Epiphyte Identification on *Kappaphycus alvarezii* Seaweed Farming Area in Arungkeke Waters, Jeneponto and the Effect on Carrageenan Quality

Sri Redjeki Hesti Mulyaningrum^{*}, Hidayat Suryanto Suwoyo, Mudian Paena, Bunga Rante Tampangallo

Research Institute for Coastal Aquaculture and Fisheries Extension JI. Makmur Dg. Sitakka No. 129 Maros 90511, South Sulawesi Email: mulyaningrum@kkp.go.id

Abstract

Kappaphycus alvarezii which is widely cultivated at sea is susceptible to other algae that drift away and attach as epiphyte. This study aims to identify epiphyte on seaweed farming K. alvarezii and its effect on carrageenan quality. The study was conducted on K. alvarezii seaweed farming area in Jeneponto, South Sulawesi. Sample of epiphyte was obtained from four stations. Epiphyte was identified in laboratory based on its morphological characteristics and calculated its density. Carrageenan yield and gel strength of healthy seaweed and those infected one were analyzed in laboratory in triplicates for each sample. Data were statistically analyzed using independent samples t-test analysis. Present study showed that there were 6 epiphytes species on the seaweed farming area, namely: Entheromorpha intestinalis, Ceramium sp., Neosiphonia apiculata, Chaetomorpha crassa, Hypnea sp., and Gracilaria sp. The average of epiphytic density in cultivation area was (24.26 ± 9.64) %. Healthy seaweed and infected one had significantly different carrageenan yield and gel strength (P<0.05). Healthy seaweed had higher carrageenan yield (48.17 ± 1.62) % and gel strength (1130.76±8.42) g.cm⁻² than infected seaweed which had carrageenan yield (42.47 ± 0.23) % and gel strength (958.22±10.85) g.cm⁻².

Keywords: epiphyte, cultivation, K. alvarezii, carrageenan

Introduction

The demand for seaweed production is increasing because of an increasing in its utilization. K. alvarezii is carrageenan-producing seaweed with high economic value. Carrageenan is widely used in global industries of various food, pharmaceutical, and cosmetics. Indonesia has big contribution to the production of tropical seaweed, especially K. alvarezii. Indonesian seaweed production continues to increase for more than 10 times, in 2005 it was less than 1 million tons increasing into 10 million tons in 2014. Indonesia contributed to the world seaweed production increasing, from 6.7% in 2005 to 36.9% in 2014 (FAO, 2016). The major mariculture commodity in Indonesia is seaweed for carrageenan production. Seaweed production accounts for 98% of total Indonesian mariculture production and 84% of value. Since 2010 until 2014, seaweed production in Indonesia increase 32% per annum (Rimmer, 2010). Seaweed production was potential to meet the sustainability of farmer's livelihood, seaweed farming has become the main income source for small-scale fishermen, particularly for some areas in South Sulawesi (Yusuf et al., 2018).

K. alvarezii is widely cultivated at sea. One of the obstacles in seaweed cultivation at sea is the difficulty of controlling epiphytes. The sea, as an open space, enables wide access for other algae to move along with the current and attached as competitor algae on cultivated seaweed. The aquatic environment is a factor that affects the occurrence of epiphytes, especially in tropical regions such as Indonesia. The tropical marine and coastal ecosystems are rich in biodiversity, as an archipelagic country with large areas for seaweed cultivation, Indonesia has advantages in the abundance of tropical seaweed resources. The commercial types of seaweed can be widely found in Indonesia (Mulvati and Geldermann. 2017) However, a tropical country with adequate sunlight and rich biodiversity has consequences on epiphytic problem in seaweed farming. Spore and propagule of other algae are possible to drift away and attach on cultivated seaweed, the availability of adequate sunlight will support the competitor algae to grow well on host. Epiphytes are common phenomenon in seaweed cultivation, and even become a major problem affecting the productivity of seaweed (Lüning and Pang, 2003; Critchley et al. 2004; Hurtado et al., 2006a; Vairappan, 2006).

Several studies have reported that epiphytes decrease the growth rate of cultivated could seaweed as well as decreasing in biomass due to competition in space and nutrients with the host (Buschmann and Gómez 1993). Moreover, epiphytes also disrupt photosynthesis process and affecting formation process of carrageenan. The the decreasing of carrageenan quality in some cases causing in economic losses. To evaluate the kind of epiphyte on seaweed farming and the effect of epiphyte on carrageenan quality, we observed some epiphytes on K. alvarezii seaweed farming area on Arungkeke waters, Jeneponto regency, South Sulawesi and analysed carrageenan quality of cultivated seaweed.

