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Marine Polysaccharide Degrading Bacteria - A Source of Nutraceuticals

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Abstract: Marine resources are very important in order to fulfil the large percentage of the nutraceutical products of the global market. The nutraceuticals are obtained from a diverse range of resources such as seaweeds, marine invertebrates, fungi, and bacteria that offer a myriad of bioactive molecules. However, emphasis on the use of polysaccharide degrading bacteria amongst other marine heterotrophic bacteria as a candidate for nutraceutical has not been laid. The polysaccharide degrading bacteria are diverse, ubiquitous, and have immense potential in the nutraceutical industry as they have previously exhibited anti-inflammatory, anticarcinogenic, and antioxidant activity. Polysaccharide degrading enzymes play a crucial role in shaping the complex marine microbial loop. Enzymes such as agarases, chitinases, xylanases, carrageenases and fucoidanases have from marine microorganisms have demonstrated the substrate degradation abilities that have been exploited to obtain nutraceuticals of industrial importance. This chapter review focuses on the properties of the role of polysaccharide degrading bacterial enzymes as nutraceuticals and discusses their symbiotic interactions. In addition, the indirect use of polysaccharide degrading bacteria as nutraceuticals has been highlighted. Finally, the challenges and scope for further research in this field has also been discussed.

Keywords: Nutraceutical, Polysaccharide-Degrading-Bacteria, Marine-Resources, Bioactive

1. Introduction

The irony of the Oceans concerning nutraceutical research is that, although the Oceans are vast and deep constituting the major area of our planet, the research on the potential resources that could be considered as therapeutic food is not completely been explored and is still shallow and at an infancy stage. Marine organisms constitute approximately 80% of the total biota with a wide range of bioactive compounds that could unlock the answers to problems regarding several diseases in human beings. The word nutraceutical was first coined by Stephen de Felice in 1989 [15] blending nutrient and pharmaceutical which simply means nutrients with medicinal abilities to cure diseases [19]. With new and emerging diseases witnessed in the last decade, there is a sharp shift from the traditional and allopathic treatment to diet based cure [2], especially for nutrition-related chronic diseases. Nutraceuticals have been designed over the years to reduce stress levels, increase longevity, and improve physical health and general wellbeing [10].

The complex polysaccharides are components of seaweeds, the exoskeleton of crustacean providing structural integrity [27]. Widely found polysaccharide in the marine environment is chitin- a naturally distributed non-toxic biopolymer that is found second most abundantly after cellulose. Alginate is a complex copolymer that is majorly found in algae and marine invertebrates mostly brown algae. Agar is another abundantly found complex polysaccharide that is present in seaweed [56]. The complex polysaccharides act as structural components in the marine ecosystem and in turn lay a platform for the epiphytic association of multiple polysaccharide degrading bacteria [42].

The marine ecosystem is known for its diversity in microorganisms especially the bacterial population that could be investigated to develop safe and effective probiotics. The probiotics obtained from marine bacteria have higher chances of novelty that could be used effectively in fighting the menace caused by antibiotic resistance [1]. Enzymes from microorganisms have unique properties that have recently gained attention. Also, the exopolysaccharides derived from a marine bacterium that is known as polysaccharide degraders have been researched as a source of new marine natural compounds. Reports have highlighted the marine organisms that have been a potential source of several bioactive compounds (Table 1).

