Journal of Biotechnology **18**(2): 321-329, 2020

CHARACTERIZATION OF KAPPA-CARAGEENAN FROM THE RED ALGA *KAPPAPHYCUS STRIATUM*

Le Dinh Hung[1], ✉, Dang Thi Huong[2]

[1]*NhaTrang Institute of Technology Research and Application, Vietnam Academy of Science and Technology*
[2]*Nguyen Van Troi High School, Nha Trang, Khanh Hoa*

✉To whom correspondence should be addressed. E-mail: ledinhhungims@yahoo.co.uk

Received: 16.9.2019
Accepted: 27.4.2020

SUMMARY

The red alga *Kappaphycus striatum* is an economically important species and extensively cultivated in Vietnam as a material source for carrageenan production. To evaluate carrageenan quality, the characterization of carrageenan extracted from this alga was investigated. As a result, chemical composition of carrageenan consists of 32.4% of 3,6 anhydrogalactose and 24.3% of sulfate. Gelling and metling temperatures are 34.4^{o}C and 55.6^{o}C, respectively. Gel strength of 1.5% is 615 g/cm^2 and average molecular weight is about 267 kDa. Furthermore, FT-IR spectrum showed intense absorption bands at 930 cm^{-1} and 850 cm^{-1} that attributed to 1,4-linked 3,6 anhydro-α-D-galactose and 1,3-linked β-D-galactose-4-sulfate of kappa-carrageenan, respectively. ^{13}C NMR spectrum indicated the signals for anomeric carbon of β-D-galactose-4-sulfate at 102.6 ppm and anomeric carbon of 3,6-anhydro-α-D-galactose at 95.3 ppm. ^{13}H NMR spectrum showed peak signals at 3.57 ppm and 5.1 ppm that corresponds with O-methyl proton of 1,3-linked 6-O-methyl-D-galactose and α-anomeric proton of 3,6 anhydro-α-D-galactose residues, respectively. The results show that the carrageenan from the red alga *Kappaphycus striatus* is kappa-carrageenan with the repeating disaccharide unit consisting of 1,3-linked 6-O-methylated, β-D-galactose-4-sulfate and 1,4-linked 3,6 anhydro-α-D-galactose and did not contain iota-carrageenan. Therefore, this alga may promise to be a good source for carrageenan production for application in food or medicine.

Keywords: *Carrageenophytes, kappa-carrageenan, Kappaphycus striatum, properties, structure*

INTRODUCTION

Carrageenan is extracted from red seaweed of the Rhodophycea family commonly from genera such as *Eucheuma, Solieria, Cripus, Agardhiella, Chondrus, Hypnea, Sarconema* and *Iridaea* (Zia *et al*., 2017). *Eucheuma* and *Kappaphycus* seaweeds are most commonly cultivated seaweed across Malaysia and Southeast Asia (Hurtado *et al*., 2014; Zuldin *et al*., 2016).

Carrageenan is the general name for a group of high molecular weight sulphated polysaccharides obtained by alternate units of D-galactose and 3,6-anhydro-galactose joined by -1,3 and -1,4-glycosidic linkage (Figure 1) (Craigie, 1990). There are three main types of carrageenan, which vary in their degree of sulfation (Prajapati *et al*., 2014; Li *et al*., 2014; Liu *et al*., 2015). Kappa-carrageenan is composed of alternating 3-linked β-D-galactose-4-sulfate and 4-linked 3,6-anhydro-α-D-galactopyranose having one sulfate group per disaccharide repeating unit. Iota-carrageenan is composed of alternating 3-linked β-D-galactose-4-sulfate and 4-linked 3,6-anhydro-α-D-

galactose-2-sulfate having two sulfate groups per disaccharide repeating unit. Lamda-carrageenan has three sulfate groups per disaccharide unit but do not exhibit any 3,6-anhydride bridges contrary to kappa- and iota-carrageenan (Palvi *et al*., 2011). The main differences which effect on the properties of different carrageenans are the number and position of ester sulfate groups and the content of 3,6-anhydro-galactose. Higher levels of ester sulfate resulted in lower gel strength and solubility temperature (Necas, Bartosikova, 2013).

