low adhesion (Marlia et al 2016; Marlia 2016). Leonardi et al (2006) states that macroepiphyte with enough strong parasitism could surpass the wall cell and corticolous cell tends to attach firmly and lasts longer, also it could give negative impact to the seaweed growth. While the epiphyte type that attach weakly and cannot surpass into the thallus wall cell tends to detach easily and cannot give an impactful effect to the seaweed. *E. flaccida* is relatively smaller compared to other macroalgae and equipped with enough short flagella and also has the characteristics to could attach firmly to other algae in the water (Lee & Garbary 1999). Greene & Grizzle (2007) stated that there are potential and complex biotic interaction that is affected by physical factors and cause parasitism succession and parasite organism community structure change.

Environmental factors had a normal value with a range that supports the growth of seaweed. Temperatures ranged from 29 to 31°C. The current velocity was high on day 20 with 0.095 m/s. Nitrate was higher on day 20, 0.007mg/L, while phosphate was lower on day 30, 0.16 mg/L (Table 1).

Table 1

Results of water physico-chemical parameters measurement at study sites

No.	*Parameter*	*Collection time*		
		Day 10	*Day 20*	*Day 30*
1	Physical			
	- Temperature (^{0}C)	29	31	30
	- Water transparency (m)	75%	81%	66%
	- Current speed (m/s)	0.0091	0.0950	0.0937
2	Chemical			
	- Salinity (ppt)	30	30	31
	- Nitrate (mg/L)	0.0040	0.0070	0.0049
	- Phosphate (mg/L)	0.0020	0.0020	0.0016
	- Dissolved oxygen (DO) (mg/L)	7.2	6.9	6.6

Current velocity that is strong enough could keep the seaweed clean from sediment so that all of the thallus part could work as its function for photosynthesis process. If the the water current is faster, more inorganic nutrients are carried along by the water and could be absorbed by plant through diffusion process. In motionless water, plants will having less nutrients, so it can interrupt the photosynthesis process. So the seaweed seed have to be planted in some area with strong current flow between 20-40 cm/sec (Sulistijo & Atmadja 1996). Based on the current speed measurement during the research, it has been found an average speed of 0.09 m/second. Basically the average of current speed in the research location can be classified as not optimal for the growth of seaweed and microepiphyte. Sulistijo & Atmadja (1996) stated that one of the requirements to determine the location of *Eucheuma* sp. is if there are current flows with the speed of 0.33-0.66 m/second. Therefore, the current speed measured in the research location was relatively slow against what was necessary. Effendi (2003), explain that nitrate is a primary nitrogen form in natural water and main nutrient for algae growth. Nitrate is extremely easy to dissolve in water and is stable. Nitrate is produced from a perfect oxidation process of nitrogen in water. Nitrogen is one of the main elements that composes organism cell in the process of protoplasm establishment. Nitrogen usually has a limited amount in water, especially in tropical climate area. Lack of nitrate in water could hamper the growth of aquatic plants, although the other nutrient elements are excessive (Patadjal 1993). Based on nitrate measurements in the research location, it showed 0.0040 mg/L in day 10 of the observation, and increase in day 20 to 0.0070 mg/L and in day 30 decreased to 0.0049 mg/L. Nitrate concentration in that research location is categorized as low for seaweed and macroepiphyte growth. This is supported by Effendi (2003) whom stated that nitrate concentration that is appropriate for seaweed growth is between 0.02-0.04 mg/L, while for macroepiphyte is between 0.001-0.012 mg/L (Raikin 2004). Phosphate is an essential element for epiphyte, plants and aquatic algae and also affects water productivity level (Kasim & Mukai 2006). There are also support interaction between plants and animals, between organics and inorganics, and between water column and surface and also substrate. For instance, some animals release a large amount of phosphate which is dissolved in their feces. Thus phosphate is dissolved into water later on and it is available for plants to use. Some of inorganic phosphates are settled as a mineral in the bottom of the ocean (Effendi 2003). Phosphate concentration in research location which was 0.0020 in day 10 and day 20, and in day 30 is 0.0016 mg/L is categorized as low for a proper seaweed growth. While for macroepiphyte growth according to phosphate level in this research can be classified as optimal. Raikin (2004) stated that phosphate level towards macropiphyte growth is 0.0012-0.055 mg/L. Phosphate (PO_4) could be a boundaries factor temporally or spatially because phosphate resource is modest in water. Dissolved oxygen is the most important element and extremely necessary for a respiration process and for decomposing organic matters by microorganism. Solubility of oxygen in water is influenced by temperature, partial pressure of gases in the air and in the water, salinity level, and elements that is easily oxidized in the water. The higher the water temperature, salinity, and dissolved gases pressure, is more likely that the oxygen solubility to decrease in water (Wardoyo 1981). Based on the dissolved oxygen (DO) measurements in the research locations, was found 7.2 mg/L in day 10, and then decreased in day 20 and day 30 to 6.9 mg/L and 6.6 mg/L respectively. Thus condition of dissolved oxygen level is normal for a coastal water area. According to Yusuf (2004), dissolved oxygen level that is appropriate for a seaweed growth is 6.9-7.1 mg/L, because if the dissolved oxygen is lower than 4 mg/L it indicates that the water area has a lack of oxygen problem caused by temperature rise in day time, night time is caused by water organism respiration.

Conclusions. There was found six species of macroepiphyte occured on thallus of *K. alvarezii* cultivated using floating cages. *E. flaccida* was the dominant macroepiphyte that attached on thallus of *K. alvarezii* with 94.35-97.25%. Species domination occured until day 10 and then started to decrease until day 20 and was replaced by *P. perforata* and *Ceramium* sp. After 30 days, *E. flaccida* repeated its decrease and was replaced by *Padina* sp.

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