Materials and Methods

Map by Peter Loud

The study was conducted at seaweed farming area in Arungkeke waters, Jeneponto, Regency, South Sulawesi in June 2018 (Figure 1.). The observation was executed when there was an epiphytic occurrence at a long line-seaweed farming area. Sample of seaweed, both healthy and epiphyte infected, were taken randomly, obtained from four sampling stations then transported to laboratory for being observed. Epiphytes were separated from the host and identified based on their morphological characteristics (Hurtado et al., 2006b). The measurement of epiphytic density was carried out based on the method of Muñoz and Fotedar (2010). Sample obtained from four stations was rinsed using sea water, while the epiphyte was separated from the host. Both seaweed and separated epiphyte were weighed, then epiphyte load was calculated

using the following formula according to Buschmann and Gómez (1993).

The quality of carrageenan was measured by analysing carrageenan yield and gel strength in healthy seaweed-free epiphytes and infected seaweed. Carrageenan yield and gel strength were analysed in laboratory in 3 replicates for each sample. Carrageenan extraction was carried out using the isopropyl alcohol precipitation (IPA) method and gel strength was analysed using the Brookfield CT3 Texture Analyser according to the Marine Colloids (1977) method. Epiphytic data was analysed descriptively, while the carrageenan yield and gel strength data were analysed statistically using independent samples t-test.

Results and Discussion

Morphological identification to competitor algae in *K. alvarezii* samples showed several species of epiphytes as presented in (Figure 2.). The distribution of competitor algae species at each station was shown in Table 1. Generally, all stations had the same species of epiphytes, it were: *Entheromorpha intestinalis, Ceramium* sp., *Hypnea musciformis, Neosiphonia apiculate, Gracilaria* sp., and *Chaetomorpha crassa*.

Kasim et al. (2017) reported there were 7 macro epiphytes species in Eucheuma spinosum seaweed cultivation in Bau-bau, Southeast Sulawesi, namely: Acanthophora spicifera, Chondrophycus papillosus, Chaetomorpha crassa, Jania longifurca,



Note: 1-4 = sampling station





Entheromorpha intestinalis



Ceramium sp.



Neosiphonia apiculata



Chaetomorpha crassa



Gracilaria sp.

Figure 2. Some epiphytes found on K. alvarezii seaweed farming area

Pomatoceros triqueter, Ulva lactuca and Turbunaria ornata. Radulovich et al. (2015) reported filamentous species Chaetomorpha sp., was ubiquitous and grew abundantly on the surface of floating objects such as logs, ropes, fish cages and cultivated seaweeds and considered as fouling pest.

Hayashi et al. (2010), mentioned several types of epiphytes in seaweed: epiphytes which attaching weakly to the surface of seaweed and does not cause any damage host tissue; epiphytes which attaching firmly to the surface of seaweed and does not damage the host tissue: epiphyte which spreading on the surface of the outer layer of the host cell wall and does not damage cortical cells; epiphyte which spreading to the outer layer of the host cell wall and associated with damage to the host cortical cell and epiphyte which attacking the host tissue and growing intercellular, being associated with cortical destruction and medullary cells. Hypnea sp. and Gracilaria sp. were classified as first type of epiphyte. C. crassa, E. intestinalis and Ceramium sp. were second type of epiphyte, while N. apiculata was classified as fifth type of epiphyte. Arisandi et al. (2013) reported epiphytes in Eucheuma cottonii were dominated by the C. crassa which had resembles yarn shape and clot, covered thallus surface, thus blocking the penetration of light and became a competitor of E. cottonii in absorbing

nutrient. C. crassa was also a suitable habitat for bacterial life. Neosiphonia sp. infection could decreased biomass and carrageenan quality. Neosiphonia sp. was known as epiphytic filamentous algae (EFA) which attached to K. alvarezii by forming primary rhizoid and secondary rhizoid in host (Hurtado et al., 2006a; Vairappan et al. 2014). Vairappan, et al. (2007) reported the early of N. apiculata infection was marked by a black stain on the cuticle surface cells which indicated the presence of epiphytic spores then developed, corresponding to the condition of the water temperature and salinity. Several epiphytes caused damage on host's thallus by attaching its spores on its host, made weakening of cultured seaweed, and made its exposure to bacteria population (Tampangallo et al., 2018).

