Vol. 24, No. 3(2025) Bio25514
Revista Mexicana de Ingeniería Química

Biotic and abiotic interactions during the decomposition of *Sargassum* spp. from the Mexican Caribbean

Interacciones bióticas y abióticas durante la degradación de *Sargassum* spp. del caribe mexicano

J.A. Domínguez-Maldonado [1], R. Tapia-Tussell [2]*, E. Olguin-Maciel [2], K.J. Azcorra-May [2], E. Sauri-Duch[3], G. Rivera-Muñoz[3], G. Lizama-Uc[3], S. E. Solís-Pereira[3]*
[1]Dirección de Gestión Tecnológica, Centro de Investigación Científica de Yucatán A. C. Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
[2]Unidad de energía Renovable del Centro de Investigación Científica de Yucatán A. C. Carretera Sierra Papacal Chuburná Puerto Km 5, c.p. 97302 Mérida, Yucatán, México.
[3] Departamento de Química y de ingeniería bioquímica, Tecnológico Nacional de México /IT de Mérida, Av., Tecnológico Km 4.5 S/N 97118 Mérida Yucatán, México.

Sent date: January 27, 2025; Accepted: August 5, 2025

Abstract

A study was conducted to examine decomposition process of *Sargassum* spp., using 1.25-liter conical reactors. This study used a multifaceted approach to analyze the roles of biotic and abiotic factors in the degradation process. The aim of this study was to identify the physicochemical changes in *Sargassum* spp. To this end, the structural composition of the biomass was analyzed during a period of 150 day. Additionally, the production of leachate and biogas during sargassum natural degradation process was measured, and the structural changes in microbial communities were evaluated. This study explored the relationship between biogas production and the microorganisms in sargassum, as well as their potential biotechnological applications. The results of this study indicated the presence of *Thermofilum* and *Methanopyrus* microorganisms during the initial degradation stage (0-30 days). These microorganisms are marine archaea that primarily generate greenhouse gases through their metabolic byproducts, including hydrogen sulfide and methane. This observation is particularly salient during the initial stages of degradation when gas production is particularly prolific. A comprehensive understanding of these foundational principles can facilitate the development of effective management strategies for the management of *Sargassum* spp., which could subsequently lead to the creation of a biotechnological tool to produce high value-added compounds, such as biogas, and fertilizers.

Keywords: *Sargassum* spp, sargassum decomposition, biotic interactions, abiotic interactions, Caribbean sargassum.

Resumen

Se realizó un estudio para examinar el proceso de descomposición de *Sargassum* spp. utilizando reactores cónicos de 1,25 litros. Este estudio utilizó un enfoque multifacético para analizar el papel de los factores bióticos y abióticos en el proceso de degradación. El objetivo fue identificar los cambios fisicoquímicos en el *Sargassum* spp. Para ello, se analizó la composición estructural de la biomasa durante un periodo de 150 días. Además, se midió la producción de lixiviado y biogás durante el proceso de degradación natural del sargassum y se evaluaron los cambios espaciotemporales de las comunidades microbianas. Este estudio exploró la relación entre la producción de biogás y los microorganismos del sargassum, así como sus potenciales aplicaciones biotecnológicas. Los resultados indicaron la presencia de microorganismos *Thermofilum* y *Methanopyrus* durante la etapa inicial de degradación (0-30 días). Estos microorganismos son Arqueas marinas que generan principalmente gases de efecto invernadero a través de sus subproductos metabólicos, incluidos el sulfuro de hidrógeno y el metano. Esto es especialmente relevante durante las fases iniciales de la degradación, cuando la producción de gases es prolífica. Una estudio de este proceso podría facilitar el desarrollo de estrategias de gestión eficaces para el *Sargassum* spp., lo que posteriormente podría conducir a la creación de herramientas biotecnológicas para la obtención de productos de alto valor añadido, como biogás y fertilizantes.

Palabras clave: *Sargassum* spp, Degradación de sargazo, interacciones bióticas, Interacciones abióticas, Sargazo del caribe.

---

*Corresponding author. E-mail: sara.sp@merida.tecnm.mx, rtapia@cicy.mx;
`https://doi.org/10.24275/rmiq/Bio25514`
ISSN:1665-2738, *issn-e*: 2395-8472

1 Introduction

During the last decade, excessive amounts of pelagic *Sargassum* spp. have arrived in the Caribbean, causing severe environmental damage and economic consequences for the tourism industries of several Caribbean countries (Chávez *et al.*, 2020; Vázquez-Delfín *et al.*, 2021). However, the long-term impacts of this phenomenon have not been fully measured or evaluated, as they could potentially lead to future changes in the Caribbean coastal zone, including alterations in pH, dissolved oxygen levels, and turbidity (Álvarez-Rocha, 2022). It is imperative to consider the possibility of an increase in nitrogen levels over time in the groundwater of the Yucatan Peninsula, as this could have significant environmental and economic consequences. This increase can be attributed to leachate production during biomass degradation (García-Sánchez et al., 2020; Leal-bautista *et al.*, 2024; John J. Milledge & Harvey, 2016b). These alterations could persist since sargassum quantities appear to increase annually. In 2019, (Wang *et al.*, 2019) estimated 20 million tons of macroalgae in the Atlantic and referred to it as the "Great Sargassum Belt". Several research have demonstrated that these massive amounts of seaweed contain toxic metals. These metals are accumulated during biomass trajectory and life cycle. This accumulation is due its hyperaccumulating nature (Leal-bautista et al., 2024; Olguin *et al.*, 2022; Rodríguez-Martínez *et al.*, 2020b).

