SECONDARY METABOLITE OF SUMBAWA ALGAE AND ITS POTENTIAL AS A NATURAL PRESERVATIVE

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ABSTRACT

Pathogenic bacterial contamination was a serious matter due to its capability in reducing food quality and health. This study aimed to select various types of algae in Luk Coast, Sumbawa Regency that have the potential to produce antibacterial compounds for natural food preservatives. Algae on Luk Coast was identified by means of morphological characters, followed by sample preparation and extraction of secondary metabolites (bioactive compounds). Algae extracts were used in antibacterial tests against food spoilage bacteria, such as *Escherecia coli*, *Staphylococcus aureus*, *Salmonella thypi*, *Enterobacter cloacae* and *Pantoea agglomerans*. Five types of algae identified were *Padina* sp., *Halimeda opuntia*, *Sargassum horneri*, *Sargassum crassifolium* and *Galaxaura rugose*. The five algae have the growth-inhibiting ability toward the tested bacteria. The highest inhibition zone was obtained from the 100% algae extract concentration.

Keywords: antimicrobial, natural preservative, secondary metabolite, Sumbawa algae

INTRODUCTION

Food spoilage and foodborne infection increasingly become issues in the food industry, due to their serious impact on food quality and safety (Vanegas *et al.* 2017). Food spoilage is caused by pathogenic microorganisms. Several types of food-spoiling microorganisms include aerobic psychrotrophic gram-negative bacteria, yeasts, molds, heterofermentative lactobacilli, and spore-forming bacteria (Rawat 2015). Food spoilage due to infection from foodborne microbes is mainly caused by *Campylobacter* spp., *Salmonella*, and *Eschercia coli* (Koluman & Dikici 2013). Contamination from these spoilage pathogenic bacteria, not only decreases food quality but also causes adverse health conditions. In order to inhibit the process of food deterioration, some people use synthetic preservatives such as formaldehyde, benzoic acid, BHA (*Butilated Hydroxyanisol*), BHT (*Butylated Hidroxytoluene*) and TBHQ (*Tertier Butylated Hydroxyanisole*), especially for food ingredients having high water content. However, the use of synthetic preservatives is not recommended by *Badan Pengawas Obat dan Makanan* (National Agency of Drug and Food Control/BPOM). Therefore, it is necessary to seek alternative food preservatives from natural ingredients.

Algae have biological properties that can be used as antibacterial (Renhoran *et al.* 2017; Basir *et al.* 2017). Algae have the ability to produce secondary metabolites which are bioactive compounds to protect themselves from unsuitable environmental conditions and to defend themselves from the threat of various diseases and predators. Bioactive compounds in algae can also act as antimicrobials, antifeedants, antihelmintic and cytotoxic agents (Al-Saif *et al.* 2014).

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Several secondary metabolites produced by marine algae include alkaloids, polyketides, cyclic peptides, polysaccharides, phlorotannins, diterpenoids, sterols, quinones, lipids and glycerols (Cabrita *et al.* 2010). Al-Saif *et al.* (2014) reported that groups of flavonoid compounds contained in marine algae include rutin, quercetin and kaempferol. These compounds can be used as natural food preservatives (Hidayati *et al.* 2017).

Algae can be easily found in Indonesia, such as on Sumbawa Island. Sumbawa Island is located in the Nusa Tenggara Barat (NTB) Province. This island has a 3,331.72 km^2 water area, rich in various types of algae, including red algae (*Rhodophyta*), brown algae (*Ocrophyta*) and green algae (*Chlorophyta*).

Cabrita *et al.* (2010) reported that *Laurencia* is a group of red algae that produce secondary metabolites such as diterpenes, sesquiterpenes, triterpenes and C15-acetogenins. Various kinds of compounds derived from phlorotannins (phloroglucinol, eckol, dieckol, etc.) that was beneficial for health were produced by brown algae such as *Ecklonia cava*, *Ecklonia stolonifera*, *Ecklonia kurome*, *Eisenia bicyclis*, *Ishige okamurae*, *Sargassum thunbergii*, *Hizikia fusiformis*, *Undaria pinnatifida* and *Laminaria japonica* (Li *et al.* 2011). Bhagavaty *et al.* (2011) reported that the bioactive compounds of green algae *Chloroccocum humicola* such as lipophilic and phenolic have antimicrobial properties against *Bacillarius subtilis*, *Staphylococcus aureus*, *Eschericia coli*, *Pseudomonas aeruginosa*, *Salmonella thypimurium*, *Klebsiella pneumoniae*, *Vibrio cholerae*. The secondary metabolites of algae might be used in inhibiting the activity of food-spoiling bacteria. However, there have been no studies examining the antimicrobial potentials of algae originating from Sumbawa Island.