The average of epiphytic density on cultivation area was $(24.26\pm9.64)\%$, the distribution of epiphytic density on each station was described in Figure 3. Hosnan *et al.* (2016) reported the prevalence of competitor algae on *E. cottonii* seaweed farming in Sumenep regency was 0.14-17.24%, meanwhile Munõz and Fotedar (2010) reported the epiphytes density on *Gracilaria cliftonii* seaweed farming up to 40% with 24 types of epiphytic species. Vairappan *et al.* (2008) measured the epiphytic density in *K. alvarezii* seaweed cultivation based on the number of epiphytes per centimetre square of seaweed specimen, it was reported that the epiphytic density of *K. alvarezii* from Tawi-tawi, Indonesia was 69 epiphytes cm⁻². Tsiresy *et al.* (2016) found the rate of epiphytes infestation varied from 42 to 78 epiphytes cm⁻².

Some factors that influence the onset of epiphytism among others are the intensity of sunlight and the movement of water. The emergence of epiphytes at the study site of seaweed farming was suspected due to the low currents in Arungkeke waters. Current becomes one of the determining factors of macro algae spreading. Current is an important factor influencing the transport of epiphytes. The vertical and horizontal spreading of macro algae is generally associated with the pattern and velocity of currents in certain regions and also the topography of each area (Komatsu et al., 2014). Epiphytism usually occurs when there was change in water temperature, wind and water movement (Johnson and Gopakumar, 2011). Munoz and Fotedar (2010) explained that the growth of epiphytes on seaweed cultivation was due to an increase of the water temperature and the occurrence of irradiation, further reported that the highest epiphytic diversity was in spring time. The availability of light in certain regions will greatly affect the algal community in waters.

Carrageenan quality

Carrageenan yield of healthy seaweed was higher $(48.17\pm1.62)\%$ than infected seaweed $(42.47\pm0.23)\%$ (Figure 4.). T-test analysis showed that carrageenan yield of healthy seaweed was

significantly different from infected seaweed (P<0.05). The gel strength of healthy seaweed was higher (1130.76 \pm 8.42) g cm⁻² than infected seaweed (958.22±10.85) g cm⁻² (Figure 5). The result of t-test analysis showed that the gel strength of healthy seaweed was significantly different with infected seaweed (P<0.05). Previous studies reported that epiphytes in K. alvarezii seaweed could decrease the yield and quality of carrageenan. Vairappan et al. (2014) reported that infected K. alvarezii by N. apiculata had lower carrageenan yield (20.5±2.5%) than healthy seaweed (65.5±4.2%), as well as its gel strength was lower (687.9±3.5 g cm⁻²) than healthy seaweed (1456.5±24.7 g cm⁻²). Pang et al. (2011) reported, epiphytic disruption in seaweed was through two methods i.e. material exchange and energy metabolic. The condition may lead to the disruption during photosynthesis process. Photosynthesis is basic anabolism process and the only mechanism for transferring sunlight energy into chemical energy for plants. Disruption during the photosynthesis process will affect the quality of carrageenan, in which the phycocolloid is result of photosynthesis. Mulyati the and Geldermann, (2017) reported that the most critical risk in carrageenan supply was poor quality of raw dried seaweed which was contaminated with other during cultivation and post-harvest materials handling.

Some studies reported some efforts have been performed to overcome epiphytes. Lüning and Pang (2003) recommended high density seaweed cultivation method to overcome epiphytes. Base on the knowledge that the competitor algae is known as a fast-growing but opportunistic organism, its growth

Sampling station	Epiphyte
1	Entheromorpha intestinalis
	Ceramium sp.
	Hypnea musciformis
	Neosiphonia apiculata
	Gracilaria sp.
2	Entheromorpha intestinalis
	Ceramium sp.
	Hypnea musciformis
	Neosiphonia apiculata
	Gracilaria sp.
3	Chaetomorpha crassa
	Entheromorpha intestinalis
	Ceramium sp.
	Neosiphonia apiculata
4	Chaetomorpha crassa
	Entheromorpha intestinalis
	Ceramium sp.
	Neosiphonia apiculata