A key feature of *Sargassum* spp. is its structural composition, which is mostly constituted by polymers such as fucoidan. These polymers are an integral part of the structure of organism and contain sulfate groups that can constitute a significant proportion of its weight, ranging from 5% to 38% depending on the season (Lim & Wan Aida, 2017). The sulfur content in sargassum is relevant because it transforms into hydrogen sulfide during degradation. Devault *et al.*, (2021) and John James Milledge *et al.*, (2020) have reported that this chemical compound induces toxicological effects when people are exposed to sargassum at concentrations between 0 and 10 ppm (Resiere *et al.*, 2020). Recent reports have documented the production of hydrogen sulfide due to sargassum degradation on beaches, with concentrations reaching up to 5 ppm (Rodríguez-Martínez *et al.*, 2024). However, the magnitude of the problem and the role of hydrogen sulfide production in confinement sites are not established.

A confinement site is a location designated for the disposal of sargassum; it often lacks security, accurate management and organization. These sites receive the substantial volumes of sargassum removed from beaches, over extended periods from months to years. Within these sites, the algae decomposes due to temperature and humidity levels, resulting in the generation of large volumes of gases and leachates (Olguin *et al.*, 2022).

The structure-function relationship of the carbohydrates varies by species (Takagi *et al.*, 2018). Their degradation depends on alginate dissolution, although other reactive compounds also impact their biological degradation (Robledo & Vázquez-Delfín, 2019). Recent reports indicate that these macroalgae contain significant amounts of lignin (Alzate-Gaviria *et al.*, 2020; Ardalan *et al.*, 2018; Thompson *et al.*, 2019). The conversion of lignin fractions into biofuels and other high value-added products remains a challenge for researchers due to the heterogeneity and complex structure of the polymer (Ponnusamy et al., 2019).

During the sargassum degradation process, the production of leachates and organic particles causes the phenomenon of brown tides. These events have been observed to cause a decrease in light penetration, oxygen levels, and pH. This, also causes an increase in temperature, sulfide levels, and organic matter and nutrient loads (López-Contreras *et al.*, 2021). The leachates produced during this process create conditions analogous to eutrophication and may contain heavy metals (Antonio-martínez *et al.*, 2020). Discharging sargassum leachates into the sea or in inadequate land deposits can contribute to toxic metal contamination of groundwater (Rodríguez-Martínez *et al.*, 2020). The Yucatan Peninsula and the Mexican Caribbean are particularly vulnerable due to the prevalence of karst soil. This has prompted early warnings regarding leachates, that can percolate into the soil and reach the groundwater systems in the area(Estrada Medina, 2019). Additionally, the decomposition of pelagic sargassum has been found to affect coral species, leading to their partial or complete death (Antonio-Martínez *et al.*, 2020). Bacteria are considered a vital component in the degradation process of sargassum due to their environmental adaptability and biochemical versatility. The diversity of microorganisms found on and within macroalgae is referred as the "microbiome" and includes bacteria, archaea, eukaryotic protists, fungi, and viruses(Egan *et al.*, 2017). A recent study analyzed the bacterial community structure of *Sargassum horneri* drifting in the Yellow Sea (Mei *et al.*, 2019). Consistent with the findings of other studies on macroalgae, Mei *et al.* (2019), demonstrated genus-level variations in bacterial composition between surrounding water and drifting algae, as well as in nearshore seaweeds. This indicates that community composition might differ based on geographical location. In a similar study, Torralba *et al.* (2017) found that sargassum complexes of two dominant surface-drifting species, *S. natans* and *S. fluitans*, in the Gulf of Mexico were associated with microbial communities dominated by the families Rhodobacteraceae and Saprospiraceae (Torralba *et al.*, 2017). Additionally, Serebryakova *et* *al*. (2018) studied the bacterial diversity of pelagic sargassum by analyzing 16S rRNA gene sequences associated with the benthic species *Sargassum muticum*. They found that the microbial community was primarily composed of Proteobacteria, Bacteroidetes, and Actinobacteria (Serebryakova *et al*., 2018). Determining the structure of microbial communities is difficult due to its dependence on the type of macroalgae and its seasonality. The microbiome associated with macroalgae is influenced by various biotic factors, such as algal growth and the production of metabolites by the macroalgae, as well as the composition of the surrounding seawater microbial community. Abiotic factors including temperature, and, pH, salinity, and oxygen availability affect microbial community, wave activity, and nutrient levels in seawater also play a role (Egan *et al*., 2017). Furthermore, bacteria have been observed to associate specifically with certain species of macroalgae and even with specific parts of the alga. Although the mechanisms underlying this specificity have not been fully elucidated, some associated ecological functions have been demonstrated. Specifically, chemical reaction mechanisms can be clearly attributed to algae or their epibionts. In many cases, these mechanisms can trigger the biosynthesis of bioactive compounds (Goecke *et al*., 2010). Other changes in the community may be associated with season, location, environmental stress, and macroalgae-specific regions (Egan *et al*. 2017). Given the scarcity of research on degradation in macroalgae in this region of the Caribbean, the objective of this work was to study some of the biotic and abiotic processes involved in the decomposition of *Sargassum* spp. To this end, the physicochemical characteristics of *Sargassum* spp., were assessed before and after 150 days of degradation. The volume of leachates and biogas produced during the natural degradation process were also measured. Additionally, the structural changes in microbial communities present in *Sargassum* spp. leachates over 150 days of natural degradation were determined and their relationship to the structural changes undergone by *Sargassum* was established. This information is relevant for developing effective management strategies for *Sargassum* spp., which could lead to the establishment of a biotechnological approach to produce high value-added products.