This study aimed to conduct a study of antimicrobial compounds extracted from several types of algae originating from Sumbawa Island, against some food-spoiling bacteria such as *Salmonella thypi*, *Staphylococcus aureus*, *Escherichia coli*, *Enterobacter cloacae* and *Pantoea agglomerans*.

MATERIALS AND METHODS

Sampling Method

Samples were taken in the Luk Coast, Sumbawa region. The samples were classified as red algae, green algae and brown algae. The samples were thoroughly cleaned from any debris and substrates by running the samples under tap water.

Algae Identification

Identification was conducted based on morphological observations and practical guidelines for marine algae identification.

Samples Extraction

The collected samples were crushed using a pestle and mortar before being extracted. Extraction was carried out using the Soxhletation method. This method was carried out to separate bioactive compounds from the samples. The extraction was conducted by using the 1:15 ratio of the sample powder and methanol. The reflux process was carried out for 15 h at 78 ^oC (methanol's boiling point temperature). Filtrates of the reflux process had the basic colors of the algae (red, green and brown). The 150 mL solvents were then separated from the extract by distilling the solvent directly using soxhlet. The purified solvents were subsequently poured into the bottles, followed by repeat heating until there is a sample substance in the flask (Fabrowska *et al.* 2017; Liswandari *et al.* 2018).

Rejuvenation and Manufacture of Test Bacterial Suspensions

Salmonella thypi, *Escherechia coli*, *Enterobacter cloacae*, *Pantoea agglomerans* and *Staphylococcus aureus* bacteria from pure cultures were taken and inoculated on slanted Nutrient Agar (NA) medium. Bacterial cultures were incubated at 37 ^oC for 24 h. The test bacteria having been rejuvenated for 24 h were taken and suspended into NB media (*Nutrient Broth*) then homogenated. Liquid cultures were incubated at 37 ^oC for 24 h. The turbidity of the media indicated the multiplication of bacterial cells.

Algae Extract Preparation in Various Concentrations

The crude extracts were poured into dilution bottles of 200 µL, 400 µL, 600 µL, 800 µL and 1000 µL, respectively, and subsequently added with distilled water until each solution amounted to 1 mL. So, the extracts obtained had respective concentrations of 20%, 40%, 60%, 80% and 100%. The five extract concentrations in the bottles were homogenized.

Antimicrobial Test

The antibacterial activity test was carried out *in-vitro* using the antibacterial sensitivity test with agar diffusion method using disc papers. Nine disc papers were immersed in respective crude extract solution and control solution for 30 min. As much as 100 µL of tested bacteria having been cultured for 24 h were leveled using triangular rods on the NA medium. The disc papers having been soaked in the crude extract and control solutions were affixed to the surface of the media. The disc papers were arranged as not too close to each other so that the inhibition zones did not intersect with each other. Each treatment was conducted with three replications. The Petri dishes were then incubated at 37 ^{o}C. The inhibition zones were observed at the 16th hour. The diameter of the inhibition zone was determined by subtracting the overall diameter (disc paper + inhibition zone) from the diameter of disc paper (6 mm) and diameter of the inhibition zone of solvent (if the solvent has an inhibition zone) (Al-Saif *et al.* 2014; Bhagavathy *et al.* 2011).

Data Analysis

The study was carried out using the Completely Randomized Design (CRD) method with three replications. Data were analyzed using ANOVA (*Analysis of variance*) with a confidence level of 95% by using SPSS 24.0.

RESULTS AND DISCUSSION

Algae Identification

There were 5 algae species identified in the Luk Coast, Sumbawa Regency. The five algae species were derived from the groups consisting of red algae, brown algae and green algae. Identification was carried out based on the morphological characters of each alga (Table 1), showing three species of brown algae and one species of red algae and green algae, respectively.

