Carrageenan, a marine sulphated polysaccharide, chemical modifications and anti-viral prospects - Review

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ABSTRACT: Marine realm constitutes diverse resource of bioactive compounds with significant bioactivity. Sulfated polysaccharides from seaweeds such as carrageenanare reported to pose excellent rheological properties. Despite thewide spectrum of usage as additives in the food, cosmetics and pharmaceutical industry, they acquire great developmental prospects in the area of virology. Modulation of structural characteristic of carrageenan have demonstrated remarkable rheological and chemical properties. This review focus on the modulation of structural characteristics of carrageenan with recent insights on enhancement of anti-viral property which gains significance for their application in the area of virology. Presently, there exists scant information on the pharmacology of carrageenan. Therefore, extensive research is required to explore them for the formulation of effective and safe drugs to combat the existing ailments and several novel diseases.Sulfated polysaccharides including carrageenan demonstrate a number of biological activities including antiviral properties.From the reviewed literature, it is concluded that chemical modulation of carrageenan structural characteristichasmanipulated its rheological and chemical properties and are found effective against several group of viruses including influenza A virus, human immunodeficiency virus and numerous others. The current mini review presents the anti-viral prospectives of carrageenan and induces insights to develop effective medication against novel viruses.

KEYWORDS: Antiviral, bioactive compounds, carrageenan,seaweeds,sulphated polysaccharides.

I. INTRODUCTION

Structurally, carrageenan exists as a high molecular weight, anionic, hydrophilic sulfated D-galactans constituted with alternating disaccharide units with repeating 3-linked α-D-glucopyranose and 4-linked α-galactopyranose. Carrageenan is classified based on the variations in their structural characteristics. Sulphation patterns and the existence of anhydrogalactose residues on D-units remain as a fundamental thumb rule for their classification. Presently 15 different carrageenan structures remain in existence [1]. Amongst the carrageenan forms; κ, ι and λ gains industrial value and market demand. The red seaweed *Kappaphycus alvarezii* serve as a paramount source of k-carrageenan extraction [2]. Their structure exists as alternating 3-linked β-D-galactose 4-sulfate and 4-linked anhydro galactose units [3].

The ι- form of carrageenan pose an extra sulfate group at C2 (O) position of the anhydrogalactose units, as a consequence they constitute two sulfates in each disaccharide repeating unit. The ι-form of carrageenan was reported to have homogeneous and flexible properties compared to k-carrageenan [4]. Structurally, the λ- form of carrageenans encompass three sulfate groups in each disaccharide unit constituting the third sulfate group aligned at the C6 position of the 4-linked unit, with no 3, 6-anhydride residues. λ-carrageenan is prepared from *Gigartina* and *Chondrus*genera predominantly [5]. Results of several studies have demonstrated that chemical modification of their structural characteristicsprovides various alternatives forms. In a recent study, in a hand sorted extraction, λ-carrageenan and their subsequent treatment with alkaline borohydride helped in conversion of the 4-linked units to the 3,6-anhydride ring form, obtaining λ-carrageenan albeit no contamination of k- or i-carrageenans were observed [6]. Natural carrageenan remains as mixtures of various hybrid types, such as k /β-hybrids [7] k /ι-hybrids [8-11], k /μ-hybrids [12], or ν/ι –hybrids [13]. Additionally, observations of some study hypothesized that solvents like pyruvic acid acetyl components and the existence of trace quantity of sugars are observed to influence their structural complexity [14].Significant chemical modifications to the carrageenan structural characteristic have modulated their rheological and chemical properties. Modification of their

structural characteristic influencing their physical and chemical properties is cited here with observations of few relevant studies. In 1978, Guisely[15] proposed the synthesis of ι-form of carrageenan i·e·,when acquired mechanical deformation, unsuitable extent of syneresis from aqueous gels of i- form of carrageenan-constituting hydroxyalkyl groups are found to be formed. Studies also demonstrated that gels formulated from hydroxyalkyl ι-carrageenan derivatives showed decreased syneresis. From many literatures, it is evident that alkali modifications gains significance as a best extraction protocol of carrageenan. In an observation from a recent study, κ- and i- form of carrageenan treated with 1 M sodium hydroxide at 80 °C pose cyclization. Ciancia *et al*. [16] observed that a pseudo first –order kinetics reactions are formed when 3, 6-anhydro-α-D-galactose units are formed from α-D-galactose 6- and ι- form of carrageenan pose this chemical change in larger extend compared to k family. Provided with milder experimental conditions gelling properties of crude ν/μ-hybrid carrageenan is increased by employing hot alkaline treatments industrially. This treatment is adopted by industries to increase the gel strength [17]. In another study, at 2 M sodium hydroxide concentration, a higher extend of cyclization was observed and employing microwave heating technique accelerated and improved the rate of κ-carrageenan reactions to a remarkable ratio [18].