2 Materials and methods

2.1 Sargassum spp. collection site

Sargassum samples were collected in September 2020 along the coast of Benito Juárez (21°4'04.1 "N,86°46'33 "W) in Quintana Roo, Mexico, using a fishing net. The fresh sargassum was preserved with its leachates in plastic bags on ice. Upon arrival in the laboratory, the pelagic sargassum was separated from seagrass vegetation, such as *Thalassia testudinum* and *Syringodium filiforme*, as well as from epibionts. A portion of the collected sargassum was used for initial characterization, while another portion was employed for the environmental degradation experiment.

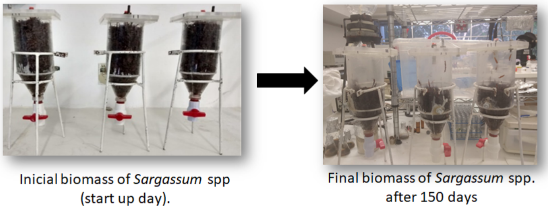

Fig. 1. Reactors used for the natural degradation of *Sargassum* spp.

2.2 Natural degradation of sargassum

The study of the natural degradation of *Sargassum* spp. was carried out as follows: Initially, 300 g of fresh *Sargassum* spp. biomass was weighed using a Torrey digital balance. Subsequently, the material was placed in three 1,250-mL conical reactors with the natural humidity of the macroalgae, i.e., in triplicate (see Figure 1). The reactors were maintained at a room temperature of 28 ± 2 °C for a duration of 150 days. To prevent evaporation while enabling gas exchange, the reactors were sealed.

2.3 Characterization of the raw material

2.3.1 Determination of lignin in Sargassum spp

The NREL/TP-510-42618 protocol was employed to quantify the lignin content (Sluiter, *et al*., 2008).

2.3.2 Elemental analysis of Sargassum spp

Elemental analysis (C, H, N, and S) was performed using a Thermo-Scientific Flash 2000 (Waltham, MA, USA) on a dried, ground, and sieved sargassum sample with 80 mesh. Approximately three to four milligrams of the sample were placed in tin containers (ThermoScientific) and 10 milligrams of vanadium pentoxide (V_2O_5) (ThermoScientific) were used as the catalyst for the sulfur peak. Each sample was analyzed in triplicate.

2.3.3 Leachates

The leachate produced by the degradation of sargassum biomass were collected at 0, 30, 60, 90, and 150 days and quantified.

Serological bottles of 250 mL with 10 g of *Sargassum* spp.

Pressure measurement with IFM sensor

Measurement with Biogas 5000

Fig. 2 Determination of greenhouse gases.

The natural moisture of the biomass was maintained as it was collected in the field, without adding more water. This approach was adopted to replicate decomposition processes occurring in beaches, confinement sites or landfills. The reactors were sealed to prevent evaporation of water from the sargassum biomass and the entry of dust. The leachates were analyzed for pH, temperature, conductivity, and salinity.

2.3.4 Determination of greenhouse gases

A biogas analyzer (Fonotest Biogas 5000) was used to measure the gases. For this purpose, ten grams of fresh sargassum were placed in 250 milliliters serological bottles (see Figure 2). To maintain anaerobic conditions, the bottles were capped with sealed septa, and the pressure was monitored using an IFM digital pressure sensor (model PN2596). This sensor was coupled to a needle capable of penetrating the septa to measure the pressure (see Figure 2). Quantification of the gases was carried out every two days; during this period, the compounds H_2 S, CH_4 and CO_2 were identified as greenhouse gasses produced by the sargassum.

2.4 DNA and metagenomic analysis

The microbial consortium was isolated as follows: First, 1 mL of sargassum leachate was inoculated into a Petri dish containing an SBM (Screening Bacteria Medium 523, Sigma-Aldrich®) culture medium. Then, the inoculum was distributed evenly across the surface of the media using an inoculation loop. The dishes were incubated in dark at 35 °C for 48 hours. This process was repeated for each leachate collection after 30, 60, 90, and 150 days.

DNA was isolated from each sample using a ZymoBIOMICSTM DNA miniprep Kit (Zymo Research, Irvine, CA, USA). The genomic DNA samples were then subjected to sequencing on the Illumina MiSeq platform (Macrogen). The library preparation was carried out using Herculase II Fusion DNA polymerase Nextera XT Index Kit V2 and the sequencing was performed by single end (SE x 300bp) with four biological samples, each with duplicates. The primers used to amplify the 16S rRNA genes from bacteria and archaea were 341F and 805R.

The raw reads were analyzed using the Galaxy platform (https://usegalaxy.org), where a quality control analysis was performed with FASTQC. The sequences were entered into Trim Galore to remove adapters (PHRED: 20, MINLEN: 50 bp). Finally, Trimmomatic (Bolger et al., 2014), was used to remove low quality sequences (SLIDINGWINDOW:4:20, MINLEN: 50 bp).

Taxonomic assignment was performed on the Galaxy platform using the Kraken2 program (Wood & Salzberg, 2014) with the single-end option. A comparison was then conducted using the Greengenes2 2022.10 database with the parameters configured by the program: k-mer-len: 35, minimizer-len: 31, minimizer-spaces: 6, load-factor: 0.7.

3 Results and discussion

3.1 Sargassum spp. characterization

The characterization of *Sargassum* spp. was conducted at the beginning and end of the 150-day experiment, which was focused on the natural degradation of the macroalgae. These parameters are essential for evaluating the environmental impact of this macroalga. Analyzing microbial metabolism during the degradation process provided critical insights that facilitated the development of management strategies. This analysis determined the degradation time in the environment and identified current deposition sites.

