water

Review

A Review of Seagrass Bed Pollution

Yong Zhang [1,†], Xinping Yu [2,3,†], Zuoyi Chen [2,4], Qiuzhen Wang [3,*], Jiulong Zuo [3], Shanshan Yu [5] and Ran Guo [3,*]

[1] Marine Environment Monitoring Central Station of Qinhuangdao, SOA, Qinhuangdao 066002, China
[2] Marine Ecological Restoration and Smart Ocean Engineering Research Center of Hebei Province, Qinhuangdao 066000, China
[3] Ocean College, Hebei Agricultural University, Qinhuangdao 066000, China; zuojiulong@126.com
[4] Marine Geological Resources Survey Center of Hebei Province, Qinhuangdao 066000, China
[5] College of Marine Resource and Environment, Hebei Normal University of Science and Technology, Qinhuangdao 066004, China
* Correspondence: qqzz1990@163.com (Q.W.); guoran@hebau.edu.com (R.G.)
† These authors contributed equally to this work.

Abstract: Due to climate change and human activities, seagrass is in crisis as the coverage of seagrass declines at an accelerated rate globally. In this paper, the severe challenges of seagrass ecosystem were briefly reviewed, including adverse effects of natural factors and human activities on seagrass beds. The research status of pollutants and pollution in seagrass bed ecosystem was reviewed, the future research directions in related fields were proposed as well. The eutrophication in coastal waters and discharge of pollutants such as sulfide, heavy metals, organic matter and microplastics caused by human activities are important reasons for seagrass loss. In addition, environmental stressors lead to reduced immunity and decreased resistance of seagrass to various pathogens, leading to seagrass wasting diseases. Future studies concerning the influence of novel pollutants, i.e., plastic waste on non-native algae, microorganisms and seagrasses, as well as their interrelationships, will be of vital importance. In addition, researches on seagrass wasting diseases and their pathogens should be much accounted in China, to fill in gaps in related fields and improve the response ability to emergent seagrass diseases. In conclusion, this review was proposed to arouse the concern about the seagrass bed pollution, and provide possible enlightening information for the protection and restoration of this significant ecosystem.

Keywords: seagrass; coastal water; plastic; heavy metal; seagrass diseases; *Labyrinthula*

Citation: Zhang, Y.; Yu, X.; Chen, Z.; Wang, Q.; Zuo, J.; Yu, S.; Guo, R. A Review of Seagrass Bed Pollution. *Water* **2023**, *15*, 3754. https://doi.org/10.3390/w15213754

Academic Editor: José Luis Sánchez-Lizaso

Received: 7 October 2023
Revised: 19 October 2023
Accepted: 21 October 2023
Published: 27 October 2023

1. Introduction

Seagrass is the only species of flowering plant that lives entirely in the marine environment, mainly in marine habitats between sub-arctic and tropical latitudes. Sea grass bed is the most widely distributed coastal ecosystem on the earth, providing a variety of important ecological service functions, such as biological conservation, coastline protection, sediment stability, water purification and nutrition cycle. Seagrass meadows are breeding and feeding places for many fish and invertebrates with important economic value, and also important habitats for threatened species [1]. The seagrass meadow has high primary productivity, with aboveground biomass (mainly leaves) accounting for 50% of the total biomass. In addition, the seagrass bed ecosystem possess abundant microbial diversity and diverse microbial communities, which play an important role in the offshore nutrient cycle [2,3]. Besides, seagrass ecosystems filter and remove the bacteria pathogens, thereby reducing the exposure of humans, fish and invertebrate to pathogens [4]. The seagrass debris with highly inert organic carbon, makes an important contribution to the global blue carbon storage and sequestration [5].

Seagrass meadows store a large amount of organic carbon, which is an important component of marine blue carbon. The organic matter reservoirs in shallow marine ecosystems are composed of exogenous organic matter, microalgae and macroplants. and seagrass sources in the waters near estuaries contribute 65% of surface sediment lignin [6]. A large number of macroalgal blooms occur due to organic matter input. The macroalgae can co-metabolize with the microbial community by providing available energy and resources, and further promote the remineralization of refractory components in seagrass debris. And this may contribute to the reduction of blue carbon storage in seagrass meadows [5]. Nutrient loads and reduced light caused by macroalgal blooms are the main reasons for the seagrass decline worldwide. Though the serious degradation trend worldwide was observed because of ocean warming and eutrophication, seagrass blue carbon is still able to effectively eliminate the CO_2 emissions and is considered as an effective natural solution to mitigate global climate change [7]. Thus, reducing coastal eutrophication could increase the conservation of seagrass meadows and further mitigate global climate deterioration. In addition, coastal environmental pollutants, such as heavy metals, refractory organic matter and microplastics, can not only pose a serious threat to the life activities of primary marine animals and plants, but also accumulate in marine food products, thus damaging human health.

Seagrass can produce natural biological fungicides such as phenols, to improve water quality by inhibiting the pathogenic bacteria, thus benefitting for humans and marine invertebrates. The phenol concentrations in seagrass were affected by seawater turbidity, temperature, ocean acidification, low salinity and heavy metal pollution. However, the opportunistic pathogen *Labyrinthula* could inhibit phenols production [8], and lead to seagrass wasting disease. Therefore, the effects of physical, chemical and biological factors should be adequately considered in environmental pollution monitoring and risk assessment of seagrass bed ecosystems to assess the ecological health of seagrass more comprehensively.