Anti-viral potential of marine sulfated polysaccharides with special reference to carrageenan: Sulfated polysaccharides extracted from marine environment are found to poses pronounced anti-viral potential. In the history of virology, there are evidences to substantiate the inhibitory effect of marine sulfated polysaccharide against several groups of viruses. Research in studying anti-viral effects of sulfated polysaccharides and various polyanionic substances from seaweed species has spurred in light of the study conducted in the recent decades. One of the prominent studies conducted [19], demonstrated the inhibitory effect of heparin, against HSV in leukocytes cultures, hypothesize this inhibitory action prevailed as a consequence of virus attachment to cellular surface interference. Consequently, the motivation to examine the progression of algal polysaccharide's anti-viral effects has spurred in few years back with the reports. From the data, observation of effects on viral replication exists by a number of chemical reactions that involve unique structural characteristic of the polysaccharides without the aid of non-specific interactions [20, 21]. Some of the recent investigations concerning the algal polysaccharide's structural necessities, carrageenan for their antiviral activity and mechanistic role in their modes of action are cited. In Vero cells, carrageenan from red seaweed *Gymnogongrus griffithsiae* inhibited DENV-2 replication [22, 23]. Their effects were found to pose least inhibitory functionality against DENV-3 and DENV-4 and they are demonstrated as inactive against DENV-1 infection [22]. The study indicated that the interference of polysaccharides with DENV-2 adsorption and their internalization to the cells and examined as effective when treated with the virus.

Chemical modification of carrageenan structural characteristics for enhanced anti-viral activity: There is a requisite for chemically modifying the structural characteristic of carrageenan for ascertaining desirable bioactivity. In the biomedical and pharmaceutical research, the usage of modified carrageenan has been explored in several ways which are subjected to be utilized in biomedical application. Here are few studies cited showing how chemical modification of carrageenan enhances anti- viral potential. Study conducted by Yamada *et al*. [24] demonstrated that, low molecular weight carrageenan and their sulfated constituents formulated by depolymerization, posed anti-HIV properties and the biological functionality have improved by the sulphation of i-form of carrageenan.Using DMF as solvent and 4-dimethyl- aminopyridine (DMAP)/tributylamine (TBA) as catalysts, the depolymerized carrageenans were acylated with the aid of carboxylic acid anhydrides [25] showed remarkable anti-viral activity, reported in another study. Additionally, the acylation of low molecular weight carrageenan demonstrated the potentiation of anti-HIV activity. In another study, a succinate diester spacer was used to allocate single molecule of AZT to a tirade of ι-form of carrageenan units. When MT-4 cells pre-incubated with the conjugate, synergistic activity of the two drugs was demonstrated prior to HIV-1-infection.The ι- form of carrageenan– AZT conjugate permitted the reduction of the doses.

Status of virology related studies in carrageenan: Recent advancements in biomedical research of carrageenan in area of virology are cited. In a study conducted by Leibbrandt[26], in view of the outbreak of flu pandemic in 2009 and the incidence of oseltamivir-resistant H1N1 influenza strains conjure the necessity for alternative treatments. As a consequence of this, ι-carrageenan formulated in a commercially containing nasal spray demonstrated pronounced inhibition against influenza virus infection. It was concluded that the nasal spray formulated withι- form of carrageenan act as an alternative to neuraminidase inhibitors. It needs to be subjected for treatment of influenza A in humans since alternative treatment for influenza were found rare. In a study [27], the administration of carrageenan nasal spray in children and adults enduring from virus common cold reduced duration of ailments, and improved viral clearance and decreased relapses of symptoms. It was observed that carrageenan nasal spray acted as a potent treatment of common cold in infants and adults. In view of these

studies, carrageenan acts as a prospective marine bioactive compound that posse better scope for screening them for anti-viral activity.