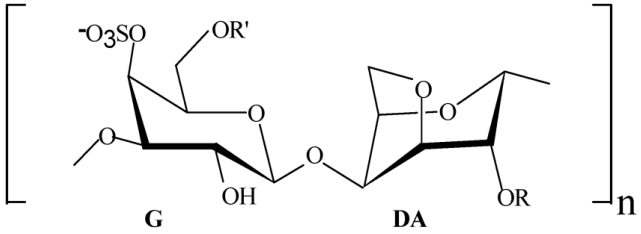

Figure 1. Idealized repeating structure of carrageenan. **G:** 1,3-linked, β-D-galactose and **DA:** 1,4-linked 3,6-anhydo-α-D-galactose; R = R' = H, kappa-carrageenan; R = SO_3^-, R' = H, iota-carrageenan; R' = CH_3, 6-*O*-methylated carrageenan (Craigie, 1990).

Carrageenans possess various pharmaceutical properties including immunomodulatory, antitumor, anticoagulant activities and anti-hyperlipidemic (Prajapati *et al*., 2014). Recently, they have been used in controlled drug release delivery systems (Liang *et al*., 2014; Zia *et al*., 2017; Yegappan *et al*., 2018), as well as enhancing dissolution rates of poorly soluble substances.

The red algae, carrageenophytes, *Kappaphycys alvarezii, Kappaphycus striatum* and *Eucheuma denticulatum* are economically important food species and extensively cultivated in Vietnam as a source of not only carrageenan, but also as a sourec of bioactive compounds for biochemical and medicinal application (Le Dinh Hung *et al*., 2009, 2011, 2015, 2019). However, little information is known about structures of carrageenans from these algae, except for the structure of carrageenan from *E. denticulatum* (Thanh Thi Thu Thuy *et al*., 2007). Thus, the objective of the present research was to elucidate the properties and structure of kappa-carrageenan from the red alga *K. striatum* cultivated at Vanphong Bay, Khanhhoa province, Vietnam, which will provide more valuable information of carrageenan group from these algae for applications in food or medicine.

MATERIALS AND METHODS

The red alga *K. striatum* (F. Schmitz) Doty ex Silva was collected at Vanphong Bay, Khanhhoa province (120°29' N, 109°10' E), Vietnam in March, 2019, brought to the laboratory, and kept at -20°C until use. Rhodizonate natri, acetal, resorcinol, D-fructose was obtained from Merck (Germany).

Extraction of carrageenan

Carrageenan was extracted according to the method of Ohno *et al*. (1994). Dry algal powder was treated in 6% KOH at 80°C for 2 h. Then, the algal powders were collected, washed by distilled water to remove excess alkali and extracted in distilled water at 90°C for 2 h. Thereafter, the solution was filtered and the extract was gellified with 0.2% KCl, frozen and thawed at least twice with distilled water to recover carrageenan, dried at 60°C and ground into power.

Prior to determine chemical and structural characterization, carrageenan sample was repurified by dialysis using a Spectrapor Membrane with Mw cut-off 12,000–14,000 Da, against milli-Q water for 24 h with frequent changes of water. After dialysis,

the sample was filtrated by filter (0.45 µm) and precipitated by absolute ethanol, dried at 40°C and milled into powder.

Determine the 3,6-anhydrogalactose content

3,6-anhydrogalactose content was determined according to the method of Yaphe, Arsenault (1965), using D-fructose as a standard. Briefly, 2 mL of a solution containing up to 0.25 µM of fructose or 3,6-anhydrogalactose (polysaccharide about 100 – 120 µg/mL) was transferred to a boiling tube and covered with a glass marble. The tubes were placed in an ice bath and 10 mL of the resorcinol reagent (9 mL of resorcinol 1.36 mM, 1 mL of acetal 2.78 mM and 100 mL of concentrated HCl) was added. The contents were mixed in the ice bath and cooled for at laest 3 min. The tubes were placed in a 20°C water bath for 4 min and then heated for 10 min at 80°C. It was then cooled for 1.5 min in an ice bath and the absorbance was mearured within 15 min at 555 nm. 3,6-anhydrogalactose content was determined by multiply to value of 1.087. The assay was carried out in triplicate for each test solution.

Determine the sulfate content

Sulfate contents were determined according to the method of Terho, Hartiala (1972), using Na_2SO_4 as a standard. Biefly, 0.5 mL each sample (polysaccharide about 100 – 120 µg/mL), standards and water are pipetted into test tubes and 2.0 mL ethanol is added to each tube. 1.0 mL $BaCl_2$ buffer (10 mL of acetic acid 2 M, 2 mL of $BaCl_2$ 0.005 M, 8 mL of $NaHCO_3$ 0.02M and 80 mL ethanol) and 1.5 mL sodium rhodizonare solution (5 mg of rhodizonate natri dissolved in 20 mL water and 80 mL ethanol) were added to each tube and shaken well. The tubes were allowed to stand 10 min in the dark at room temperature and measured at 520 nm in 30 min. The assay was carried out in triplicate for each test solution.