2. Challenges Confronted by Seagrass Bed

Due to the global climate change and disturbance of human activities, seagrass meadows are decreasing rapidly [9]. Reasons leading to the seagrass degradation could be attributed to natural and human factors. Natural factors include typhoons, earthquakes, volcanic eruptions, etc. In recent years, the frequency and intensity of hurricanes, oceanic heat waves and other extreme climates have gradually increased, causing serious damage to the global seagrass meadow [10–12]. When hurricanes and typhoons occurred, the whole seagrass tissue was buried in the sediment or uprooted, causing serious damage to the seagrass meadows. In addition, increased seawater turbidity, decreased light intensity and decreased salinity indirectly led to further degradation of seagrass meadows. In 2019, the super-strong typhoon “Liqima” reduced the distribution area of Japanese *Zostera japonica* by more than half in the Yellow River Delta of China, with the loss of soil organic carbon exceeding 35% and soil total nitrogen exceeding 65%. Even if the environmental factors were suitable for the growth, Japanese eel grass species was still unable to recover, owing to the lack of seagrass seeds and overwintering twigs, as well as the small residual distribution area [12]. Human activities, such as coastal development, reclamation, trawling and agricultural runoff affect the coastal marine ecological environment widely and are key threats to the loss of seagrass [13–15]. Severe meadow loss could reduce the availability of marine habitats, damage the service functions of seagrass ecosystems, and lead to the gradual loss of blue carbon storage, which is re-emitted to the atmosphere and thus increase the carbon concentration [16]. Considering severe damage and degradation risk of the seagrass meadows, further effective protection and remediation should be executed from the natural and social perspectives by exploring the critical pressure systematically.

3. Pollution Status of Seagrass Beds

3.1. Eutrophication

Coastal nutrient input is one of the key factors contributing to the seagrass degradation worldwide. Nutrient enrichment resulted in the excessive reproduction of epiphytic algae and macroalgae in the seagrass bed, along with light occlusion by the high density of algae organisms, which caused decreased photosynthetic rate of seagrass and insufficient photosynthetic oxygen production. Besides, the oxidation process of high-concentration sulfide is further limited by oxygen deficiency, which inhibits the growth and metabolism, or even leads to death of seagrass [5]. Seagrass debris, as well as large amounts of debris produced by the death of saprophytic and macroalgae, would float on the seawater surface or be deposited in surface sediments and transported by wind and water to coastlines. Algal debris is usually composed of unstable organic carbon (LOC) and is more easily ingested and utilized by microorganisms. Eutrophication could lead to the increase of sediment organic carbon from algae in seagrass beds, increase the composition of active organic carbon in sediments, promote the growth and metabolic activity of microorganisms, and accelerate the utilization and transformation of sediment organic carbon [17]. Therefore, coastal eutrophication could alter the uptake, conversion, and storage function of coastal blue carbon in the seagrass bed ecosystem.

3.2. Sulphides

The coastal sediments inhabited by seagrass are characterized by low concentration of oxygen and high concentration of toxic and reducing substances (such as iron, manganese and sulfide) [18]. Sulfide even at low concentrations (1 to 10 μmol/L) is toxic to the cells of eukaryotes such as seagrass, while seagrass could still survive in such high sulfide ranges [19]. Seagrass can avoid root oxygen hypoxia and sulfide invasion through root oxygen leakage (radial oxygen loss). In other words, oxygen produced by photosynthesis spreads through the ventilated tissue to breathe and leaks through the root tip to maintain oxygen in the rhizosphere [20–22]. *Zostera marina* has formed two major sulfide detoxification strategies with the help of seagrass microbes. Sulfide is oxidized and precipitated to elemental sulfur in the aerating tissue, or to thiols and sulfate in the plant. Then elemental sulfur and thiols are stored in the rhizomes and roots, while sulfate is transported from the tissue underground to that overground. Besides, the underground tissue possess the highest detoxification capacity (86%), especially the rhizome (61%), as the main buffer for detoxifying sediment sulfide, could protect the fragile meristem in the leaves [19]. However, due to climate change and human activity, future increases in surface water temperature, hypoxia and sediment sulfide levels would further elevate the sulfide pressure in seagrass bed ecosystem, possibly even exceeding the sulfide tolerance and detoxification [19,23,24]. It is known that microorganisms respond quickly to changes in their living environment. By detecting the abundance of sulfur cycling genes, such as sulfate reduction genes, in the sediment microbial communities of seagrass. Thus, the sulfur content could be indirectly detected by molecular biological methods, which may be used as a potential monitoring indicator in coastal habitats with sulfur stress in the future [25].

3.3. Heavy Metals

Seagrass meadows have been seriously polluted by a series of pollutants including heavy metals and pesticides, and are becoming an important sink of anthropogenic pollutants in coastal areas [26,27]. Simultaneously, as a significant habitat and food source for a variety of marine animals such as green turtles, seagrass meadows could transfer heave metals accumulated to consumers with higher nutritional levels as well [28]. Trace metal pollution in estuarine seagrass meadows has been observed worldwide, with most studies conducted in the Caribbean [29], Italy [28], India [30], Fiji [31], Australia [32] and Republic of Korea [33]. Marine angiosperms generally have a high bioaccumulation capacity for trace metals because of the interactions between waters and sediments in marine environment directly through leaves and root-rhizomes, where ion uptake occurs [34]. Seagrass is confronted with greater anthropogenic pressure than other marine communities. Changes in seagrass coverage, decreased growth rate and slow development signify environment variation [29,34]. Therefore, trace element level in seagrass beds can be a useful indicator of harmful pollutants in seagrass ecosystems.

Trace metals, i.e., copper, iron, manganese and zinc are necessary for plants by promoting their growth and metabolism within a certain concentration range. However, many artificial activities can increase their accumulation in the natural environment. Once the concentration of trace elements reaches the threshold level, it would damage the root cell structure and photosynthesis [35]. Seagrass is the largest reservoir of heavy metals, the contents of which could be increased through fallen leaves and food chain transmission. However, in the vegetation-free sediment, heavy metal contents were relatively constant [28]. In addition, exploring the cumulative characteristics of trace elements in the whole and each parts (such as root, rhizome, leaves), as well as between the surrounding seawater and sediment is of great significance for understanding the cycle mechanism of trace elements in the seagrass ecosystem.