Table 1. *Nutraceuticals from marine organisms.*

Nutraceutical	Application	Marine organism	Mode of action	Reference
fucoidan-like polysaccharide	Inflammatory diseases	*Sargassum horneri*	Activation of NF-κB, p50, p65, and p38, and the phosphorylation of extracellular signal-regulated kinases.	Sanjeewa et al., 2017 [48]
Sulfated polysaccharide	Antidiarrheal effects on cholera toxin	*Gracilaria caudate*	cholinergic mechanisms	Costa et al., 2016 [12]
Water-soluble polysaccharide	Functional feeds for the enhancement of average weight and specific growth rate when included in the diets of *Penaeus monodon*	*M. pyrifera* and *U. pinnatifida*	Scavenging activity in midgut gland	Díaz et al., 2017 [17]; Niu et al., 2015 [41]
Sulfated polysaccharide	An inexpensive dietary supplement to everyday food intake	Red seaweed *Laurencia papillosa*	Dose-dependent antiproliferative activity to human breast cancer cell line MCF-7.	Ghannam et al., 2018 [20]
Exopolysaccharide of brown Seaweed	Anticancer activity toward a colon cancer cell line	*Sargassum longifolium*	Encapsulation with a nanoemulsion and nanostructured lipid carrier ensures slow and steady release	Shofia et al., 2018 [50]
Sulfated polysaccharide	lipid-lowering and antiobesity properties in rats	*Caulerpa cupressoides* var. lycopodium	Reduction in total cholesterol, low-density lipoprotein cholesterol (LDL-C), and triglyceride levels and an increase in high-density lipoprotein cholesterol (HDL-C)	Kolsi et al., 2015 [35]
Sulfated polysaccharide		Brown seaweed *Ascophyllum nodosum*	Antitumor activity toward sarcoma-180	Jiang et al., 2014 [31]
Sulfated polysaccharide		*U. lactuca*	Improvements in lipid profiles and to exhibit antioxidant activity	Hassan et al., 2011 [24]
ι-, κ-, λ-, μ-, and ν-carrageenans	Antiviral against herpes simplex viruses type 1 (HSV-1) and type 2 (HSV-2)	*Gigartina skottsbergii*	Antiviral activities	Carlucci et al., 1999 [8]

Marine life forms such as complex polysaccharide degrading bacteria have evolved to survive and thrive in an extreme environment such as high salinity, low nutrients, high pressures, and temperatures. In the bargain, these bacteria have developed unique metabolic and physiological properties and house several unique and unraveled metabolites that are usually not found in the terrestrial ecosystem [51, 16, 38]. The bioactive compounds produced by marine polysaccharide degrading bacteria can be polysaccharides, proteins, peptides, enzymes, fatty acids, probiotics, polyphenols, vitamins, and minerals.

2. Polysaccharide Degrading Enzymes

The degradation or hydrolysis of complex polysaccharides such as agar, chitin, xylan, alginate, and carrageenan occur due to the combined effect of carbohydrate-active enzymes or CAZymes that are produced by multiple polysaccharide degrading bacteria [26, 27].

2.1. Agarases

Is obtained from multiple polysaccharide degrading bacteria that use this enzyme to degrade agar- a stabilizing and gelling agent that has a promising role as a functional food and nutraceutical. Agarases is isolated from bacterial species belonging to the genus *Cellulophaga, Microbulbifer*, *Pseudoalteromonas, and Pseudomonas* [45]. Agar has been studied for its ability to decrease blood glucose levels and prevents the aggregation of red blood cells. The role of agar in the suppression of pro-inflammatory cytokines and nitric oxide (NO)-producing enzymes has also been realized. Likewise, the antioxidant as well as anti-tumor activity of agar explains the need for the enzymes that can hydrolyze the otherwise insoluble complex polysaccharide [51]. In the nutraceutical industry, agar has several applications as a gelling and stabilizing agent as it maintains viscosity was the first phycocolloid to be used in the food industry [25].

2.2. Chitinases

An enzyme degrading chitin that has previously been isolated from several saprophytic multiple polysaccharides degrading bacteria that include species of *Pseudomonas, Alteromonas, Acinetobacter*, and many more. The chitinase is produced by the bacteria and is excreted out into the culture medium, allowing the efficient use of chitin and associated products. For example, the enzyme N-acetyl glucosamine deacetylase enzyme from bacteria belonging to *Alteromonas* sp. is a potent drug and biocatalyst in the production of D glucosamine which exhibits anti-tumor and anti-microbial properties [45]. The partial de-acetylation of chitin by the bacterial enzymes gives rise to the formation of chitosan which is the only known natural cationic polysaccharide.

Both Chitin and Chitosan have interesting anti-microbial, antiviral hypocholesterolemic, antioxidant, and wound healing properties that could be utilized by the nutraceutical industry [4]. The enzyme chitinases play an important role in catalytic conversion of the insoluble chitin into its monomeric form [52]. Chitinase has a lot of potential applicability in the nutraceutical business as chitin is non-toxic, biocompatible, and biodegradable [30].