Determine gelling and melting temperatures of k-carrageenan

Gelling and melting temperatures were determined according to the method of Hellebust, Craige (1978). Gelling temperature was measured with 1.5% kappa-carrageenan solution in 0.2% KCl by a thermometer, corresponded to the introduced glass beads (diam: 4.30 mm; wt: 430 mg) which failed to sink to the bottom of the test tube at an interval of 0.5°C. The melting temperature corresponded to the temperature at which glass beads (diam: 4.30 mm; wt: 430 mg) gradually sank to the bottom of the test tube; temperature gradually was raised at an interval of 0.5°C. The assay was carried out in triplicate for each test solution.

Determine the viscosity-average molecular weight and gel strength

The viscosity-average molecular weight (MW) was obtained from viscometry and extrapolation. The sample was dissolved at initial concentration of 0.1% in 0.1 M NaCl. Viscosity measurements at different dilluted concentrations of kappa-carrageenan in 0.1 M NaCl were done at 25 ± 0.1°C. From efflux time of polymer solution (t) and that of solvent 0.1 M NaCl (t_o), relative viscosity rel = t/t_o was obtained. Specific viscosity was calculated from the relationship sp = rel - 1. Reduced viscosity for a set of polymer solutions was calculated at different concentrations (g/mL). Intrinsic viscosity was then obtained from common ordinate intercept on extrapolation of plots of reduced viscosity versus concentration (Pal *et al*., 2008). The Mw was calculated according to the Mark–Houwink equation for kappa-carrageenan in 0.1 M NaCl at 25°C.

$$[\eta] = K \cdot M_w^{\alpha}$$

$K = 8.84 \times 10^{-3}$ and $\alpha = 0.86$, according to the report of Vreeman *et al*. (1980).

Gel strength was measured on a Rheometer (CR-500DX; Sun Scientific, Tokyo, Japan), using 1.5% kappa-carrageenan solution in 0.2% KCl. The assay was carried out in triplicate for test solution.

Determine structures

Fourier-Transform Infrared (FT-IR)

spectrum of carrageenan sample was recorded on on Bruker mode ALPHA at Nhatrang University.

^{13}C NMR and ^{1}H NMR spectra of carrageenan sample were measured at 80°C with D_2O solvent on Bruker AVANCE 500MHz at Institute of Chemistry (VAST), using aceton as internal standard.

RESULTS AND DISCUSSION

Carrageenan yield, 3,6 anhydrogalactose and sulfate contents, gelling and melting temperatures, gel strength and viscosity-average molecular weight of kappa-carrageenan were shown in Table 1. The data in this study are in range of those reported for the red algae containing carrageenan with 3,6 anhydrogalactose content from 15 to 40%, ester sulfate from 23.1 to 34.5% (Hayashi *et al*., 2007; Nanaki *et al*., 2010; Zia *et al*., 2017), gelling temperature from 32.7 to 34.5°C, melting temperature from 52 to 56°C (Mendoza *et al*., 2002), gel strength from 503 - 1004 g/cm^2 (Le Dinh Hung *et al*., 2009), carrageenan content from 32.5 to 54.3% (Mendoza *et al*., 2002; Zuldin *et al*., 2016) and molecular weight from 100 to 700 kDa (Necas, Bartosikova, 2013). The differences in chemical composition and physical properties between carrageenan samples can be attributed to the extraction methodology used in each study, and the time of algal harvest.

Table1. Characterization of kappa-carrageenan from *K. striatum.*

Carrageenan yield (% dry alga)	3,6-AG content[a] (% carrageenan)	Sulfate content (% carrageenan)	Gelling temp (°C)	Melting temp (°C)	Gel strength (g/cm^2)	MW[b] (kDa)
39.2 ± 3.5	32.4 ± 0.5	24.3 ± 0.8	34.4 ± 0.9	55.6 ± 1.6	615 ± 45	267

[a] AG: anhydrogalactose; [b] MW: viscosity-average molecular weight. Mean ± SEM (n = 3).