3.4. Refractory Organic Compounds

Coastal environmental pollution, especially persistent organic pollution, led to severe decline of seagrass meadows, among which polycyclic aromatic hydrocarbons (PAHs) attract much attention due to their persistence, toxicity, mutagenicity and carcinogenicity. The lipophilic character of organic pollutants makes it easy to penetrate the cytoplasmic membrane to accumulate in marine organisms. Human activities such as combustion of fossil fuels cause terrestrial import of PAH, leading to increased PAH content in coastal sediments and seagrass. PAHs are transmitted along the food chain and ultimately damage human health [36]. The abundant microbial communities distributed in seagrass bed ecosystems play a key role in organic substance degradation, certain consortia of which could degrade hydrocarbon through nitrogen fixation [37]. The effect of PAHs addition on bacterial communities in the sediment of *Enhalus acoroides* seagrass showed that different strains and bacterial group behaved differently in response to PAH exposure. It is worth noting that microbial community structure of seagrass sediment was sensitive to PAH-induced stress and susceptible to PAHs contamination, which can be used as a potential indicator of PAHs contamination [38]. The fungi in sediments of seagrass *Enhalus acoroides*, such as the phylum Ascomycota and Basidiomyces, had the potential to degrade PAHs. It was found that low concentration (100 mg/kg) PAHs contamination can increase the fungal diversity in a short time. While the fungal diversity reduced with high concentration (1000 mg/kg) PAHs contamination in a short time, which however could be utilized in a long term (7~28 d) by the fungi [39]. Thus, microbial community in seagrass sediment could be used as an early monitoring indicator to identify PHAs contamination in seagrass bed ecosystems.

To date, the effects of PAHs addition on seagrass functional microorganisms such as nitrogen-fixing bacteria and sulfate-reducing bacteria have been less studied. Limited studies have shown that the addition of PAH can inhibit both nitrogen-fixing bacteria and sulfate-reducing bacteria communities [40]. The seagrass *Posidonia oceanica* could accumulate PAHs in tissue. However, the nitrogen-fixing bacterial combination underwent a huge transition under the stress induced by PAHs addition [36,41]. Some seagrass nitrogen-fixed bacteria are known to degrade PAHs with low concentrations, while sulfur-reducing bacteria exhibit a resistance to high concentrations of PAHs [40]. Moreover, microorganisms that degrade hydrocarbons by nitrogen fixation can be applied to oil spill cleaning, especially in areas with limited nitrogen content [37].

3.5. Microplastics

Marine microplastics is an important pollutant in the global coastal and marine environment. Plastics floating on the surface or submerged in water have serious negative effects on marine life and marine ecosystems. Plastic adhered to contaminated organic matter and sediment particles would be sink to the sediment surface colonized by seagrass and macroalgae plants. These macrophyte would accumulate in large amounts and create a new micro-habitat furtherly by changing the nonbiological conditions (such as environmental pH, temperature and oxidation-reduction), microbial community composition and biogeochemical cycle process [42–45]. This poses a threat to the survival of some marine organisms, and disrupts balance of the seagrass bed ecosystem through the cascade and amplification effect.

Of the 70 seagrass species that exist worldwide, only seven species of seagrass were detected for the microplastics, which include *Z. noltei*, *Z. marina*, *E. acoroides*, *C. nodosa*, *C. serrulate*, *C. rotundata* and *Thalassia hemprichii* [46]. The microbial biofilm on the surface of seagrass leaves can promote the adsorption and trapping of microplastics, and thus increase the microplastics content in the leaves. In addition, the microplastics content was related to the seagrass species and characteristics of the leaves and differs in the leaves and sediments of the same seagrass species among different coastal areas [46]. This can be attributed to the differentiation of environmental conditions and microplastics types, as well as the measurement methods which is the key factor in assessing the microplastic distribution. Therefore, the standardization of microplastics determination methods is of great significance for revealing the distribution of microplastics in seagrass bed ecosystems around the world and their impact on coastal ecosystems.

Marine organisms living in seagrass bed ecosystems that feed on seagrass leaves can accumulate microplastics in the body through feeding, and transmit through the food chain to higher organisms. This will eventually damage human health seriously by accumulating high content of microplastics in human body. Existing studies have detected the adverse effects of microplastics on marine organisms in the physiological, metabolic and genetic levels [47]. Thus it can be predicted that the biotoxicity and environmental risks would amplify with the increase of microplastics abundance. Furthermore, coastal microplastics could be input mainly by terrestrial and marine routes. The former mainly input microplastics including microplastic debris and chemical fiber products as well as cosmetic wastes rich in plastic particles into the offshore environment through rivers, sewage discharge and other ways. While the marine input mainly exists in fishing activities such as aquaculture and marine transport. Therefore, it is of great significance to figure out and block the input routes of microplastics in the specific coastal areas as well as develop new degradable plastic products for the protection of coastal seagrass ecosystems and human health.

Microplastic types detected in the seagrass bed ecosystem include polypropylene (PP), polyethylene (PE), polyethylene terephthalate (PET) and polyamide (PA), polystyrene (PS), and polyvinyl chloride (PVC), with the first four occurring most frequently [46]. In addition, both high-density polyethylene (HDPE) and biodegradable starch-based plastics can be present in marine sediments of temperate regions for at least 18 months [48]. Both plastics could change the structure of seagrass *Cymodocea nodosa* (such as the biomass and length of rhizomes), and reduce plant coverage in the meadow. Besides, HDPE had a greater effect than biodegradable starch-based plastics [48]. The HDPE deposition may not only reduce the seagrass habitat by promoting the spread of non-native macroalgae in the meadows, but also affect the interaction intensity between the two algae and promote the spread of macroalgae in the sea meadow [48]. In addition, degradable starch-based bio-plastic bags and compostable plastic bags can transform the interaction between species from neutral to competitive by forming a physical barrier and changing the sediment quality [49]. Therefore, it is critical to evaluate the effects of plastic wastes and their deposition on non-native algae and seagrass as well as their relationship in the future [48].