Table 1. Distribution of epiphytes on each sampling stations



Figure 3. Epiphytic load on four sampling stations



Figure 4. Carrageenan yield of healthy and epiphyte infected K. alvarezii



Figure 5. Gel strength of healthy and epiphyte infected *K. alvarezii*

is very dependent on the sunlight. Thus, in highdensity cultivation, competitor algae will be covered by cultivated seaweed and do not get enough sunlight for its growth. Borlongan et al. (2011) overcome epiphytes by using AMPEP (Acadian Plant Extract Powder), found Marine that Neosiphonia growth was reduced up to 6-50% by AMPEP application. Hurtado and Critchley et al. (2013) explained that AMPEP could reduce damage on K. alvarezii seaweed caused by Neosiphonia epiphytic. The use of this Ascophyllum nodosum extract was claimed as an efficient and effective tool to help reduce Neosiphonia epiphytism (Hurtado et al., 2014). The other epiphytism control technique is by maintaining a certain light intensity that favours the growth of cultivated seaweed but reduces the epiphytic algae. Changing the seasonal rhythms of seaweed cultivation also helps to control epiphytism (Titlyanov and Titlyanova, 2010). Farmers have a simple way to prevent epiphytes, i.e. by shaking the rope line of seaweed frequently, so that the propagules or spores of epiphytes do not stick to cultivated seaweed and cultivation facilities. This method is effective enough to avoid epiphytic disorders in seaweed cultivation.

Conclusion

Epiphytes found on K. alvarezii seaweed farming Arungkeke area in waters were Entheromorpha intestinalis, Ceramium sp., Neosiphonia apiculata, Chaetomorpha crassa, Hypnea sp., and Gracilaria sp. Epiphyte affected the quality of carrageenan, where healthy seaweed had higher carrageenan quality than infected one.

References

- Arisandi, A., Farid, A., Wahyuni, E.A., & Rokhmaniati, S. 2013. Impact of Ice-ice Infection and Epiphyte to Eucheuma cottonii Growth. Ilmu Kelautan, 18(1): 1–6. doi: 10.14710/ik.ijms. 18.1.1-6
- Borlongan, I.A.G., Tibubos, K.R., Yunque, D.A.T., Hurtado, A.Q & Critchley, A.T. 2011. Impact of AMPEP on the growth and occurrence of epiphytic *Neosiphonia* infestation on two varieties of commercially cultivated *Kappaphycus alvarezii* grown at different depths in the Philippines. *J. Appl. Phycol.*, 23(3): 615-621. doi: 10.1007/s108 11-010-9649-9.
- Buschmann, A. & Gómez, P. 1993. Interaction mechanisms between *Gracilaria chilensis* (Rhodophyta) and epiphytes. *Hydrobiologia*, 260–261: 345–351. doi: 10.1007/BF000490 39.
- Critchley, AT., Largo, DB., Wee, W., Bleicher, LG., Hurtado, AQ. 2004. A preliminary summary on *Kappaphycus* farming and the impact of epiphytes. *Japanese J. Phycol.* 52: 231–232.
- FAO. 2016. The State of World Fisheries and Aquaculture 2016. Contributing to food security and nutrition for all. Rome. 200 pp.
- Hayashi, L. Hurtado, A.Q., Msuya, F.E., Bleicher-Lhonneur, G., & Critchley, A.T. 2010. A review of *Kappaphycus* farming: prospects and constraints *in*: seaweeds and their role in globally changing environments. Springer. New York. 283 p.

- Hosnan, M.H., Arisandi, A. & Hafiludin. 2016. Identifikasi spesies alga competitor *Eucheuma cottonii* pada lokasi yang berbeda di Kabupaten Sumenep. *Pros. Seminar Nasional Kelautan Universitas Trunojoyo Madura*. p. 334-338.
- Hurtado, A.Q., Critchley, A.T., Trespoey, A. & Lhonneur, GB. 2006a. Occurrence of *Polysiphonia* epiphytes in *Kappaphycus* farms at Calaguas Is., Camarines Norte Phillippines. *J Appl Phycol* 18: 301–306. doi: 10.1007/s108 11-006-9032-z.
- Hurtado, A.Q., Luhan, M.R.J. & Guanzon Jr, N.G. 2006b. Seaweed of Panay. Second edition. Aquaculture Department Southeast Asian Fisheries Development Center. Tigbauan, Iloilo, Philippines. 50 p.
- Hurtado, A.Q. & Critchley, A.T. 2013. Impact of Acadian Marine Plant Extract Powder (AMPEP) in *Kappaphycus* Production. Malaysian Journal of Science (SCS Sp Issue) 32: 239-252.
- Hurtado, A.Q., Gerung, G.S., Yasir, S. & Critchley, A.T. 2014. Cultivation of tropical red seaweed in the BIMP-EAGA region. *J. Appl. Phcol.* 26: 707-718. doi: 10.1007/s10811-013-0116-2.
- Johnson, B. & Gopakumar, G. 2011. Farming of the seaweed *Kappaphycus alvarezii* in Tamil Nadu coast-status and constraints. Marine Fisheries Information Service T&E Ser., No. 208. p.1-5.
- Kasim, M., Jamil, M.R. & Irawati, N. 2017. Occurrence of macro-epiphyte on *Eucheuma spinosum* cultivated on floating cages. *AACL Bioflux*, 10(3): 633-639.
- Komatsu, T., Mizuno, S., Natheer, A., Kantachumpoo, A., Tanaka, K., Marimoto, A., Hsiao, S.T., Rothäusler, E.A., Shishidou, H., Aoki, M. & Ajisaka, T. 2014. Unusual distribution of floating seaweeds in the east China sea in the early spring of 2012. *J. Appl. Pycol.* 26(2): 1169-1179. doi: 10.1007/s1081 1-013-0152-y.
- Lüning, K. & Pang, S. 2003. Mass cultivation of seaweeds: current aspects and approaches. *J. Appl. Phycol.*, 15: 115–119. doi: 10.1023/A:10 23807503255.
- Marine Colloids, FMC Corp. 1977. Carrageenan. Marine Colloid Monograph Number One. Marine Colloid Division FMC Corporation. Springfield, New Jersey. USA.
- Mulyati, H. & Geldermann, J. 2017. Managing risks in the Indonesian seaweed supply chain. *Clean*