II. CONCLUSION

An effective medication against existing and emerging novel viral infections remain as a problematic scenario in biomedicine during these days. In view of this problem, this review confers on the advancement of research in carrageenan holding emphasis to area on the significance of carrageenan in the context of virus-related studies. This is an ingenious approach by giving a briefing on how bioactive potential including anti-viral property that are ascertained by chemical modification of structural characteristic of biomolecules. The literatures cited here apparently supports the anti-viral potential of carrageenan against few viruses including influenza A virus and pandemic H1N1. These observations are really a milestone in biomedical research. Therefore, extensive research is required in the area of chemical modification of carrageenan to develop sequence of pronounced bioactivity. It is well evident from the studies that modifying the sequence quality of carrageenan or chemical modifications to oligopeptide level may enhance anti-viral activity against broad spectrum of virus to compact several novel diseases including COVID-19.These approaches would definitely lead to advancements in pharmaceutical applications and in the field of biomedicine today and in future.

Abbreviations:AZT:Aazidothymidine; *DMF: Dimethylformamide;* DENV-2: Dengue virus type 2; H1N1: Influenza A virus subtype H1N1; HSV: Herpes simplex virus.

REFERENCES

1. Lahaye M (2001) Developments on gelling algal galactans, their structure and physico-chemistry. J App Phyco 13(2) 173-184.https://doi.org/10.1023/A:1011142124213.
2. Anderson NS, Dolan TC and Rees DA (1973). Carrageenans. Part VII. Polysaccharides from Eucheuma spinosum and Eucheumacottonii. The covalent structure of ι-carrageenan. **J Chem Soc Perkin Trans** 1 2173-2176.https://doi.org/10.1039/P19730002173**.**
3. Estevez JM, Ciancia M, and Cerezo AS (2000). The system of low-molecular-weight carrageenans and agaroids from the room-temperature-extracted fraction of Kappaphycus alvarezii. Carbohydr Res 325(4), 287-299. https://doi.org/10.1016/S0008-6215(00)00006-9.
4. Funami T, Hiroe M, Noda S, Asai I, Ikeda S Nishinari K (2007) Influence of molecular structure imaged with atomic force microscopy on the rheological behavior of carrageenan aqueous systems in the presence or absence of cations. Food Hydrocoll. 21(4) 617-629. https://doi.org/10.1016/j.foodhyd.2006.07.013
5. Zhou G, Sheng W, Yao W and Wang C (2006). Effect of low molecular λ-carrageenan from Chondrus ocellatus on antitumor H-22 activity of 5-Fu. Pharmacol. Res. 53(2), 129-134. https://doi.org/10.1016/j.phrs.2005.09.009.
6. Doyle JP, Giannouli P, Rudolph B and Morris ER (2010) Preparation, authentication, rheology and conformation of theta carrageenan. Carbohydr. Pol. 80(3), 648-654.https://doi.org/10.1016/j.carbpol.2009.10.029
7. Yang B, Yu G, Zhao X, Ren W, Jiao G, Fang L, Wang Y, Du G, Tiller C, Girouard G and Barrow CJ (2011). Structural characterisation and bioactivities of hybrid carrageenan-like sulphated galactan from red alga Furcellarialumbricalis. Food chem. *124*(1), 50-57.https://doi.org/10.1016/j.foodchem.2010.05.102
8. Van de Velde F, Antipova AS, Rollema HS, Burova TV, Grinberg NV, Pereira L, Gilsenan PM, Tromp RH, Rudolph B and Grinberg VY (2005) The structure of κ/ι-hybrid carrageenans II. Coil–helix transition as a function of chain composition. Carbohydr. Res. 340(6) 1113-1129.https://doi.org/10.1016/j.carres.2005.02.015
9. Chopin T, Kerin BF and Mazerolle R. (1999). Phycocolloid chemistry as a taxonomic indicator of phylogeny in the Gigartinales, Rhodophyceae: a review and current developments using Fourier transform infrared diffuse reflectance spectroscopy. Phycol. Res. 47(3) 167-188.https://doi.org/10.1046/j.1440-1835.1999.00170.x.
10. Hilliou L, Larotonda FDS, Abreu P, Ramos AM, Sereno AM and Gonçalves MP (2006). Effect of extraction parameters on the chemical structure and gel properties of κ/ι-hybrid carrageenans obtained from Mastocarpusstellatus. Biomol. Eng. 23(4) 201-208.https://doi.org/10.1016/j.bioeng.2006.04.003.
11. Hilliou L, Larontonda FDS, Sereno AM and Gonçalves MP (2006). Thermal and viscoelastic properties of κ/ι-hybrid carrageenan gels obtained from the Portuguese seaweed Mastocarpusstellatus. J. Agric. Food Chem, *54*(20), 7870-7878.https://doi.org/10.1021/jf0612934.
12. Jouanneau D, Guibet M, Boulenguer P, Mazoyer J, Smietana M and Helbert W (2010) New insights into the structure of hybrid κ-/μ-carrageenan and its alkaline conversion. Food Hydro. 24(4) 452-461.https://doi.org/10.1016/j.foodhyd.2009.11.012
13. van de Velde F (2008) Structure and function of hybrid carrageenans. Food Hydrocoll 22(5) 727-734.https://doi.org/10.1016/j.foodhyd.2007.05.013