3.6. Pathogenic Bacteria

Due to human impacts and global climate change, emerging infectious marine diseases are becoming more widespread and serious. At present, four known eukaryotic genera that can cause seagrass diseases are *Labyrinthula*, *Phytophthora*, *Halophytophthora* and *Physoyxea* [50]. Among them, the genus *Labyrinthula* is a heterotrophic and halophytic protist causing seagrass wasting diseases (SWD). It is also the most studied seagrass pathogen to date (Table 1). *Labyrinthula* strains isolated from seagrass leaves showed varying degrees of virulence using laboratory infection tests and related phylogenetic analysis. Isolates with high virulence were able to invade leaf cells of living plants and cause black leaf lesions, a diagnostic feature of SWD [49–51]. However, *Labyrinthula* is also a ubiquitous symbiont that decomposes marine plants and algal wastes, one of which hosts the lawn grass [51]. Since the 1930s, the North American and European Atlantic coastal *Zostera* has been severely affected by consumptive disease, when it killed 90% of the North Atlantic *Zostera* population [52]. Muehlstein et al. [53] (1991) first identified *L. zosterae* (Labyrinthulomycetes) as the pathogen causing wasting diseases of *Zostera*, and later found that in many countries such as Australia, Mexico, and Republic of Korea (Table 1). However, the research on seagrass consumptive disease and its pathogenic bacteria in China is still blank.

Table 1. Studies on *Labyrinthula* causing meadow wasting diseases.

Host	*Labyrinthula*	Sampling Site	Sampling Time	Main Content	References
Halophila australis	*Labyrinthula* SR_Ha_C	Victoria, Australia	March 2016 (early autumn, Australia)	The genomic sketch and predicted protein group of the pathogenic isolate *Labyrinth* SR_HA_C were proposed. Phylogeny and cross-phylum comparisons reveal the evolutionary history of stramenopiles	[54]
Zostera marina	*Labyrinthula zosterae*	Republic of Korea	April–September 2013	The first report on *L. zosterae*, a causative agent of consumptive disease of seagrass *Z. marina*, in Republic of Korea	[55]
Thalassia testudinum	*Labyrinthula* sp.	Florida	May 2015	A sensitive qPCR method was established with universal applicability to the seagrass pathogen *Labyrinthula* worldwide. A host immunization panel was developed that could evaluate factors that may affect host, *Labyrinthula* and environment.	[56]
Eelgrass	*Labyrinthula zosterae*	Southern bog in Charleston, Oregon, USA	2017 and 2019	Abnormally large amounts of DHA was observed in marine parasite *L. zosterae* which may be an unknown source of long-chain polyunsaturated fatty acids in the eel grass ecosystem	[57]
Amphibolis antarctica, *Halophila australis*, *Heterozostera nigricaulis*, *Posidonia australis*, *Zostera muelleri*	*Labyrinthula*	Southeast Australia	March 2016 (early autumn, Australia)	The *Labyrinthula* isolate was first cultured, genotyped and pathogenicity identified in Australia, and thus provide a preliminary ecological understanding of consumptive diseases in Australia	[58]
Zostera muelleri, *Halophila ovalis*, *Heterozostera nigricaulis*, *Posidonia australis*	*Labyrinthula* sp.	Southeast Australia	March 2014–October 2015	Isolation and characterization of *Labyrinthula* in southeastern Australia for the first time in Australia	[59]
Turfgrass	*Labyrinthula* spp.	New Mexico and Arizona	2011 and 2012	Genetic diversity, pathogenicity and morphological differences of *Labyrinthula* were determined for rapid blight in lawn grass from new Mexico and Arizona	[60]

Table 1. *Cont.*

Host	*Labyrinthula*	Sampling Site	Sampling Time	Main Content	References
Thalassia testudinum	*Labyrinthula* sp.	Florida	June 2010	Phenols and potential novel, unspecified non-phenolic metabolites from seagrass *Thalassia testudinum* Banks ex Konig were demonstrated to have anti-*Labyrinthula* activity by in vitro bioassay	[61]
Zostera marina	*Labyrinthula zosterae*	Northern Europe (Portugal, Germany, Denmark, southern Norway and western Sweden)	2010–2012	The quantitative PCR (QPCR) technology was used to quantitatively analyze the abundance and prevalence of the pathogen causing the wasting disease in 19 phyllostachys species in the coastal area of northern Europe using the species-specific primers designed for the internal transcribed spacer (ITS1) of *L. zosterae*	[62]
Zostera marina	*Labyrinthula zosterae*	Northern Europe	August–October 2010	The 18S rDNA (1400 bp) of *L. zosterae* isolates (N = 41) from 6 sites in northern Europe and 1 site in the south (Adriatic) were identified to assess the identity and potential diversity of their endogenous protists	[63]
Turfgrass	*Labyrinthula terrestris*	United States of America	2003–2007	The most commonly used *Labyrinthula* growth medium serum seawater agar (SSA) was modified, and the modified SSA (MSSA) and grass extract SSA (GESSA) media were designed	[64]
Poa annua	*Labyrinthula terrestris*	Adams county, Colorado, USA	April 2009	The first report of rapid wasting disease by *L. terrestris* on *Poa annua*, Colorado, as well as the pathological features of plants and the pathogenic characteristics	[65]
Poa trivialis L., *Lolium perenne* L.	*Labyrinthula terrestris*	Arizona, USA	February 2003	The isolation, morphological characteristics and growth curve of the new *Labyrinthula* species which were named *L. terrestris*	[66]

The genus *Labyrinthula*, belonging to the phylum Labyrinthulomycetes, is genetically related to thraustochytrids, a protist famous for their DHA production potential. And both of them widely distribute in marine environment and play significant role in marine ecosystems [67,68]. Unlike *Labyrinthula*, thraustochytrids is not pathogenic to plants such as seagrass, but it can be pathogenic to some marine mollusks [69]. In our previous study, several *Labyrinthula* strains were isolated from the coastal mangrove habitats in southern China. The colonies of *Labyrinthula* sp. was white and round, and uniform spindle-shaped cells were observed under the microscope. Simultaneously, we found that *Labyrinthula* sp. could synthesize polyunsaturated fatty acids (PUFAs) such as docosahexaenoic acid (DHA), with potential for industrial application [70]. However, the pathogenicity of these *Labyrinthula* strains was not investigated in our previous work. Therefore, it would be interesting to clarify the relationship between the lipid metabolism and their pathogenicity from the perspective of molecular biology and omics in the future.