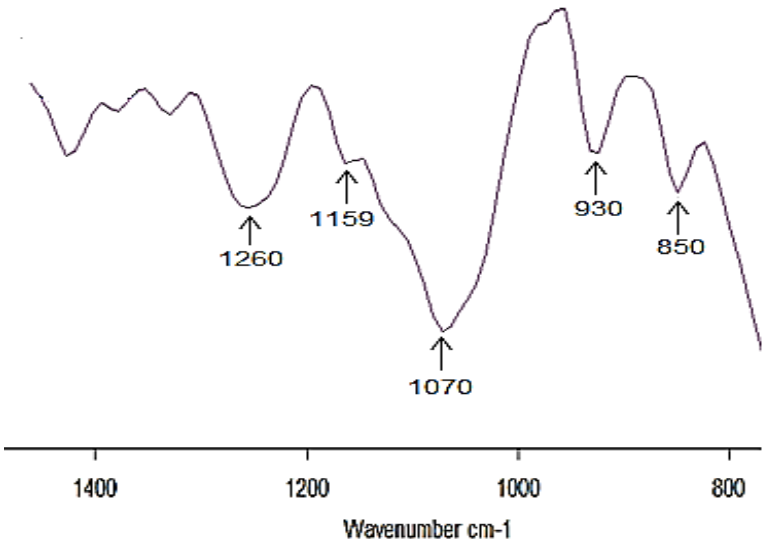

Figure 2. FT-IR spectrum of carrageenan extracted from *K. striatum.*

The infrared spectrum of kappa-carrageenan was shown in Figure 2. As reported previously, the bands at 1260 cm^{-1}, 850 cm^{-1} were assigned respectively to O=S=O symmetric vibration and C_4–O–S stretching vibration, respectively. They stood for total –SO_4 and C_4–O–S of β-D-galactose, respectively. The band at 930 cm^{-1} proved the existence of C–O–C of 3,6-anhydro-

D-galactose (Silva *et al*., 2010). It also displayed an absorbance band at 1159 cm^{-1} due to bridge –O stretch and band at 1070 cm^{-1} was reported to be related to C–O stretch (Mendoza *et al*., 2002; Tranquilan-Aranilla *et al*., 2012). The native carrageenan extracts were composed predominantly of kappa carrageenan with trace amounts of iota-carrageenan that showed at absorption band of 805 cm^{-1} for 3,6 anhydro-α-D-galactose-2-sulfate (Mendoza *et al*., 2002). However, the infrared spectrum in this study showed no appearance of absorption band at 805 cm^{-1}, indicating that kappa-carrageenan sample did not contain iota-carrageenan.

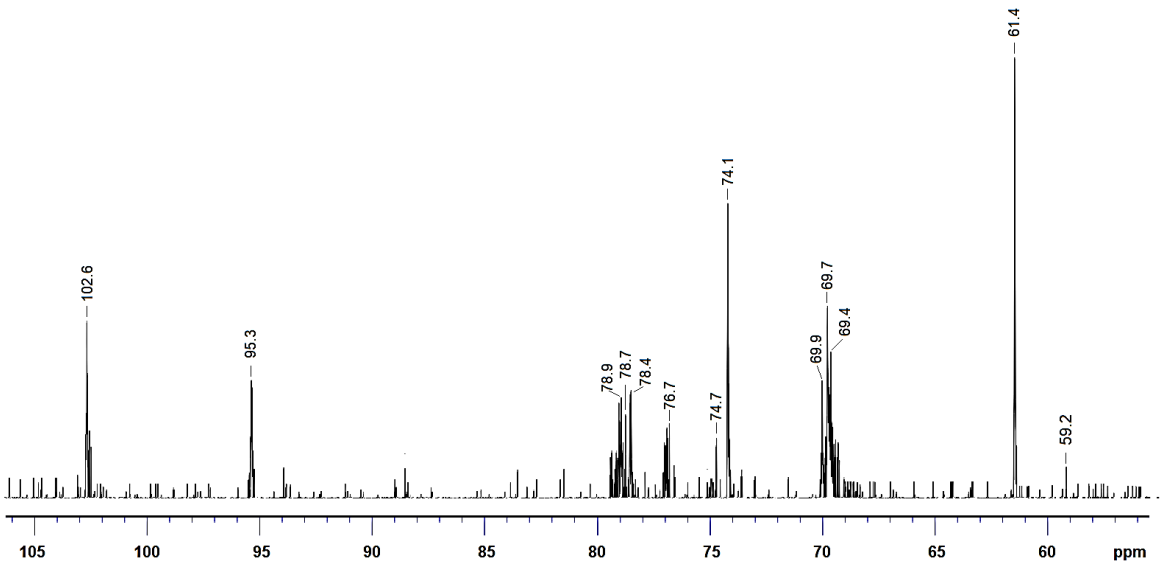

Figure 3. ^{13}C NMR spectrum of carrageenan extracted from *K. striatum.*

Table 2. Chemical shifts (ppm) in the ^{13}C NMR spectrum of kappa-carrageenan extracted from *K. striatum*