2.3. Xylanases

Is isolated from polysaccharide degrading bacteria, for example, *Thermotoga maritima*, that caused the production of a thermostable xylanase B which has been reported to be used in the clarification of fruit juices as well as exhibits a nutraceutical activity in the production of partially baked bread [31, 32, 11] Xylanases have been used as a dietary supplement as a treatment for poor digestion. In addition, xyalnases catalyses the production of xylo-oligosaccharides that have gained recent importance as a gut bacterial stimulant and also in the formulation of prebiotics [2]. The xylo-oligosaccharides have proven to be beneficial in improving gastrointestinal health and in reducing the risks of colon cancer [53]. Xylan has a low-calorie value that makes them ideal candidates for anti-obesity diets and in the food processing industry, xylan oligosaccharides have shown resistance to both heats as well as acids, thus giving them the advantage of being used in low-pH juices and drinks [2]. Among the chief physiological roles, xylan helps in the reduction of cholesterol, improved availability of calcium, inhibition of starch retrogradation, and enhancing the nutritional and sensory food value [40].

2.4. Carrageenases

This enzyme act upon the linear sulfated polysaccharides that are obtained from marine red seaweeds. The degradation by carrageenases results in the production of oligosaccharides that have exhibited antiviral, antitumor, and anticoagulation activities [49, 46]. Carrageenases degrading bacteria belong to genus *Cytophaga, Pseudomonas,* and *Alteromonas*. Carrageenans have good protein-binding properties especially to milk protein, and the stabilizing properties of the carrageenans make them valuable to the dairy and meat-processing industries. As a thickening agent, carrageenans can be used to control the viscosity of puddings and dairy products [7]. Carrageenans have demonstrated several biological activities such as the λ-carrageenans from Chondrus ocellatus have exhibited antitumor and immunomodulatory activities [58]. In addition, the role of carageenan as an antiviral agent has also been studied [34].

2.5. Fucoidanases

The enzyme degrades fucoidans that is a complex sulfated polysaccharide, used to activate a human epidermal keratinocyte activator. This enzyme was reported from marine bacteria belonging to genus *Pseudomonas* and *Pseudoalteromonas* [54]. Fucoidans that were extracted from *Ecklonia cava* exhibited an increase in anticoagulant activity. The sulfate content in the polysaccharide fucoidan had a direct effect on the antithrombin activity mediated by heparin [14]. There are several reports on the pre-treatment with fucoidan that blocked the activation of caspase-3 and caspase-9. Caspase 3 and caspase-9 have been known to facilitate the terminal stages of neuronal apoptosis [13]. The enzyme fucoidanase mediates the breakdown of fucoidan which have anticoagulant, antivirus, antitumor, antioxidant, antithrombotic, anti-inflammatory and immunomodulatory functions There are several properties of fucoidans such as regulation of glucose uptake, apoptosis of cancerous cells and antimetastatic function that make the enzymes that break the polysaccharide into simpler forms gain importance from the nutraceutical point of view.

3. Polysaccharide Degrading Bacteria

Bacteria belonging to genus *Saccharophagus* and *Microbulbifer* predominate as multiple polysaccharides degrading bacteria in the marine ecosystem. The bacteria are opportunistic pathogens as they voracious feed on decomposing seaweeds and break down the insoluble complex polysaccharides owing to their polysaccharide degrading enzymes [27] that have potential to be exploited as a nutraceutical.

Lactobacillus is known for utilizing the sugar lactose however thy also degrade starch [18] and *Bifidobacteria* amongst several polysaccharides are known to degrade arabinan, arabinogalactan, and arabinoxylan derived from plant cell wall polysaccharides [43]. As a nutraceutical, they have great potential applications as the bacteria belonging to genus *Lactobacillus* and *Bifidobacteria* have demonstrated anti-tumour and anti-mutagenic activity [21].

However, the ability of polysaccharide degrading bacteria to withstand the extreme marine environmental conditions is due to the production of unique bioactive compounds that may have applications as an API (active pharmaceutical ingredient) as well as a food supplement in various nutraceuticals. Studies conducted by Inagi and workers [28] reported bacteria communities - JS1 isolated from deep marine sediments from the Pacific Ocean margin that could degrade only monosaccharide and / or oligosaccharides.

The antibiotic, anti-inflammatory, and anti-cancer properties of bioactive compounds of the coastal bacteria growing in saline conditions were realized. However, there are no reports on polysaccharide degrading bacteria as a direct source of nutraceutical suggesting the room for further research in this area. *Pseudomonas marincola* KMM 3042 [47] was isolated from the deep Sea of Fiji. However, although this marine bacterium could produce an array of bioactive antimicrobial compounds such as pyrroles, phloroglucinol, quinolone, benzaldehyde, it won't be discussed further due to the inability of the bacteria to degrade polysaccharides rendering it as out of the scope of the chapter.