A comprehensive characterization of sargassum was conducted; as shown in Table 1, in which, relative humidity, total ash content, and lignin content were presented, the values can vary among different *Sargassum* species, location and temporality (Ardalan *et al.*, 2018; Bikker *et al.*, 2016; Borines *et al.*, 2013; Rabemanolontsoa & Saka, 2013). The analysis showed that pelagic sargassum has unique characteristics, it has a low relative humidity content when ground and dried., contrasting with the findings reported by other authors, such as Biswas *et al*. 2017, who documented higher values (13.5%) for brown algae *Sargassum terrarium* (Biswas *et al*., 2017). However, the authors do not specify the time when the biomass was collected, where it was collected from (offshore or on the beach), or the method used to grind and dry the sample.

These factors are important when comparing moisture and ash results because the season and the locality where the seaweed was collected can influence sargassum characterization results. Several studies have noted that the ash, moisture, and mineral content can vary depending on the season in which the seaweed was collected (Antúnez-Argüelles *et al*., 2020; John James Milledge *et al*., 2020; Oliveira *et al*., 2015; Saldarriaga-Hernandez *et al*., 2021). The particle size is another factor that influences this process, as it has been observed that screening with a fine mesh can affect water absorption in the sample. For instance, Ali and Bahadar subjected the *Sargassum* spp., to a drying process involving dry air for a period of four hours. Thereafter, they proceeded to grind and screen the material, reducing its size to bellow 0.4 mm, obtaining a moisture content of 9.8 ± 1.1 wt% for this macroalgae (Ali & Bahadar, 2017).

The results of this study also indicate a high lignin content (Table 1), suggesting the adaptability of the macroalgae as they undergo changes from the Atlantic Ocean to the Mexican Caribbean. A histological staining method was used by Alzate *et al*, (2020), to characterized complex polyphenolic compounds in *Sargassum* spp. This work established a foundational framework for enhancing the comprehension of the architecture and arrangement of heteropolymers within the cell wall of these macroalgae. It also explored the potential factors that may contribute to this phenomenon, focusing on the abiotic stresses, such as UV radiation and high temperatures, which are adverse conditions generated by climate change (Alzate-Gaviria *et al*., 2020).

One of the most descriptive analyses of sargassum biomass is the organic elemental analysis (C, N, H and S), since it provides insight into the composition of the biomass. Through elemental analysis of dry sargassum, the following concentrations were obtained: carbon concentration of 32.47%, a hydrogen concentration of 4.8%, a sulfur concentration of 0.91%, and a nitrogen concentration of 1.005% (see Table 2).

The carbon content of the samples was observed to range from 30.63% to 33.72%, this is consistent with the findings reported in other studies of brown algae. Biswas *et al.* (2017) found that the carbon content of the macroalgae *Sargassum tenerrimum* ranged from 30 to 32%, respectively. However, the nitrogen content is noteworthy for its lower values compared to those reported by Milledge *et al.* (2020), in which nitrogen levels were found up to 1.71% for sargassum in the Caribbean. Regarding sulfur content, the values were higher compared with other authors.

The nitrogen-carbon ratio was around 30, indicating a nitrogen deficiency for the composting context, for which the recommended is 10 to 20. However to initiate microbial growth, the ratio must be between 20 and 30 carbon atoms per nitrogen atom (Méndez *et al*., 2011).

Table 1. *Sargassum* spp. characterization.

Material	% Residual moisture	% Ash	% lignin	Reference
Sargassum tenerrimum	13.5	23.2	ND	(Biswas *et al*., 2017)
Sargassum sp	11.16 ±0.01	26.19 ± 0.07	less than 1	(Borines *et al*., 2013)
Ulva lactuca	ND	17.30	6.90	(Bikker *et al*., 2016)
Sargassum sp	ND	17.10	6.0	(Rabemanolontsoa & Saka, 2013)
Sargassum angustifolium	ND	35.5-43.8	11.8-25.8	(Ardalan *et al*., 2018)
Sargassum sp	ND	ND	4.60	(Oliveira *et al*., 2015)
Macroalgae consortium	ND	23.1	24.1	(Antúnez-Argüelles *et al*., 2020)
***Sargassum* spp.**	**5.06 ±0.5**	**24.48 ±1.75**	**30.14**	**This study**

Table 2. Elemental analysis of *Sargassum* spp.

Day 0	N (%)	C (%)	H (%)	S (%)
Reactor 1	1.21	30.62	4.87	1
Reactor 2	0.55	33.72	5.02	0.78
Reactor 3	1.24	33.06	4.71	0.95
Mean	1	32.5	4.87	0.91
Standard deviation	0.39	1.63	0.155	0.115
Day 150	N (%)	C (%)	H (%)	S (%)
Reactor 1	2.53	23.27	2.44	1.63
Reactor 2	2.98	23.39	2.58	1.77
Reactor 3	2.76	23.33	2.51	1.7
Mean	2.76	23.3	2.51	1.7
Standard deviation	0.225	0.06	0.07	0.07

Table 3. Comparative analysis of the elemental composition of different sargassum studies.

Sample	C (%)	H (%)	N (%)	S (%)	C/N	Reference
Sargassum spp	**33.159**	**4.784**	**1.173**	**0.962**	**28.269**	**This study**
S. fluitans, y natans	27.41	3.13	1.71	0.21	16.029	Milledge (2020)
Sargassum spp.	25.35	3.81	1.37	N.D	18.504	Ali and Bahadar (2017)
Sargassum spp.	27.5	4.16	1.21	0.82	22.727	Thompson et al. (2020)
Sargassum spp	27.68	2.72	1.64	ND	16.878	Nielsen (2021)

N. D.; Not detected.

As shown in Table 2, the increase in nitrogen content may be due the activity of the cyanobacteria found in the macroalgae. Numerous studies have shown that marine epiphytic bacteria are a significant source of fixed nitrogen for algae (Goecke *et al.*, 2010). Several epiphytic cyanobacteria (*Calothrix* sp., *Anabaena/Dolichospermun* sp. and *Phormidium* sp.) that fix nitrogen and supply it to *Codium* species (Chlorophyta) have been identified in certain areas. Another nitrogen-fixing species, *Dichothrix fucicola*, has been observed in association with *Sargassum natans* and *S. fluitans* in the Sargasso Sea and Gulf Stream (Goecke *et al.* 2010). This phenomenon may have arisen during the degradation of *Sargassum* spp.