4. Conclusions and Future Prospects

In summary, the challenges confronted by the seagrass bed ecosystem are becoming increasingly serious under the impact of global climate change and human activities. The impacts of human activities on seagrass ecosystems are particularly prominent. Coastal nutrient enrichment alters the organic carbon storage of sediments by changing the activities of algae and microorganisms. Though sulfide, heavy metals and refractory organic substances have toxic effects on seagrass organisms, seagrass and the epiphytic microorganisms have certain tolerance and degradation ability to resist them in a certain concentration range. However, the terrestrial input from microplastics will carry large amounts of pollutants and sediment particles, greatly changing the biological and non-biological conditions of the seagrass ecosystem, and the resulting cascade effect needs to be further studied.

The seagrass beds are comprehensively affected by various factors, and their complex coupling effects could not be ignored. In the future, comprehensive analysis of various factors using mathematical models will be of great significance for revealing the environmental pressure faced by the seagrass beds more accurately and formulating the scientific protection and restoration strategies for seagrass beds.

In addition, many marine eukaryotes can cause seagrass diseases. At present, *Labyrinthula*, the most widely studied pathogen causing seagrass consumptive diseases worldwide, has not attracted much attention in some regions such as China, and relevant research processes need to be accelerated in the future. Current urgent studies on the seagrass pathogen *Labyrinthula* include: (1) isolation of *Labyrinthula* strains from decayed and diseased seagrass by pure culture techniques, analyzing their pathogenicity and ecological functions from the perspective of genome and transcriptomics, to reveal the potential of *Labyrinthula* causing seagrass wasting diseases in some unstudied coastal waters; (2) elucidating the diversity and distribution characteristics of *Labyrinthula* using molecular biology methods to explore the pathogenic potential of pathogenic *Labyrinthula* on different species of seagrass and their ecological function as well as adaptive evolution mechanisms in various habitats of seagrass beds. These could provide basic information for the protection and restoration for seagrass bed ecosystem.

Author Contributions: Q.W., Conceptualization, Project Administration, Supervision, Writing Original Draft, Review and Editing; R.G., Project Administration, Supervision, Review and Editing, Funding acquisition; Y.Z., Conceptualization, Writing—Original Draft Preparation, Writing Original Draft, Review and Editing; X.Y., Writing Original Draft, Review and Editing, Software; Z.C., Writing Original Draft, Review and Editing; J.Z., Review and Editing, Software. S.Y., Software. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the National Natural Science Foundation of China (32100003), the Open Fund of Marine Ecological Restoration and Smart Ocean Engineering Research Center of Hebei Province (HBMESO2304) and Marine Science and Technology Project of North Sea Bureau of Ministry of Natural Resources of China (202308).