14. Yu G, Hu Y, Yang B, Zhao X, Wang P, Ji G, Wu J and Guan H (2010) Extraction, isolation and structural characterization of polysaccharides from a red alga Gloiopeltisfurcata. J. Ocean Univ. China. 9(2) 193-197.https://doi.org/10.1007/s11802-010-0193-7
15. Guiseley KB (1978) U.S. Patent No. 4,096,327. Washington, DC: U.S. Patent and Trademark Office
16. Ciancia M, Noseda MD, Matulewicz MC and Cerezo AS (1993) Alkali-modification of carrageenans: mechanism and kinetics in the kappa/iota-, mu/nu-and lambda-series. Carbohydr. Polym. 20(2), 95-98.https://doi.org/10.1016/0144-8617(93)90083-G
17. Viana AG, Noseda MD, Duarte MER and Cerezo AS (2004) Alkali modification of carrageenans. Part V. The iota–nu hybrid carrageenan from Eucheumadenticulatum and its cyclization to iota-carrageenan. Carbohydr. Poly. 58(4) 455-460.https://doi.org/10.1016/j.carbpol.2004.08.006
18. Navarro DA and Stortz CA (2005) Microwave-assisted alkaline modification of red seaweed galactans. Carbohydr. Poly. 62(2) 187-191.https://doi.org/10.1016/j.carbpol.2005.07.009
19. Nahmias AJ and Kibrick S (1964). Inhibitory effect of heparin on herpes simplex virus. J Bacteriol 87(5) 1060-1066.doi: 10.1128/JB.87.5.1060-1066.1964.
20. Witvrouw M and De Clercq E (1997) Sulfated polysaccharides extracted from sea algae as potential antiviral drugs. Gen Pharmacol 29(4) 497-511.https://doi.org/10.1016/S0306-3623(96)00563-0
21. Ghosh T, Chattopadhyay K, Marschall M, Karmakar P, Mandal P and Ray B (2009) Focus on antivirally active sulfated polysaccharides: from structure–activity analysis to clinical evaluation. Glycobiology 19(1) 2-15.https://doi.org/10.1093/glycob/cwn092
22. Talarico LB, Pujol CA, Zibetti RGM, Faria PCS, Noseda MD, Duarte MER and Damonte EB (2005) The antiviral activity of sulfated polysaccharides against dengue virus is dependent on virus serotype and host cell. Antiviral Res 66(2-3) 103-110.https://doi.org/10.1016/j.antiviral.2005.02.001
23. Talarico LB, Duarte ME, Zibetti RG, Noseda MD and Damonte EB (2007) An algal-derived DL-galactan hybrid is an efficient preventing agent for in vitro dengue virus infection. Planta Med 2007; 73(14): 1464-1468.DOI: 10.1055/s-2007-990241
24. Yamada T, Ogamo A, Saito T, Watanabe J, Uchiyama H and Nakagawa Y (1997). Preparation and anti-HIV activity of low-molecular-weight carrageenans and their sulfated derivatives. Carbohydr Pol 32(1) 51-55.https://doi.org/10.1016/S0144-8617(96)00128-2
25. Yamada T, Ogamo A, Saito T, Uchiyama H and Nakagawa Y (2000) Preparation of O-acylated low-molecular-weight carrageenans with potent anti-HIV activity and low anticoagulant effect. Carbohydr Poly, 41(2), 115-120.https://doi.org/10.1016/S0144-8617(99)00083-1
26. Leibbrandt A, Meier C, König Schuster M, Weinmüllner R, Kalthoff D, Pflugfelder B, Graf P, Frank Gehrke B, Beer M, Fazekas T and Unger H (2010) Iota-carrageenan is a potent inhibitor of influenza A virus infection. PloS one 5(12) e14320.https://doi.org/10.1371/journal.pone.0014320
27. Koenighofer M, Lion T, Bodenteich A, PrieschlGrassauer E, Grassauer A, Unger H, Mueller CA and Fazekas T (2014) Carrageenan nasal spray in virus confirmed common cold: individual patient data analysis of two randomized controlled trials. MultidiscipResp Med 9(1) 57.https://doi.org/10.1186/2049-6958-9-57