As illustrated in Table 3, this study presents a comparative analysis of the elemental compositions of various sargassum studies. This analysis reveals that the consortium in question has the highest Sulphur content, compared with findings from other researchers.

The C/N ratio is a crucial factor in determining the suitability of macroalgae for biogas production. It is important to note that this ratio can vary significantly based on nutrient viability. For example, studies have shown that the average C/N ratio in the deep open ocean is 47, whereas in shallower neritic waters is 27. However, no significant differences were observed between the C/N ratios of *S. fluitans* and *S. natans* (Milledge *et al.*, 2020). Although elemental determinations of macroalgae are not a common practice, recent studies have revealed signs of nitrogen deficiency in macroalgae, with the C/N ratio displaying seasonal fluctuations (Nielsen *et al.*, 2021). A study on *Sargassum muticum* revealed that summer samples had a C/N ratio closer to optimal levels reported in the literature, while spring samples exhibited deficiency. This finding is particularly relevant in the context of anaerobic digestion, where the rapid consumption of carbon leads to the accumulation of volatile fatty acids (VFA) (Thompson *et al.*, 2020). Excess of nitrogen in the substrate promotes ammonia formation, which is toxic for various groups of bacteria at high concentrations, thereby reducing the potential of substrate for methane production.

3.2 Leachates

The leachates generated after 150 days from the *Sargassum* spp. inside the conical bottom reactor were 0.302 ± 0.02 L/kg. Approximately, 50% of the initial biomass was lost, likely due to the loss of water and organic compounds from the cell walls, including alginates, fucoidans, proteins, and lipids. These substances are degraded by the enzymatic system of microorganisms (Alderkamp *et al.*, 2007; Morrissey et al., 2021). The highest volume of leachate (25 mL) was collected on day 30, while the lowest volume (7 mL) was collected at the end of the 150 days. This phenomenon can be attributed to the natural process of *Sargassum* spp. losing water and organic matter over time, as discussed by Antúnez-Argüelles *et al* 2020 and Alzate-Gaviria *et al*, 2020. The recalcitrance of compounds such as lignin results in their greater resistance to degradation.

Research on leachate generation during algal decomposition, particularly in the Mexican Caribbean, is limited. Currently, there is a lack of data on the volume of leachate produced by this macroalgae during decomposition. Milledge and Harvey (2016) used ensiling as a conservation technique for *Sargassum muticum* and observed leachate generation of 7.8% w/w after 60 days(Milledge & Harvey, 2016a). Subsequently, Antonio-Martínez *et al.* (2020) examined the impact of leachate on coral larvae, reporting a leachate generation time of 3.5 days in 0.4 x 0.4 x 1 m tanks, yielding a leachate volume of 6.6 L/kg of biomass. However, these studies were not conducted over an extended period, nor did they examine leachate production of other species of macroalgae. Therefore, this study is the first to investigate the mechanisms of sargassum degradation by microbial communities and their influence on leachate production. Understanding the structure of microbial communities during sargassum degradation can provide valuable information for establishing confinement sites and determining the subsequent treatment of this macroalgae. Previous studies have documented that the algae removed from beaches are disposed of in areas that are not adequately prepared to prevent leachates from reaching the aquifer (Rodríguez-Martínez *et al.*, 2020).

3.3 *Summary of the initial and final characterization of environmental degradation of Sargassum spp*

After 150 days of *Sargassum* spp. degradation, the characterization parameters were evaluated again, as shown in Table 4. The table provides an overview of the parameters measured during the degradation process. It reveals that the initial mass loss was 146 g, corresponding to 48.46% of the original weight. By the end of the 150-days period, a total of 90.83 milliliters of leachates had been produced, representing 30.28% of the initial weight. This calculation assumes that the density of the leachates is equivalent to that of water (1 g/cm^3). This data indicates a significant decrease in moisture content within the macroalgae tissue, which was 30.28% when considering the loss of water exclusively attributable to the production of leachates over the 150-day period. Other studies have indicated that leachate generation varies depending on the collection site (Leal et al., 2024). The *Sargassum* spp. consortium was reported to generate 58 milliliters of leachate, which was designated as leachate per se. This consortium was collected at sea, similar to the present study. However, this study focused on the leachate generated by the decomposition of the macroalgae. A notable finding is the loss of carbon, as evidenced by a 9.2% reduction in total carbon content compared to the initial level. This was followed by a decline in hydrogen content, from 4.87% to 2.51%. These changes may be attributed to the loss of organic structures, as indicated by a decrease in lignin from 30.13% to 12.14%. This decline suggests a loss of approximately half of the initial biomass. Regarding the nitrogen and sulfur content, there was an increase from 1% to 2.76% for nitrogen and from 0.9% to 1.7% for sulfur, respectively.

3.4 *Microbial diversity in leachates*

More than 250 genera were identified with OTUs during the degradation of *Sargassum* spp. at room temperature. However, as shown in Figure 3, the composition of these genera changed over time; the composition of the leachates also changed significantly over time. Initially, *Thermophilum* dominated, accounting for 84% of the relative abundance, it was followed by *Methanopyrus* (9%), *Hydrogenophilus* (3%), and *Thermoproteus* and *Thermoplasma* (1% each). After 30 days, a major shift occurred, with *Fischerella* and *Hydrogenophilus* becoming the dominant genera, accounting for 60% and 26%, respectively. Conversely, the least abundant genera were *Mycobacterium*, *Thermophilum*, and *Thermoproteus*, with only 1% of the relative abundance.