Data Availability Statement: Data will be provided on request.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Unsworth, R.K.F.; Cullen-Unsworth, L.C.; Jones, B.L.H.; Lilley, R.J. The planetary role of seagrass conservation. *Science* **2022**, *377*, 609–613. [CrossRef] [PubMed]
2. Bagwell, C.E.; La Rocque, J.R.; Smith, G.W.; Polson, S.W.; Friez, M.J.; Longshore, J.W.; Lovell, C.R. Molecular diversity of diazotrophs in oligotrophic tropical seagrass bed communities. *FEMS Microbiol. Ecol.* **2002**, *39*, 113–119. [CrossRef] [PubMed]
3. Hamisi, M.I.; Lyimo, T.J.; Muruke, M.H.; Bergman, B. Nitrogen fixation by epiphytic and epibenthic diazotrophs associated with seagrass meadows along Tanzanian coast, Western Indian Ocean. *Aquat. Microb. Ecol.* **2009**, *57*, 33–42. [CrossRef]
4. Ugarelli, K.; Laas, P.; Stingl, U. The microbial communities of leaves and roots associated with turtle grass (*Thalassia testudinum*) and manatee grass (*Syringodium filliforme*) are distinct from seawater and sediment communities, but are similar between species and sampling sites. *Microorganism* **2019**, *7*, 4–23. [CrossRef]
5. Liu, S.; Trevathan-Tackett, S.M.; Lewis, C.J.E.; Huang, X.; Macreadie, P.I. Macroalgal blooms trigger the breakdown of seagrass blue carbon. *Environ. Sci. Technol.* **2020**, *54*, 14750–14760. [CrossRef]
6. Nakakuni, M.; Watanabe, K.; Kaminaka, K.; Mizuno, Y.; Takehara, K.; Kuwae, T.; Yamamoto, S. Seagrass contributes substantially to the sedimentary lignin pool in an estuarine seagrass meadow. *Sci. Total Environ.* **2021**, *793*, 148488. [CrossRef]
7. Stankovic, M.; Ambo-Rappe, R.; Carly, F.; Dangan-Galon, F.; Fortes, M.D.; Hossain, M.S.; Kiswara, W.; Van Luong, C.; Minh-Thu, P.; Mishra, A.K.; et al. Quantification of blue carbon in seagrass ecosystems of Southeast Asia and their potential for climate change mitigation. *Sci. Total Environ.* **2021**, *783*, 146858. [CrossRef]
8. Groner, M.; Burge, C.; Kim, C.; Rees, E.; Van Alstyne, K.; Yang, S.; Wyllie-Echeverria, S.; Harvell, C. Plant characteristics associated with widespread variation in eelgrass wasting disease. *Dis. Aquat. Org.* **2016**, *118*, 159–168. [CrossRef]
9. Lopez, B.; Hinesp, P.J.; Ash, C. The unrecognized value of grass. *Science* **2022**, *377*, 590–591. [CrossRef]
10. Qin, L.-Z.; Kim, S.H.; Song, H.-J.; Kim, H.G.; Suonan, Z.; Kwon, O.; Kim, Y.K.; Park, S.R.; Park, J.-I.; Lee, K.-S. Long-term variability in the flowering phenology and intensity of the temperate seagrass *Zostera marina* in response to regional sea warming. *Ecol. Indic.* **2020**, *119*, 106821–106831. [CrossRef]
11. Tomasko, D.; Alderson, M.; Burnes, R.; Hecker, J.; Iadevaia, N.; Leverone, J.; Raulerson, G.; Sherwood, E. The effects of Hurricane Irma on seagrass meadows in previously eutrophic estuaries in Southwest Florida (USA). *Mar. Pollut. Bull.* **2020**, *156*, 111247. [CrossRef] [PubMed]
12. Yue, S.; Zhang, X.; Xu, S.; Liu, M.; Qiao, Y.; Zhang, Y.; Liang, J.; Wang, A.; Zhou, Y. The super typhoon Lekima (2019) resulted in massive losses in large seagrass (*Zostera japonica*) meadows, soil organic carbon and nitrogen pools in the intertidal Yellow River Delta, China. *Sci. Total Environ.* **2021**, *793*, 148398. [CrossRef] [PubMed]
13. Unsworth, R.K.; Ambo-Rappe, R.; Jones, B.L.; La Nafie, Y.A.; Irawan, A.; Hernawan, U.E.; Moore, A.M.; Cullen-Unsworth, L.C. Indonesia's global significant seagrass meadows are under widespread threat. *Sci. Total Environ.* **2018**, *634*, 279–286. [CrossRef] [PubMed]
14. Hu, W.; Zhang, D.; Chen, B.; Liu, X.; Ye, X.; Jiang, Q.; Zheng, X.; Du, J.; Chen, S. Mapping the seagrass conservation and restoration priorities: Coupling habitat suitability and anthropogenic pressures. *Ecol. Indic.* **2021**, *129*, 107960. [CrossRef]
15. Stankovic, M.; Hayashizaki, K.-I.; Tuntiprapas, P.; Rattanachot, E.; Prathep, A. Two decades of seagrass area change: Organic carbon sources and stock. *Mar. Pollut. Bull.* **2021**, *163*, 111913. [CrossRef]
16. Macreadie, P.I.; York, P.H.; Sherman, C.D.; Keough, M.J.; Ross, D.J.; Ricart, A.M.; Smith, T.M. No detectable impact of small-scale disturbances on 'blue carbon' within seagrass beds. *Mar. Biol.* **2014**, *161*, 2939–2944. [CrossRef]
17. Liu, S.L.; Jiang, Z.J.; Wu, Y.C.; Zhang, J.P.; Zhao, C.Y.; Huang, X.P. Carbon storage mechanism of marine litter and its response to eutrophication. *Sci. Bull.* **2017**, *62*, 3309–3318.
18. Terrados, J.; Duarte, C.M.; Kamp-Nielsen, L.; Agawin, N.S.R.; Gacia, E.; Lacap, D.; Fortes, M.D.; Borum, J.; Lubanski, M.; Greve, T. Are seagrass growth and survival constrained by the reducing conditions of the sediment? *Aquat. Bot.* **1999**, *65*, 175–197. [CrossRef]
19. Hasler-Sheetal, H.; Holmer, M. Sulfide intrusion and detoxification in the seagrass Zostera marina. *PLoS ONE* **2015**, *10*, e0129136. [CrossRef]
20. Pedersen, O.; Binzer, T.; Borum, J. Sulphide intrusion in eelgrass (*Zostera marina* L.). *Plant Cell Environ.* **2004**, *27*, 595–602. [CrossRef]
21. Jensen, S.I.; Kuhl, M.; Prieme, A. Different bacterial communities associated with the roots and bulk sediment of the seagrass *Zostera marina*. *FEMS Microbiol. Ecol.* **2007**, *62*, 108–117. [CrossRef] [PubMed]
22. Frenderiksen, M.S.; Glud, R.N. Oxygen dynamics in the rhizosphere of *Zostera marina*: A two-dimensional planar optode study. *Limnol. Oceanogr.* **2006**, *51*, 1072–1083. [CrossRef]
23. Orth, R.J.; Carruthers, T.J.B.; Dennison, W.C.; Duarte, C.M.; Fourqurean, J.W.; Heck, K.L.; Hughes, A.R.; Kendrick, G.A.; Kenworthy, W.J.; Olyarnik, S.; et al. A global crisis for seagrass ecosystems. *Bioscience* **2006**, *56*, 987–996. [CrossRef]
24. García, R.; Holmer, M.; Duarte, C.M.; Marbà, N. Global warming enhances sulphide stress in a key seagrass species (NW Mediterranean). *Glob. Chang. Biol.* **2013**, *19*, 3629–3639. [CrossRef] [PubMed]