Unit	Carbon atom	Kappa-carrageenan	Ref [a]	Ref [b]	Ref [c]
D-Galactose-4-sulfate	C-1	102.6	102.6	102.7	102.9
	C-2	69.7	69.6	69.9	70.1
	C-3	78.7	78.9	79.1	79.5
	C-4	74.1	74.1	74.3	74.5
	C-5	74.7	74.7	75.0	75.2
	C-6	61.4	61.2	61.5	61.7
3,6-anhydro-D-galactose	C-1	95.3	95.2	95.4	95.7
	C-2	69.9	69.9	70.1	70.4
	C-3	78.9	79.1	79.4	79.6
	C-4	78.4	78.2	78.5	78.7
	C-5	76.7	76.7	77.0	77.2
	C-6	69.4	69.4	69.7	69.9

C1–6 shows the carbon numberings.
[a, b, c] Referenced to Mendoza *et al*. (2002), Kolender, Matulewicz (2004) and Tranquilan-Aranilla *et al*. (2012), respectively.

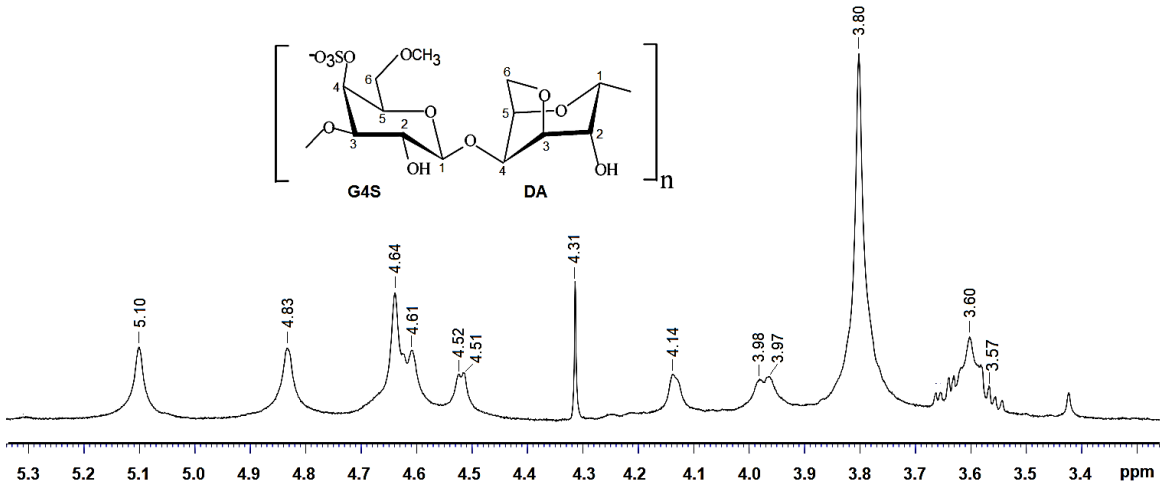

Figure 4. ^{1}H NMR spectrum of carrageenan extracted from *K. striatum.*

Table 3. Chemical shifts (ppm) in the ^{1}H NMR spectrum of carrageenan extracted from *K. striatum.*

Unit	Proton	Kappa-carrageenan	Ref [a]	Ref [b]	Ref [c]
D-Galactose-4-sulfate (G4S)	H-1	4.64	4.75	4.75	5.19
	H-2	3.60	3.74	3.50	3.67
	H-3	3.98	4.10	3.90	3.96
	H-4	4.83	4.95	4.83	4.84
	H-5	3.80	3.93	3.71	3.72
	H-6	3.97	3.93	3.71	3.71
3,6-anhydro-D-galactose (DA)	H-1	5.10	5.24	5.01	5.01
	H-2	4.14	4.12	4.05	4.01
	H-3	4.51	4.30	4.44	4.45
	H-4	4.52	4.75	4.51	4.52
	H-5	4.61	4.65	4.56	4.58
	H-6	4.31	4.25	4.23	4.13

H1–6 shows the proton numberings

[a, b, c] Referenced to Campo *et al*. (2009), Abad *et al*. (2011) and Mahmood *et al*. (2014), respectively.