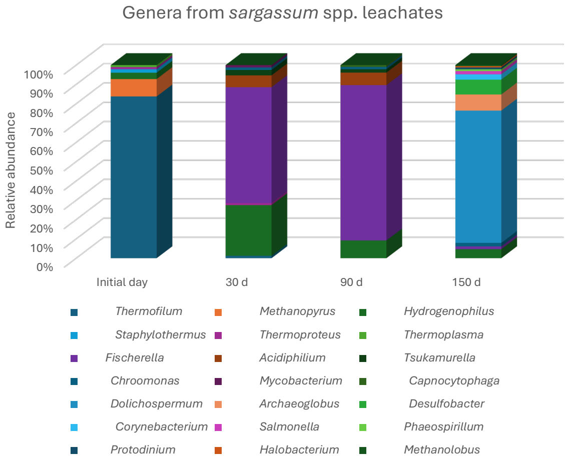

Fig. 3 Microbial communities present in *Sargassum* spp.

Table 4. Summary of the initial and final characterization of environmental degradation of *Sargassum* spp from Caribbean.

Parameter	Initial (Day 0)	Final (Day 150)	Difference
Mass (g)	300	154.6	146
Leacheates (mL)	0	90.83	-90.83
C (%)	32.47	23.27	9.2
H (%)	4.87	2.51	2.36
S (%)	0.91	1.7	-0.79
N (%)	1	2.76	-1.76
Moisture (%)	86.15	55.87	30.28
Ash (%)	24.48	26.34	-1.86
Lignin (%)	30.137	17.99	12.147
Conductivity	100mS	100mS	0
pH	7.6	8.2	0.6

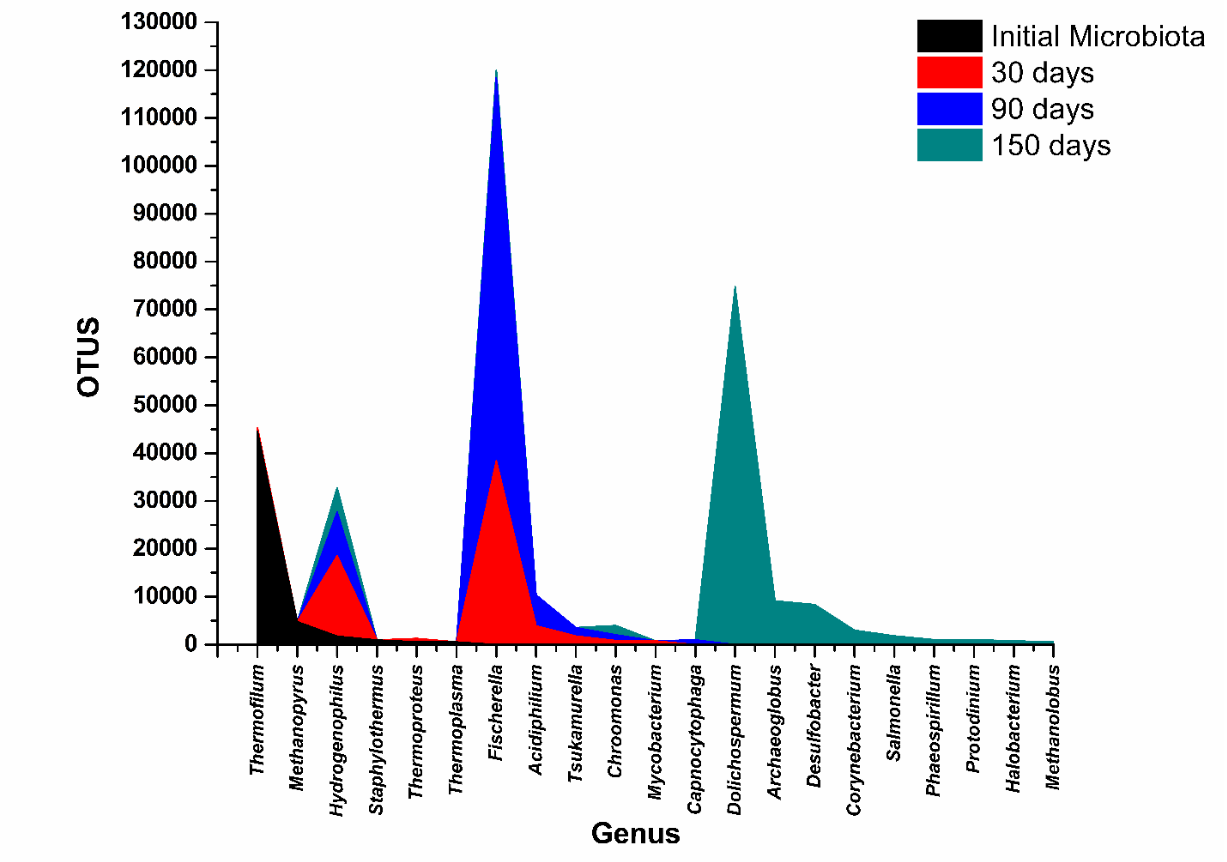

Fig. 4 Relative abundance over time of the main genera presents in *Sargassum* spp leachates.