25. Fraser, M.W.; Martin, B.C.; Wong, H.L.; Burns, B.P.; Kendrick, G.A. Sulfide intrusion in a habitat forming seagrass can be predicted from relative abundance of sulfur cycling genes in sediments. *Sci. Total Environ.* **2023**, *864*, 161144. [CrossRef] [PubMed]
26. Bonanno, G.; Orlando-Bonaca, M. Trace elements in Mediterranean seagrasses and macroalgae, A review. *Sci. Total Environ.* **2018**, *618*, 1152–1159. [CrossRef]
27. Bonanno, G.; Venezaiano, V.; Orlando-Bonaca, M. Comparative assessment of trace element accumulation and biomonitoring in seaweed Ulva lactuca and seagrass Posidonia oceanica. *Sci. Total Environ.* **2020**, *718*, e137413. [CrossRef]
28. Amado, F.G.M.; Creed, J.C.; Andrade, L.R.; Pfeiffer, W.C. Metal accumulation by *Halodule wrightii* populations. *Aquat. Bot.* **2004**, *80*, 241–251.
29. Govers, L.L.; Lamers, L.P.; Bouma, T.J.; Eygensteyn, J.; de Brouwer, J.H.; Hendriks, A.J.; Huijbers, C.M.; van Katwijk, M.M. Seagrasses as indicators for coastal trace metal pollution: A global meta-analysis serving as a benchmark, and a Caribbean case study. *Environ. Pollut.* **2014**, *195*, 210–217. [CrossRef]
30. Immaculate, J.K.; Lilly, T.T.; Patterson, J. Macro and micronutrients of seagrass species from Gulf of Mannar, India. *MOJ Food Process. Technol.* **2018**, *6*, 391–398.
31. Singh, S.; Lal, M.M.; Southgate, P.C.; Wairiu, M.; Singh, A. Trace metal content in sediment cores and seagrass biomass from a tropical southwest Pacific Island. *Mar. Pollut. Bull.* **2021**, *171*, 112745. [CrossRef] [PubMed]
32. Prange, J.A.; Dennision, W.C. Physiological responses of five seagrass species to trace metals. *Mar. Pollut. Bull.* **2000**, *40*, 327–336. [CrossRef]
33. Jeong, H.; Choi, J.Y.; Choi, D.-H.; Noh, J.-H.; Ra, K. Heavy metal pollution assessment in coastal sediments and bioaccumulation on seagrass (*Enhalus acoroides*) of Palau. *Mar. Pollut. Bull.* **2021**, *163*, 11912. [CrossRef] [PubMed]
34. Bonanno, G.; Di Martino, V. Trace element compartmentation in the seagrass Posidonia oceanica and biomonitoring applications. *Mar. Pollut. Bull.* **2017**, *116*, 196–203. [CrossRef]
35. Ambo-Rappe, R.; Lajus, D.L.; Schreider, M.J. Heavy metal impact on growth and leaf asymmetry of seagrass Halophila ovalis. *J. Environ. Chem. Ecotoxicol.* **2011**, *3*, 149–159.
36. Apostolopoulou, M.-V.; Monteyne, E.; Krikonis, K.; Pavlopoulos, K.; Roose, P.; Dehairs, F. Monitoring polycyclic aromatic hydrocarbons in the Northeast Aegean Sea using *Posidonia oceanica* seagrass and synthetic passive samplers. *Mar. Pollut. Bull.* **2014**, *87*, 338–344. [CrossRef]
37. Foght, J. *Handbook of Hydrocarbon and Lipid Microbiology: Nitrogen Fixation and Hydrocarbon-Oxidizing Bacteria*; Springer: Berlin/Heidelberg, Germany, 2010; pp. 1661–1668.
38. Ling, J.; Jiang, Y.-F.; Wang, Y.-S.; Dong, J.-D.; Zhang, Y.-Y.; Zhang, Y.-Z. Responses of bacterial communities in seagrass sediments to polycyclic aromatic hydrocarbon-induced stress. *Ecotoxicology* **2015**, *24*, 1517–1528. [CrossRef]
39. Ling, J.; Zhang, Y.; Wu, M.; Wang, Y.; Dong, J.; Jiang, Y.; Yang, Q.; Zeng, S. Fungal community successions in rhizosphere sediment of seagrasses *Enhalus acoroides* under PAHs stress. *Int. J. Mol. Sci.* **2015**, *16*, 14039–14055. [CrossRef]
40. Ling, J.; Zhou, W.; Yang, Q.; Lin, X.; Zhang, Y.; Ahmad, M.; Peng, Q.; Dong, J. Effect of PAHs on nitrogen-fixing and sulfate-reducing microbial communities in seagrass *Enhalus acoroides* sediment. *Arch. Microbiol.* **2021**, *203*, 3443–3456. [CrossRef]
41. Sun, F.-L.; Wang, Y.-S.; Sun, C.-C.; Peng, Y.-L.; Deng, C. Effects of three different PAHs on nitrogen-fixing bacterial diversity in mangrove sediment. *Ecotoxicology* **2012**, *21*, 1651–1660. [CrossRef]
42. Green, D.S.; Boots, B.; Blockley, D.J.; Rocha, C.; Thompson, R. Impacts of discarded plastic bags on marine assemblages and ecosystem functioning. *Environ. Sci. Technol.* **2015**, *49*, 5380–5389. [CrossRef] [PubMed]
43. Balestri, E.; Menicagli, V.; Vallerini, F.; Lardicci, C. Biodegradable plastic bags on the seafloor: A future threat for seagrass meadows? *Sci. Total Environ.* **2017**, *605–606*, 755–763. [CrossRef] [PubMed]
44. Cozzolino, L.; Nicastro, K.R.; Zardi, G.I.; Santos, C.B.d.L. Species-specific plastic accumulation in the sediment and canopy of coastal vegetated habitats. *Sci. Total Environ.* **2020**, *723*, 138018. [CrossRef] [PubMed]
45. Seeley, M.E.; Song, B.; Passie, R.; Hale, R.C. Microplastics affect sedimentary microbial communities and nitrogen cycling. *Nat. Commun.* **2020**, *11*, 2372. [CrossRef] [PubMed]
46. Li, C.; Zhu, L.; Li, W.-T.; Li, D. Microplastics in the seagrass ecosystems: A critical review. *Sci. Total Environ.* **2023**, *902*, 166152. [CrossRef] [PubMed]
47. Guzzetti, E.; Sureda, A.; Tejada, S.; Faggio, C. Microplastic in marine organism: Environmental and toxicological effects. *Environ. Toxicol. Pharmacol.* **2018**, *64*, 164–171. [CrossRef]
48. Menicagli, V.; Balestri, E.; Vallerini, F.; De Battisti, D.; Lardicci, C. Plastics and sedimentation foster the spread of a non-native macroalga in seagrass meadows. *Sci. Total Environ.* **2020**, *757*, 143812. [CrossRef]
49. Brakel, J.; Reusch, T.B.; Bockelmann, A.C. Moderate virulence caused by the protist *Labyrinthula zosterae* in ecosystem foundation species Zostera marina under nutrient limitation. *Mar. Ecol. Prog. Ser.* **2017**, *571*, 97–108. [CrossRef]
50. Sullivan, B.K.; Trevathan-Tackett, S.M.; Neuhauser, S.; Govers, L.L. Host-pathogen dynamics of seagrass diseases under future global change. *Mar. Pollut. Bull.* **2018**, *134*, 75–88. [CrossRef]
51. Martin, D.L.; Chiari, Y.; Boone, E.; Sherman, T.D.; Ross, C.; Wyllie-Echeverria, S.; Gaydos, J.K.; Boettcher, A.A. Functional, phylogenetic and host-geographic signatures of *Labyrinthula* spp. provide for putative species delimitation and a global-scale view of seagrass wasting disease. *Estuaries Coasts* **2016**, *39*, 1403–1421. [CrossRef]
52. Muehlstein, L.K. Perspectives on the wasting disease of eelgrass *Zostera marina*. *Dis. Aquat. Org.* **1989**, *7*, 211–221. [CrossRef]