Techn. Environ. Policy 19:175–189. doi: 10.10 07/s10098-016-1219-7.

- Muñoz, J. & Fotedar, R. 2010. Epiphytism of Gracilaria cliftonii (Withell, Millar & Kraft) from Western Australia. J. Appl. Phycol. 22: 371– 379. doi: 10.1007/s10811-009-9469-y
- Pang, T., Liu, J., Liu, Q. & Lin, W. 2011. Changes of Photosynthetic Behaviors in Kappaphycus alvarezii Infected by Epiphyte. Evidence-Based Complementary and Alternative Medicine: 1-7. doi: 10.1155/2011/658906.
- Radulovich, R., Umanzor, S., Cabrera, R. & Mata, R. 2015. Tropical seaweeds for human food, their cultivation and its effect on biodiversity enrichment. *Aquaculture*, 436: 40–46. doi: 10.1016/j.aquaculture.2014.10.032.
- Rimmer, M.A. 2010. Mariculture development in Indonesia; prospect and constraints. *Indo. Aquacul. J.* 5(2):187-201. doi: 10.15578/iaj. 5.2.2010.187-201.
- Tampangallo, B.R., Suwoyo, H.S., Paena, M. & Mulyaningrum, S.R.H. 2018. Population of bacteria and epiphytes on *Kappaphycus alvarezii* seaweed in Pandang, Jeneponto regency, South Sulawesi. Paper presented at the 3rd International CEDUS-UNHAS Seaweed Symposium. Makassar.
- Titlyanov, E.A. & Titlyanova, T.V. 2010. Seaweed Cultivation: Methods and Problems. *Russian J. Mar. Biol.* 36(4): 227–242. doi: 10.1134/ S1063074010040012.
- Tsiresy, G., Preux, J., Lavitra, T., Dubois, P., Lepoint, G. & Eeckhaut, I. 2016. Phenology of farmed seaweed Kappaphycus alvarezii infestation by the parasitic epiphyte Polysiphonia sp. in Madagascar. J. Appl. Phycol. 28(5): 2903-2914 doi: 10.1007/s10811-016-0813-8.
- Vairappan, C.S. 2006. Seasonal occurrences of epiphytic algae on the commercially cultivated red alga *Kappaphycus alvarezii* (Solieriaceae, Gigartinales, Rhodophyta). *J. Appl. Phycol.* 18: 611–617. doi: 10.1007/s10811-006-9062 -6.
- Vairappan, C.S., Chung, C.S., Hurtado, A.Q., Soya, F.E., Lhonneur, G.B. & Critchley A. 2008. Distribution and symptoms of epiphyte infection in major carrageenophyte-producing farms. J. Appl. Phycol. 20: 477-483. doi: 10.1007/978-1-4020-9619-8_4.
- Vairappan, C.S., Chung, C.S. & Matsunaga, S. 2014. Effect of epiphyte infection on physical and

chemical properties of carrageenan produced by *Kappaphycus alvarezii* Doty (Soliericeae, Gigartinales, Rhodophyta). *J. Appl Phycol.* 26: 923–931. doi: 10.1007/s10811-013-0126-0.

Yusuf, S., Arsyad, M. & Nuddin, A. 2018. Prospect of seaweed development in South Sulawesi

through a mapping study approach. *IOP Conf. Series: Earth and Environ. Sci.* 157 012041 doi: 10.1088/1755-1315/157/1/012041.