Figure 4 illustrates that the genera *Methanopyrus* and *Thermofilum*, which are hyperthermophiles, belong to the phyla *Euryarchaeota* and *Crenarchaeota*, respectively, and were the most abundant at the beginning. These microorganisms are also the most abundant in oceans. Thus, it is hypothesized that they were found in the fresh leachates of *Sargassum* spp. which were collected from the sea at the beginning of the experiment. Methanopyrus metabolism has been linked to methane production from algal biomass, as documented for other *Euryarchaeota* microorganisms (Hervé *et al.*, 2021). These genera play a pivotal role in degradating *Sargassum* spp. and the producing leachate, since they utilize macroalgal polysaccharides (Devault *et al.*, 2021). *Methanopyrus* has been observed to capture carbon dioxide and hydrogen in its metabolic processes and produce methane. Additionally, *Thermofilum*, a sulfur-dependent anaerobic heterotroph (Fenner, 2008), has been implicated in the degradation of sulfur-containing polymers, such as fucoidan. These organisms produce methane and hydrogen sulfide when degrading *Sargassum* spp., either on beaches or at storage sites. Consequently, these organisms are present at the onset of leachate collection.

As shown in Figure 4, the most prevalent genera (*Thermofilum, Fischerella,* and *Dolichospermum*) are represented by black, blue, and green, respectively. A less abundant red peak is followed by *Hydrogenophilus, Methanopyrus,* and *Desulphobacter* as the second most prevalent genera, corresponding to a time between zero and 30 days. Subsequent analysis of the abundance of these genera over time revealed *Thermofilum* and *Methanopyrus* as the most prevalent in the initial phase. It is noteworthy that these genera are known to metabolize sulfur and hydrogen, suggesting a potential role in the degradation of sulfur-containing sugars, such as fucoidan, which are characteristic of brown algae

(Egan *et al.*, 2017). This observation aligns with the onset of sargassum degradation, suggesting a possible link between the abundance of these genera and the initiation of this ecological process.

The genera *Hydrogenophilus* and *Fischerella* were the most prevalent after 90 days, while *Dolichospermum* and *Desulfobacter* dominated until the end of the 150 days. Figure 4 illustrates the predominance of *Methanopyrus* and *Thermofilum*, which are hyperthermophiles microorganisms classified within the *Euryarchaeota* and *Crenarchaeota* phyla, respectively. The cyanobacteria found in the sargassum leachates at 150 days are likely to be related to their decomposition, as they normally, like other photosynthetic organisms, require nutrients such as nitrogen and phosphorus for their growth, causing massive growth under favorable conditions such as high temperature and nutrient availability.

3.5 *Greenhouse gas production*

Figure 5 shows the production of greenhouse gasses during the degradation of *Sargassum spp*. Methane (CH_4), hydrogen sulfide (H_2S) and carbon dioxide (CO_2) were analyzed for 35 days until no pressure measurements could be taken in the 250 ml serological bottles. The results show that during the *Sargassum spp.* degradation, carbon dioxide and hydrogen sulfide were the most abundant reaching values of 28 and 0.4% respectively, compared to methane at 35 °C. These results indicate that the microorganisms present in the biomass are more active at warmer temperatures. As observed in Figure 4, greenhouse gases production occurs during the first 10 days of decomposition with microorganisms endemic to sargassum. It is worth mentioning that hydrogen sulfide is striking because it is the most dangerous gas produced during this process. Figure 5 shows that more than 4,400 ppm is produced for every 10 grams of sargassum in a 250-milliliters volume under anaerobic conditions. Thus, the fresh biomass that accumulates on the Caribbean beaches will contribute large amounts of greenhouse gases. The greenhouse gas concentrations obtained in this study are higher than those reported by other authors. For example, Rodríguez-Martínez *et al*., (2024) reported 10 ppm H_2S in degraded sargassum on the beach. While, Resiere *et al*., (2020) studied the effects of this gas on sargassum workers, indicating effects at 10 ppm and above. Our results are higher as we conducted the measurement in a fully enclosed, controlled environment that favors anaerobic digestion. Consequently, this maximizes the potential damage caused by *Sargassum* decomposition.

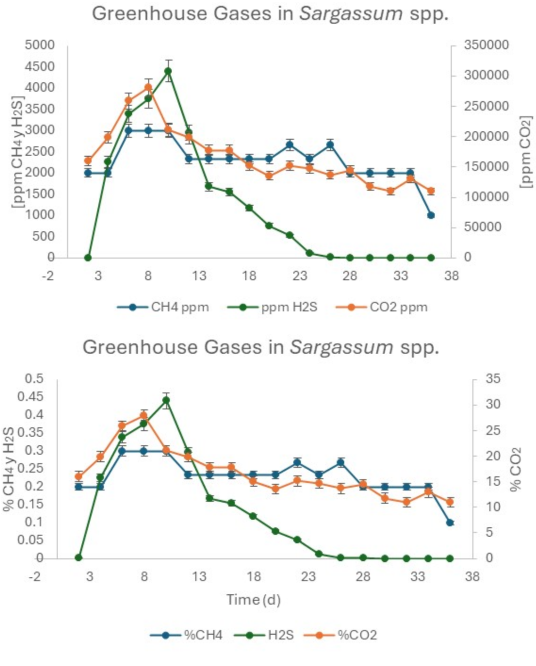

Fig. 5 Determination of greenhouse gases in *Sargassum* spp.

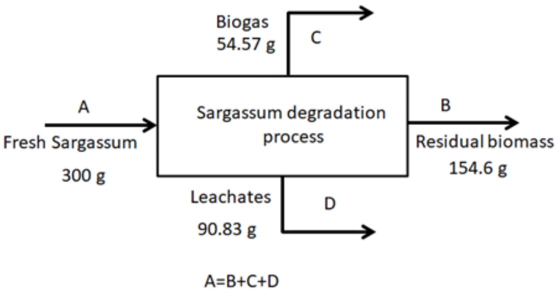

Fig. 6 General balance of degraded sargassum biomass.

The highest gas production occurs during the initial days of degradation. This finding is supported by our metagenomic analysis. During the initial stage of degradation, the most active microbial communities were the *Thermofilum* and *Methanopyrus* genera. These sulphate-reducing bacteria are responsible of the degradation of the consortium, since they use sulfur polymers to produce the gases.