Figure 3 showed the chemical shifts in the ^{13}C NMR spectrum of kappa-carrageenan that corresponded strong intensity signals of the twelve carbon atoms in the disaccharide repeating unit indicated in Table 2, and agreed closely with reported values (Mendoza *et al*., 2002; Tranquilan-Aranilla *et al*., 2012). On the other hand, the signals at 102.6 ppm and 95.3 ppm of kappa-carrageenan spectra have been reported for anomeric carbon of D-galactose-4-sulfate and 3,6-anhydro-D-galactose, respectively (Mendoza *et al*., 2002). A weak signal at 59.2 ppm is clearly seen in the spectra, which corresponds to the methoxyl group on C-6 of the β-D-galactose-4-sulfate units reported (Mendoza *et al*., 2002). Figure 4 showed the chemical shifts in the ^{13}H NMR spectrum of kappa-carrageenan that corresponded strong intensity signals of the protonss in the disaccharide repeating unit indicated in Table 3 and agreed closely with reported values (Campo *et al*., 2009; Abad *et al*., 2011; Mahmood *et al*.,

2014). On the other hand, the signal at 3.57 ppm corresponds to O-methyl proton of 3-linked 6-O-methyl-D-galactose in kappa-carrageenan, indicated methylation in the C-6 position of D-Galactose-4-sulfate. Similar chemical shift (3.56 ppm, 3.58 ppm and 3.58 ppm) for such O-methyl proton was also reported for 6-O-methyl-D-galactose residue (Abad *et al*., 2011; Mahmood *et al*., 2014), respectively.

Small variations of the chemical shift values in NMR spectra may be due to the samples collected from different sources or experiments were performed in different conditions (Velde *et al*., 2004).

The result showed that structure of carrageenan from *K. striatum* consists of repeating disaccharide unit of β-D-galactose-4-sulfate and 3,6 anhydro-α-D-galactose (Figure 4). Similar structures have been reported for carrageenans from the red algae *Eucheuma denticulatum* (Thanh Thi Thu Thuy *et al*., 2007) and *E. gelatinae* (Tran Dinh Toai *et al*., 2004), except for difference in structure of iota-carrageenan from *Eucheuma* had sulfate group at position C2 of 3,6-anhydro-α-D-galactose residue.

CONCLUSION

Carrageenan from the red seaweed of *K. striatum* is kappa-carrageenan with the repeating disaccharide unit consisting of 1,3-linked 6-O-methylated, β-D-galactose-4-sulfate and 1,4-linked 3,6 anhydro-α-D-galactose. The chemical and structural characteristics of kappa-carrageenan used in this study were similar well to those of the red algae reported, indicating that the red alga *K. striatum* may promise to be a good source for carrageenan production to apply in food or medicine.

Acknowledgements: *This research is supported by Vietnam National Foundation for Science and Technology Development (NAFOSTED) under grant number 106-YS.06-2015.16.*

REFERENCES

Abad L, Saiki S, Nagasawa N, Kudo H, Katsumura Y, Dela Rosa A (2011) NMR analysis of fractionated irradiated -carrageenan oligomers as plant growth promoter. *Rad Physics Chem* 80: 977–982.

Campo VL, Kawano DF, da Silva DB, Carvalho I (2009) Carrageenans: Biological properties, chemical modifications and structural analysis - A Review. *Carbohydr Polym* 77: 167-180.

Craigie JS (1990) Cell walls. In: Cole K.M. and Sheath R.G. (eds), Biology of the red algae. *Cambridge University Press, Cambridge*, pp: 221–257.

Hayashi L, Paula EJD, Chow F (2007) Growth rate and carrageenan analyses in four strains of *Kappaphycus alvarezii* (Rhodophyta, Gigartinales) farmed in the subtropical waters of São Paulo State, Brazil. *J Appl Phycol* 19: 393–399.

Hellebust JA, Craige JS (1978) Handbook of phycological methods. Physiological and biochemical methods. *Cambridge University Press, London* pp: 110–131.

Hurtado AQ, Gerung GS, Yasir S, Critchley AT (2014) Cultivation of tropical red seaweeds in the BIMP-EAGA region. *J Appl Phycol* 26: 707–718.

Kolender A, Matulewicz M (2004) Desulfation of sulfated galactans with chlorotrimethylsilane. Charaterization of B-carrageenan by ^{1}H NMR spectroscopy. *Carbohydr Res* 339: 1619–1629.