In another study, a multiple polysaccharide degrading bacterium of *Pseudoalteromonas* was discovered to produce bioactive substances that were reported to inhibit the methilicin resistant *Staphylococcus aureus* strains due to the presence of a brominated biphenyl compound and also inhibit the growth of protists [29]. Bhatnagar and Kim demonstrated the production of novel Tertiary-amine modified bentonite (TAMB) that has antifungal, anti-tumorigenic, anti-microbial, immunosuppressive, and anti-proliferative activity [5].

4. Symbiotic Interaction Between Marine Polysaccharide Degrading Bacteria

The mutual symbiotic association between marine polysaccharide degrading bacteria has been studied for the production of secondary metabolites. Also, the epiphytic/ associated bacteria with marine algae and invertebrates has also led to the production of secondary metabolites. The antibacterial activity against pathogenic bacteria such as Escherichia coli, Staphylococcus aureus, Bacillus sp., and Candida was reported [3].

Sponge associated bacterium *Nocardiopsis dassonvillei* that degrades sugars such as cellobiose, xylose, glucose, galactose, sucrose, trehalose have been shown to exhibit great chemical potential for generating novel pharmaceutical leads [55, 23]. *Bacillus amyloliquefaciens* isolated from, South China Sea demonstrated antibacterial activity against Escherichia coli and *Staphylococcus aureus*. *Bacillus amyloliquefaciens* is a polysaccharide degrading bacterium that degrades chitin [44, 22]. Zeng demonstrated the antimicrobial activity of 42 marine polysaccharide degrading bacteria belonging to the genus *Ateromonas* [56]. *Pseudomonas, Bacillus*, and *Flavobacterium.* The marine polysaccharide degrading bacteria are rich sources of new and potent bioactive compounds and enzymes that hold promising applications in the field of medicine, pharmaceutical, and nutraceutical [57].

5. Indirect Use of Polysaccharide Degrading Bacteria as Nutraceuticals

Marine polysaccharide degrading bacteria are known to feed avidly on the dead, decomposing, and detritus matter such as seaweeds. The saprophytic activity of the polysaccharide degrading bacteria helps in the breakdown of the otherwise indigestible cellulosic fibre. A study by *Jonnadula* proved the protoplasmic detritus from *Laminaria japonica* by a multiple polysaccharide degrading bacterium – *Microbulbifer* strain CMC5 which degrades xylan, carboxymethyl cellulose, agar, alginate, carageenan and chitin thus enhancing the nutraceutical potential [33].

6. Challenges and Scope

Although there is a lot of scope for marine polysaccharide degrading bacteria to be considered as an ideal candidate in formulating nutraceuticals, owing to their unique properties as previously described, there is still a long way before the successful implementation. One main difficulty in using polysaccharide degrading bacteria as a nutraceutical is the isolation and identification of non-pathogenic strains with effective and promising nutraceutical ability. The second difficulty lies in the fact that very few (less than 1%) of the marine polysaccharide degrading bacteria is culturable. This may be the reason for extremely limited research in this area.

Despite the challenges, it is noteworthy that the multiple polysaccharide degrading bacteria are ubiquitous and hardy [9] as they can withstand the extremities of marine environmental conditions due to the combination of their unique physiochemical properties along with biochemical adaptations to the dynamic marine ecosystems.

7. Conclusion

Biotechnology is an important tool to obtain resources from polysaccharide degrading bacteria, as controlling the growth conditions in a bioreactor while tailoring the production of biologically active compounds [36]. The marine heterotrophic bacteria utilize the polysaccharides due to enzymatic hydrolysis of the complex insoluble polysaccharides by glycoside hydrolases and polysaccharide lyases [37]. Among the *Gammaproteobacteria*, the genus *Pseudoalteromonas* consist of marine polysaccharide degrading bacteria that are usually producers of bio-active extracellular compounds and are also an active producer of biofilms [6]. They often secrete anti-microbial and/ or anti-viral agents and display anti-fouling properties making them excellent candidates as a nutraceutical. Also, the bacterial enzymes could be used as food ingredients as the enzymes can affect aspects of spoilage, processing, storage, and safety. Enzymes such as agarases, chitinases, fucoidanase carrageenases, and xylanases can be used as food ingredients as well as in food processing owing to due to their specificity, diversity, salt tolerance, high activity at mild pH [39].

Conflicts of Interest

The author declared no conflict of interest.

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