One identified type of brown algae was *Padina* sp. This species is also found in several Indonesian Waters such as in the Makassar Strait (Hamrun *et al.* 2019), Parigi Moutong Coast, Padang (Dharmayanti *et al.* 2019) and Belitung Lengkuas Coast (Jannah *et al.* 2021). According to Aulia *et al.* (2021), *Padina* sp., is fan-shaped with a diameter of 3-4 cm, yellowish-brown or whitish due to calcification, consisting of flexible rhizoids for attaching themselves to a surface. The tip of the blade is thin-widens and forms a lobe. The lobe is segmented and has white circular stripes, which were not visible, so the talus structure consists only of a blade and a holdfast. The holdfast has the slabs form. *Padina* sp. grows on sandstone substrates.

Besides *Padina* sp. other types of brown algae found were *Sargassum horneri* and *Sargassum crassifolium*. These two types of algae have similar physical characteristics. *Sargassum crassifolium* has a cylindrical or flattened brown thallus with a lush and crossing branch, similar to terrestrial plants. The leaves' shape is broad, oval, or sword-like. On the edges of the jagged leaves, there are some bubbles called vesicles. The function of these air bubbles is to keep the leaves on the surface of the water. The branching alternates regularly. The leaf has an oval and elongated shape with a thallus length of 13.5 - 14 cm. The thallus grows closely resembling a stem. The leaves are sparse and

Table 1 Classification of the five algae species in the Luk Coast

Algae classification					
No.	Class	Ordo	Familia	Genus	Spesies
1	*Ocrophyta*	*Dyctyotales*	*Dyoctaceae*	*Padina*	*Padina* sp.
2	*Chlorophyta*	*Bryopsidales*	*Halimedaceae*	*Halimeda*	*Halimeda opuntia*
3	*Ocrophyta*	*Fucales*	*Sargassaceae*	*Sargassum*	*Sargassum horneri*
4	*Ocrophyta*	*Fucales*	*Sargassaceae*	*Sargassum*	*Sargassum crassifolium*
5	*Rhodophyta*	*Nemaliales*	*Galaxauraceae*	*Galaxaura*	*Galaxaura rugose*

wavy. The tips are curved or tapered. This type of algae is able to grow on coral substrates in choppy areas (Ode 2013). Meanwhile, *Sargassum horneri* has a branched and wide thallus, and lush branching. On the leaves, there were serrations and bubbles. *Sargassum horneri* has a slightly flat and smooth thallus, but the main stem is rounded and slightly rough. The length of the pinnatus alternates is 30-50 cm. According to Gazali *et al.* (2018) *Sargassum* sp. contains bioactive compounds such as alkaloids, phenols and triterpenoids. *Sargassum* sp. also contains bioactive compounds such as saponins, tannins, flavonoids, and phenols (Pangestuti *et al.* 2019). These compounds are able to inhibit the growth of *S. aureus* and *E. coli*.

Green algae, namely *Halimeda opuntia,* was also found on Luk Coast. *Halimeda opuntia* grows on sandy and coral substrates in intertidal to subtidal areas (Kepel *et al.* 2018). *Halimeda opuntia* has the characteristics of a green thallus and is very stiff because it is formed from filaments of branched shiphonus; segmented by tricotome branching; segments form triangles, and segments appear in basal segments. The adhesive device is a filament that comes out of the basal segment and grips the substrate. Segments are calcareous, very rigid, and have a three-indented shape. The arrangements are overlapping, irregular and not located in one branching, so the thallus is not located on one side. *Halimeda opuntia* produces several types of bioactive compounds such as alkaloids, flavonoids and triterpenoids (Hudaifah *et al.* 2020).

The last type of algae found on Luk Coast was *Galaxaura rugose,* which belongs to the red algae group. The algae are beneficial because it contains compounds such as sterols, triterpenoids, flavonoids, tannins and coumarins (Al-Enazi *et al.* 2018). *Galaxaura rugose* has a thick thallus, stiff, dark red-brown color. Branches are cylindrical and have a holdfast to attach to coral reefs. This species is short clumps and reaches 5-7 cm in height. *Galaxaura rugose* grows on every coral reef.