Le Dinh Hung, Hori K, Huynh Quang Nang, Tran Kha, Le Thi Hoa (2009) Seasonal changes in growth rate, carrageenan yield and lectin content in the red alga *Kappaphycus alvarezii* cultivated in Camranh Bay, Vietnam. *J Appl Phycol* 21: 265-272.

Le Dinh Hung, Sato Y, Hori K (2011) High-mannose N-glycan-specific lectins from the red alga *Kappaphycus striatum* (Carrageenophyte). *Phytochem* 72: 855-861.

Le Dinh Hung, Hirayama M, Bui Minh Ly, Hori K (2015) Biological activity, cDNA cloning and primary structure of lectin KSA-2 from the cultivated red alga *Kappaphycus striatum* (Schmitz) Doty ex Silva. *Phytochem Let* 14: 99-105.

Le Dinh Hung, Le Thi Hoa, Le Nhu Hau, Dinh Thanh (2019) The lectin accumulation, growth rate, carrageenan yield, and quality from the red alga

Kappaphycus striatus cultivated at Camranh Bay, Vietnam. *J Appl Phycol* 31: 1991-1998.

Li L, Ni R, Shao Y, Mao S (2014) Carrageenan and its applications in drug delivery. *Carbohydr Polym* 103: 1–11.

Liang W, Mao X, Peng X, Tang S (2014) Effects of sulfate group in red seaweed polysaccharides on anticoagulant activity and cytotoxicity. *Carbohydr Polym* 101: 776–785.

Liu J, Zhan X, Wan J, Wang Y, Wang C (2015) Review for carrageenan-based pharmaceutical biomaterials: favourable physical features versus adverse biological effects.

Carbohydr Polym 121: 27–36.

Mahmood WAK, Khan MMR, Yee TC (2014) Effects of reaction temperature on the synthesis and thermal properties of carrageenan ester. *J Phys Sci* 25: 123–138.

Mendoza WC, Montano NE, Ganzon-Fortes ET, Villanueva RD (2002) Chemical and gelling profile of *ice-ice* infected carrageenan from *Kappaphycus striatum* (Schmitz) Doty "sacol" strain (Solieriaceae, Gigartinales, Rhodophyta). *J Appl Phycol* 14: 409–418.

Nanaki S, Karavas E, Kalantzi L, Bikiaris D (2010) Miscibility study of carrageenan blends and evaluation of their effectiveness as sustained release carriers. *Carbohydr Polym* 79: 1157–1167.

Necas J, Bartosikova L (2013) Carrageenan: a review. *Vet Med* 58: 187–205.

Ohno M, Largo DB, Ikumoto T (1994) Growth rate, carrageenan yield and gel properties of cultured kappa-carrageenan producing red alga *Kappaphycus alvarezii* (Doty) Doty in the subtropical waters of Shikoku, Japan. *J Appl Phycol* 6: 1–5.

Pal S, Sen G, Mishra S, Dey RK, Jha U (2008) Carboxymethyl tamarind: Synthesis, characterization and its application as novel drug-delivery agent. *J Appl Polym Sci* 110: 392–400

Prajapati DV, Maheriya PM, Jani GK, Solanki HK (2014) Carrageenan: a natural seaweed polysaccharide and its applications. *Carbohydr Polym* 105: 97–112.

Pavli M, Baumgartner S, Kos P, Kogej K (2011) Doxazosin-carrageenan interactions: A novel approach for studying drug-polymer interactions and relation to controlled drug release. *Int J Pharm* 421: 110–119.

Silva FRF, Dore CMPG, Marques CT, Nascimento MS, Benevides NMB, Rocha HAO (2010) Anticoagulant activity, paw edema and pleurisy induced carrageenan: Action of major types of commercial carrageenans. *Carbohydr Polym* 79: 26–33.

Terho TT, Hartiala K (1972) Method for determination of sujphate content of glycosaminoglycans. *Anal Biochem* 41: 471-476.

Thanh Thi Thu Thuy, Tran Thị Thanh Van, Bui Minh Ly, Pham Duc Thinh (2007) Structure of carrageenan from *Eucheuma denticulatum***.** *Tuyển tập Báo cáo Hội nghị Quốc gia "Biển Đông-2007"*: 207-212.