53. Muehlstein, L.K.; Porter, D.; Short, F.T. *Labyrinthula Zosterae* sp. Nov., the causative agent of wasting disease of eelgrass, *Zostera Marina*. *Mycol.* **1991**, *83*, 180–191. [CrossRef]
54. Tan, M.H.; Loke, S.; Croft, L.J.; Gleason, F.H.; Lange, L.; Pilgaard, B.; Trevathan-Tackett, S.M. First genome of *Labyrinthula* sp., an opportunistic seagrass pathogen, reveals novel insight into marine protist phylogeny, Ecology and CAZyme Cell-Wall Degradation. *Genes Genomes* **2021**, *82*, 498–511. [CrossRef] [PubMed]
55. Lee, S.J.; Shim, J.B.; Lee, S.R. First report of *Labyrinthula zosterae* (Labyrinthulomycetes) as the causal pathogen of wasting disease in the seagrass Zostera marina in Korea. *Plant Dis.* **2021**, *105*, 8. [CrossRef]
56. Duffin, P.; Martin, D.L.; Lohan, K.M.P.; Ross, C. Integrating host immune status, *Labyrinthula* spp. load and environmental stress in a seagrass pathosystem: Assessing immune markers and scope of a new qPCR primer set. *PLoS ONE* **2020**, *15*, e0230108. [CrossRef]
57. Yoshioka, R.M.; Schram, J.B.; Galloway, A.W.E. Eelgrass pathogen *Labyrinthula zosterae* synthesizes essential fatty acids. *Dis. Aquat. Org.* **2019**, *135*, 89–95. [CrossRef]
58. Trevathan-Tackett, S.M.; Sullivan, B.K.; Robinson, K.; Lilje, O.; Macreadie, P.I.; Gleason, F.H. Pathogenic *Labyrinthula* associated with Australian seagrasses: Considerations for seagrass wasting disease in the southern hemisphere. *Microbiol. Res.* **2018**, *206*, 74–81. [CrossRef]
59. Sullivan, B.K.; Robinson, K.L.; Trevathan-Tackett, S.M.; Lilje, E.S.; Gleason, F.H.; Lilje, O. The first isolation and characterisation of the protist *Labyrinthula* sp. in Southeastern Australia. *J. Eukaryot. Microbiol.* **2017**, *64*, 504–513. [CrossRef]
60. Chitrampalam, P.; Goldberg, N.; Olsen, M.W. *Labyrinthula* species associated with turfgrasses in Arizona and New Mexico. *Eur. J. Plant Pathol.* **2015**, *143*, 485–493. [CrossRef]
61. Trevathan-Tackett, S.M.; Lane, A.L.; Bishop, N.; Ross, C. Metabolites derived from the tropical seagrass *Thalassia testudinum* are bioactive against pathogenic *Labyrinthula* sp. *Aquat. Bot.* **2015**, *122*, 1–8. [CrossRef]
62. Bockelmann, A.C.; Tams, V.; Ploog, J.; Schubert, P.R.; Reusch, T.B.H. Quantitative PCR reveals strong spatial and temporal variation of the wasting disease pathogen, *Labyrinthula zosteraein* in Northern European eelgrass (*Zostera marina*) beds. *PLoS ONE* **2013**, *8*, e62169. [CrossRef] [PubMed]
63. Bockelmann, A.C.; Beining, K.; Reusch, T.B.H. Widespread occurrence of endophytic *Labyrinthula* spp. in northern European eelgrass *Zostera marina* beds. *Mar. Ecol. Prog. Ser.* **2012**, *445*, 109–116. [CrossRef]
64. Yadagiri, K.K.; Kerrigan, J.; Martin, S.B. Improved methods for axenic culture of *Labyrinthula terrestris*, causal agent of rapid blight of turfgrasses. *Can. J. Microbiol.* **2012**, *58*, 1230–1235. [CrossRef] [PubMed]
65. Hyder, N.; Wong, F.P.; Koski, A.; Tisserat, N.; Stowell, L. First report of rapid blight caused by Labyrinthula terrestris on Poa annua in Colorado. *Plant Dis.* **2010**, *94*, 919. [CrossRef] [PubMed]
66. Bigelow, D.M.; Olsen, M.W.; Gilbertson, R.L. *Labyrinthula terrestris* sp. nov., a new pathogen of turf grass. *Mycologia* **2005**, *97*, 185–190. [CrossRef] [PubMed]
67. Bai, M.; Sen, B.; Wang, Q.; Xie, Y.; He, Y.; Wang, G. Molecular detection and spatiotemporal characterization of Labyrinthulomycete protist diversity in the coastal waters along the Pearl River Delta. *Microb. Ecol.* **2018**, *77*, 394–405. [CrossRef]
68. Popova, O.V.; Belevich, T.A.; Golyshev, S.A.; Kireev, I.I.; Aleoshin, V.V. *Labyrinthula diatomea* n. sp.—A Labyrinthulid associated with marine diatoms. *J. Eukaryot. Microbiol.* **2020**, *67*, 393–402. [CrossRef] [PubMed]
69. Hassett, B.T. A widely distributed thraustochytrid parasite of diatoms isolated from the Arctic represents a gen. and sp. nov. *J. Eukaryot. Microbiol.* **2020**, *67*, 480–490. [CrossRef]
70. Wang, Q.; Ye, H.; Xie, Y.; He, Y.; Sen, B.; Wang, G. Culturable diversity and lipid production profile of Labyrinthulomycete protists isolated from coastal mangrove habitats of China. *Mar. Drugs* **2019**, *17*, 268–285. [CrossRef]