Antimicrobial Activity of Algae Extracts

An antimicrobial activity test is a method to determine the ability of a substance to inhibit bacterial growth. This test was carried out on five types of pathogenic bacteria causing food spoilage, such as *Salmonella thypi*, *Staphylococcus aureus*, *Escherichia coli*, *Enterobacter cloacae* and *Pantoea agglomerans*. The test was carried out on each identified algae extract.

The test results indicated that *Padina sp.* has growth-inhibiting ability against all tested pathogenic bacteria (Table 2). The antimicrobial activities of *Padina* sp. against *P. agglomerans*, *S. aureus* and *S. thypi* showed significantly different values at each treatment concentration with the highest result obtained by the 100% extract concentration of *Padina* sp. In testing against *E. coli*, the 60% and 80% concentrations were not significantly different. The highest inhibiting ability was also shown by the 100% extract concentration. The testing against *E. cloacae* showed that the diameter of the inhibition zone for the 20% and 40% concentrations were not significantly different and the highest inhibiting ability was shown by the 100% extract concentration.

The antimicrobial activity of *Halimeda opuntia* extract also showed growth inhibiting ability against all tested bacteria (Table 3). The formed inhibition zone showed significantly different results for each treatment concentration against all tested bacteria. The highest inhibition zone for all tested bacteria was obtained by the 100% extract concentration.

Table 2 Results of testing the antimicrobial activity of *Padina* sp. extract

Concentration of each treatment (%)	Bacterial inhibition zone diameter (mm)				
	P. agglomerans	*S. aureus*	*E. coli*	*E. cloacae*	*S. thypi*
20	11.67±0.58^{b}	9.33±0.58^{b}	12.33±0.00^{b}	12.67±0.00^{b}	13.00±0.00^{b}
40	15.67±0.58^{c}	12.00±1.00^{c}	13.67±0.58^{c}	13.67±0.58^{b}	14.67±0.58^{c}
60	19.33±0.58^{d}	17.33±0.58^{d}	16.67±0.58^{d}	16.33±1.15^{c}	17.33±1.15^{d}
80	22.67±0.58^{e}	18.67±0.58^{e}	17.67±0.58^{d}	20.00±0.58^{d}	20.33±0.58^{e}
100	25.00±1.00^{f}	20.33±0.58^{f}	22.33±0.58^{e}	22.67±1.15^{e}	22.67±1.15^{f}
K+	29.00±1.00^{g}	28.33±0.58^{g}	28.67±0.58^{f}	28.67±0.58^{f}	28.67±0.58^{g}
K-	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}

Notes: * = Numbers followed by the same letter at the same column indicate no significant difference based on the Duncan test at the level α = 0.05 and n = 3; **K+ = positive control; K-= negative control.

Table 3 Results of testing the antimicrobial activity of *Halimeda opuntia* extract

Concentration of each treatment (%)	Bacterial inhibition zone diameter (mm)				
	P. agglomerans	*S. aureus*	*E. coli*	*E. cloacae*	*S. thypi*
20	8.33± 0.58^{b}	8.67±0.58^{b}	8.67±0.58^{b}	10.33±0.58^{b}	10.67±0.58^{b}
40	11.33±0.58^{c}	13.67±0.58^{c}	14.67±0.58^{c}	14.33±0.58^{c}	14.67±0.58^{c}
60	13.00±0.00^{d}	16.67±0.58^{d}	18.00±0.00^{d}	14.67±0.58^{c}	15.67±0.58^{d}
80	16.00±0.00^{e}	18.00±1.00^{e}	19.33±0.58^{e}	16.33±0.58^{d}	18.33±0.58^{e}
100	21.00±1.00^{f}	22.33±0.58^{f}	22.67±0.58^{f}	20.33±0.58^{e}	21.67±0.58^{f}
K+	28.00±0.00^{g}	28.33±0.58^{g}	28.33±0.58^{g}	28.67±0.58^{f}	29.00±0.00^{g}
K-	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}

Notes: * = Numbers followed by the same letter at the same column indicate no significant difference based on the Duncan test at the level α = 0.05 and n = 3; **K+ = positive control; K-= negative control.