Trần Đình Toại, Nguyễn Xuân Nguyên, Phạm Hồng Hải, Nguyễn Bích Thủy, Trần Thị Hồng (2003) Nghiên cứu carrageenan từ Rong Hồng vân *Eucheuma gelatinae* vùng biển Việt Nam. *Báo cáo Khoa học, Hải Phòng*: 185-204.

Tranquilan-Aranilla C, Nagasawa N, Bayquen A, Rosa AD (2012) Synthesis and characterization of carboxymethyl derivatives of kappa-carrageenan. *Carbohydr Polym* 87: 1810–1816.

Velde F, Pereirac L, Rollema HS (2004) The revised NMR chemical shift data of carrageenans. *Carbohydr Res* 339: 2309–2313.

Vreeman HJ, Snoeren THM, Payens TAJ (1980) Physicochemical investigation of k-carrageenan in the random state. *Biopolym* 19: 1357–1374.

Yaphe W, Arsenault GP (1965) Improved resorcinol reagent for the determination of fructose, and of 3,6-anhydrogalactose in polysaccharides. *Anal Biochem* 13: 143–148.

Yegappan R, Selvaprithiviraj V, Amirthalingam S, Jayakumar R (2018) Carrageenan based hydrogels for drug delivery, tissue engineering and wound healing. *Carbohydr Polym* 198: 385-400.

Zia KM, Tabasum S, Nasif M, Sultan N, Aslam N, Noreen A, Zuber M (2017) A review on synthesis, properties and applications of natural polymer based carrageenan blends and composites. *Int J Biol Macromol* 96: 282–301.

Zuldin WH, Yassir S, Shapawi R (2016) Growth and biochemical composition of Kappaphycus (Rhodophyta) in customized tank culture system. *J Appl Phycol* 28: 2453–2458.

Journal of Biotechnology **18**(2): 321-329, 2020

MÔ TẢ ĐẶC TÍNH CỦA KAPPA-CARRAGEENAN TỪ RONG ĐỎ *KAPPAPHYCUS STRIATUM*

Lê Đình Hùng[1], Đặng Thị Hương[2]

[1] Viện Nghiên cứu và Ứng dụng Công nghệ Nha Trang, Viện Hàn lâm Khoa học và Công nghệ Việt Nam
[2] Trường Trung học Phổ thông Nguyễn Văn Trỗi, Nha Trang, Khánh Hòa

TÓM TẮT

Rong đỏ *Kappaphycus striatum* là loài rong kinh tế đang được nuôi trồng rộng rãi ở Việt Nam làm nguồn nguyên liệu để sản xuất carrageenan. Để đánh giá chất lượng carrageenan, đặc tính của carageenan được chiết từ mẫu rong này đã được khảo sát. Kết quả cho thấy rằng thành phần hóa học của carrageenan bao gồm 32,4% 3,6-anhydrogalactose và 24,3% sulfate. Nhiệt độ tạo gel và nhiệt độ tan là 34,4^{o}C và 55,6^{o}C, tương ứng. Độ bền gel 1,5% là 615 g/cm^{2} và khối lượng phân tử trung bình là 267 kDa. Phổ FT-IR đã cho thấy các dải hấp thụ mạnh ở 930 cm^{-1} và 850 cm^{-1} tương ứng với 3,6 anhydro-α-D-galactose liên kết ở vị trí 1,4 và β-D-galactose-4-sulfate liên kết ở vị trí 1,3 của kappa-carrageenan. Phổ ^{13}C NMR đã chỉ ra các tín hiệu cho các carbon anomer của β-D-galactose-4-sulfate ở 102,6 ppm và 3,6-anhydro-α-D-galactose ở 95,3 ppm. Phổ ^{13}H NMR đã cho thấy các đỉnh tín hiệu ở 3,57 ppm và 5,1 ppm tương ứng với proton O-methyl của 6-O-methyl-D-galactose liên kết ở vị trí 1,3 và proton α-anomer của 3,6 anhydro-α-D-galactose. Các kết quả này cho thấy rằng carrageenan từ rong đỏ *K. striatum* là kappa-carrageenan với các nhóm disaccharide lập lại bao gồm 6-O-methyl-β-D-galactose-4-sulfate liên kết 1,3 và 3,6 anhydro-α-D-galactose liên kết 1,4 và không chứa iota-carrageenan. Vì vậy, rong này hứa hẹn sẽ là một nguồn giá trị để sản xuất carrageenan cho sử dụng trong thực phẩm hoặc y học.

Từ khóa: *Carrageenophytes, cấu trúc, kappa-carrageenan, Kappaphycus striatum, tính chất*