Table 4 Results of testing the antimicrobial activity of *Sargassum horneri* extract

Concentration of each treatment (%)	Bacterial inhibition zone diameter (mm)				
	P. agglomerans	*S. aureus*	*E. coli*	*E. cloacae*	*S. thypi*
20	8.00±0.58^{b}	10.67±0.58^{b}	11.67±0.58^{b}	8.33±0.58^{b}	7.33±0.58^{b}
40	10.75±0.58^{c}	14.33±0.58^{c}	15.33±0.58^{c}	13.00±1.00^{c}	13.33±0.58^{c}
60	12.50±1.15^{d}	16.00±1.00^{d}	17.00±1.00^{c}	16.33±0.58^{d}	16.33±0.58^{d}
80	17.75±0.58^{e}	23.67±0.58^{e}	23.00±1.00^{d}	18.33±0.58^{e}	17.67±0.58^{e}
100	19.00±0.58^{f}	25.67±0.58^{f}	26.00±1.00^{e}	23.67±0.58^{f}	22.67±0. 58^{f}
K+	21.25±0.58^{g}	28.33±0.58^{g}	28.67±0.58^{e}	28.67±0.58^{g}	28.33±0 58^{g}
K-	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}

Notes: * = Numbers followed by the same letter at the same column indicate no significant difference based on the Duncan test at the level α = 0.05 and n = 3; **K+ = positive control; K-= negative control.

The antimicrobial activity of *Sargassum horneri* extract against *P. agglomerans*, *S. aureus*, *E. cloacae* and *S. thypi* showed significantly different results at all concentrations (Table 4). Slightly different results were shown in observations against *E. coli*. The diameter of the inhibition zone showed no significant difference at concentrations 40% and 60%. The 100% extract concentration of *Sargassum horneri* has the same effectiveness with positive control in inhibiting the activity of *E. coli*.

The *Sargassum crassifolium* extract showed growth inhibiting ability against all tested bacteria (Table 5). The diameter of the inhibition zone formed in the four tested bacteria, i.e., *S. aureus*, *E. coli*, *E. cloacae*, and *S. thypi* showed significantly different results in

Table 5 Results of testing the antimicrobial activity from *Sargassum crassifolium* extract

Concentration of each treatment (%)	Bacterial inhibition zone diameter (mm)				
	P. agglomerans	*S. aureus*	*E. coli*	*E. cloacae*	*S. thypi*
20	10.67±0.58^{b}	7.67±0.58^{b}	10.33±0.58^{b}	9.00±1.00^{b}	8.67±0.58^{b}
40	13.67±0.58^{c}	12.00±0.00^{c}	12.00±0.00^{c}	12.67±0.58^{c}	13.33±0.58^{c}
60	18.33±0.58^{d}	14.67±0.58^{d}	14.00±1.00^{d}	16.00±1.00^{d}	16.67±0.58^{d}
80	24.00±0.00^{e}	17.67±0.58^{e}	18.00±0.00^{e}	19.00±0.00^{e}	21.67±0.58^{e}
100	27.00±1.00^{f}	22.33±0.58^{f}	22.33±0.58^{f}	23.67±0.58^{f}	23.67±0.58^{f}
K+	28.67±0.58^{f}	28.33±0.58^{g}	29.00±0.00^{g}	29.00±0.00^{g}	28.33±0.58^{g}
K-	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}

Notes: * = Numbers followed by the same letter at the same column indicate no significant difference based on the Duncan test at the level α = 0.05 and n = 3; **K+ = positive control; K-= negative control.

each extract concentration with the highest inhibition zone obtained by the 100% extract concentration of *Sargassum crassifolium*. However, there was a slight difference in observations against *P. agglomerans* test bacteria, showing that the 100% extract concentration was not significantly different from the control treatment. It means that the 100% concentration of *Sargassum crassifolium* extract has the same inhibitory effect against the *P. agglomerans* test bacteria with the control.

The antimicrobial activity test using *Galaxaura rugose* extract against *E. coli*, *E. cloacae*, and *S. thypi* showed significantly different results among extract concentrations with the highest inhibiting ability obtained by 100% *Galaxaura rugose* extract (Table 6). The 40% and 60% extract concentrations against the *P. agglomerans* and *S. aureus* test bacteria showed a non-significant difference effect, with the highest inhibiting ability shown by the 100% extract concentration.

The five algae extracts showed the ability to inhibit the growth of all tested bacteria. The largest inhibition zone formed was shown by the 100% extract concentration with an inhibition zone diameter range of 19.00-27.00 mm. According to Davis and Stout (1971), the bacterial inhibition zone less than 5 mm was categorized as weak, the 5 - 10 mm zone of inhibition was categorized as moderate, the 10 - 20 mm zone of inhibition was categorized as strong and more than 20 mm was categorized as very strong. It means that the 100% extract concentration of the five algae was in the very strong category.

Table 6 Results of testing the antimicrobial activity from *Galaxaura rugose* extract

Concentration of each treatment (%)	Bacterial inhibition zone diameter (mm)				
	P. agglomerans	*S. aureus*	*E. coli*	*E. cloacae*	*S. thypi*
20	10.67±0.58^{b}	10.33±0.58^{b}	8.00±1.00^{b}	11.67±0.58^{b}	11.67±0.58^{b}
40	12.00±0.00^{c}	13.00±0.00^{c}	9.67±1.53^{c}	14.67±0.58^{c}	14.67±0.58^{c}
60	12.67±0.58^{c}	13.67±1.15^{c}	13.33±0.58^{d}	17.00±0.00^{d}	18.00±0.00^{d}
80	15.33±0.58^{d}	16.33±0.58^{d}	15.00±0.00^{e}	19.33±0.58^{e}	20.67±0.58^{e}
100	19.67±0.58^{e}	20.33±0.58^{e}	20.00±0.00^{f}	23.33±0.58^{f}	24.33±0.58^{f}
K+	28.00±0.00^{f}	28.67±0.58^{f}	28.67±0.58^{g}	28.67±0.58^{g}	28.67±0.58^{g}
K-	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}	0±0^{a}

Notes: * = Numbers followed by the same letter at the same column indicate no significant difference based on the Duncan test at the level α = 0.05 and n = 3; **K+ = positive control; K-= negative control.

The formation of the inhibition zone indicates that the algae extracts from *Padina* sp., *Halimeda opuntia, Sargassum horneri*, *Sargassum crassifolium* and *Galaxaura rugose* have the potential as antibacterial agents. The five algae have capabilities to produce bioactive compounds such as steroids, terpenoids and eicosanoid acid. Different phytochemical contents in *Padina* sp., *Halimeda opuntia, Sargassum horneri*, *Sargassum crassifolium* and *Galaxaura rugose* determine the differences in inhibition abilities because each bioactive compound contained in algae extract has a different action mechanism against bacteria. According to Dewi (2010), flavonoids are able to inhibit the growth of pathogenic bacteria by damaging the cell wall components so that the cell wall layer is not intact causing cell death. Steroids can damage bacterial cell membranes by increasing cell permeability, resulting in cell leakage followed by the release of intracellular material (Cowan 1999). Alkaloid compounds can inhibit the growth of gram-positive and gram-negative bacteria, wherein alkaloids can cause cell lysis and changes in bacterial morphology (Katou 2006). Terpenoids have an antibacterial mechanism by reacting with porin (transmembrane protein) on the outer membrane of the bacterial cell wall, forming strong polymer bonds resulting in the breakdown of porin. The damage of porin, i.e., the entrance and exit for compounds, reduces the permeability of the bacterial cell wall resulting in the bacterial cell being deficient in nutrients so that bacterial growth is inhibited or dies (Cowan 1999).

The ability of the five algae extracts to inhibit the growth of pathogenic bacteria shows that the five algae have the potential to be developed and utilized as natural food preservatives. The bioactive compounds produced through the secondary metabolism of algae were proven by this study to have the ability for inhibiting the growth of food-spoiling pathogenic bacteria. The use of algae as natural food preservatives is expected to replace synthetic food preservatives. Food preservatives derived from natural ingredients are relatively safe to consume and do not cause side effects with the same effectiveness as food preservatives as synthetic ones.

CONCLUSION

The *Padina* sp., *Halimeda opuntia*, *Sargassum horneri*, *Sargassum crassifolium* and *Galaxaura rugose* algae were able to inhibit the growth of pathogenic bacteria such as *Pantoea agglomerans, Staphylococcus aureus*, *Escherecia coli*, *Enterobacter cloacae* and *Salmonella thypi.* Antibacterial activities in the five types of algae have an inhibition zone ability ranging from 19.00 to 27.00 mm at 100% extract concentration with a category of very strong inhibition zone capability. This means that the five types of algae have the potential to be used as natural food preservatives.

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