By conducting a general balance of the biomass consortium (see Figure 6), we can obtain valuable information for estimating future sargassum upwelling damage. Starting from an initial biomass of 300 grams, it was found that the production of leachates and gases is dependent on the present microbial community. After 150 days, it was found that 18.2% of the biomass had transformed into gases and water evaporation, while 30.27% had degraded as leachate. For instance, in the case of the great Atlantic Sargassum belt (Wang *et al* 2019), it was estimate that of the 20 million tons of sargassum calculated in 2018, 18.2% by weight into gases and water evaporation and the degradation of 30.27% as leachate after 150 days. It can be extrapolated that the degradation of sargassum in the Atlantic Ocean results in the production of approximately 6 million tons of leachates and 3.63 million tons of greenhouse gases. This situation is problematic, and it is necessary to find new applications for the biomass that reaches the Mexican Caribbean. In 2022, the Mexican Navy reported that 52,647 tons of sargassum were collected in Quintana Roo, Mexico. This suggests that approximately 15,936.24 tons of leachate and a substantial amount of greenhouse gases were produced, depending on the anaerobic conditions that can form in the sargassum piles that reach the Caribbean Sea. The issue of greenhouse gas and heavy metal production, as a result of leachates and brown tides, represents a grave environmental concern in the Mexican Caribbean region, as previously highlighted.

The description of this biodiversity provides a basis for understanding the role that microbial communities play in the degradation of sargassum. As previously documented, epiphytic communities associated with sargassum decomposition have been reported in both marine and terrestrial ecosystems (Hervé *et al*., 2021). However, a more comprehensive understanding of the functionality of phycosphere is imperative, as it appears that the degradation of sargassum is influenced not only by microorganisms but also by a variety of environmental factors that were not thoroughly addressed in this study. This work serves as an approximation to the complex reality of the degradation of this macroalgae. It is evident that microorganisms play a significant role in the production of greenhouse gases and leachates. However, further research is required to explore the decomposition process in marine and terrestrial environments in greater depth. It is imperative to consider the elevated temperature that is generated in the sargassum compost once it has reached the beach, as well as the parameters in the sea, including the salinity, oxygen content, epiphytic organisms of the macroalgae, and those that are associated with the phycospere.

Conclusions

This study examined the natural degradation of a consortium of *Sargassum* spp. 150 days to investigate the structural and physical changes that occur during degradation. The study identified spatiotemporal changes in the microbial communities involved in the degradation of pelagic sargassum. After 150 days, the macroalgae exhibited a 29% loss in carbon content, which was attributed to the loss of carbon structures resulting from the production of leachates and greenhouse gases. Conversely, there was a significant increase in nitrogen content, modifying the C/N ratio due to nitrogen capture by the cyanobacteria group. Among the changes observed in the microbial community, Archaea belonging to the genera *Thermofilum* and *Methanopyrus* were identified in the initial stages of degradation (0-30 days). These archaea are capable of reducing sulfate and have been identified as the primary producers of greenhouse gases.

The populations of *Thermofilum* and *Acidiphilum* have been identified as key factors in the depolymerization of macroalgae. These genera were detected following a 30-day degradation period, suggesting their role as primary agents responsible for this form of degradation. This process enables the release of organic compounds, as well as leachates and heavy metals, which can have a detrimental effect on water bodies. The production of greenhouse gases approaches 5,000 ppm of H_2S, exceeding the regulatory limits established for this gas. Meanwhile, CO_2 and methane levels decline over time. Although the production of these gases is substantial, it pales in comparison to H_2S given its status as the most hazardous gas for the environment and human health. The above findings are highly relevant for the management of sargassum and the implementation of new policies for managing this macroalgae. This is a significant issue for the Mexican government, as these types of studies will help to make the management of marine resources more efficient and effective.

Acknowledgements

The authors would like to thank the TecNM 20185.24-P project, and IQ. Alberto Cortes, for his support in molecular biology techniques, Tanit Toledano Thompsom for the conservation of the microbial communities and technical support provided.

References

A. Sluiter, B. Hames, R. Ruiz, C. S., Slui, J., & ter, D. Templeton, and D. C. (2008). Determination of Structural Carbohydrates and Lignin in Biomass: Laboratory Analytical Procedure (LAP). *Technical Report NREL/ TP -510 -42618, January*, 1–15.

Alderkamp, A., Rijssel, M. Van, & Bolhuis, H. (2007). *Characterization of marine bacteria and the activity of their enzyme systems involved in degradation of the algal storage glucan laminarin*. *59*, 108–117. `https://doi.org/10.1111/j.1574-6941.2006.00219.x`

Ali, I., & Bahadar, A. (2017). Red Sea seaweed (*Sargassum* spp.) pyrolysis and its devolatilization kinetics. *Algal Research*, *21*(January 2019), 89–97. `https://doi.org/10.1016/j.algal.2016.11.011`

Álvarez-Rocha, M., & Ortegón-Aznar, I. (2022). Can the *Sargassum* sp. drift influence on cover and morphology of Syringodium filiforme (Cymodoceaceae) in the Mexican Caribbean? *Hidrobiologica*, *32*(1), 51–58. `https://doi.org/10.24275/uam/izt/dcbs/hidro/2022v32n1/Alvarez`

Alzate-Gaviria, L., Domínguez-Maldonado, J., Chablé-Villacís, R., Olguin-Maciel, E., Leal-Bautista, R. M., Canché-Escamilla, G., Caballero-Vázquez, A., Hernández-Zepeda, C., Barredo-Pool, F. A., & Tapia-Tussell, R. (2020). Presence of Polyphenols Complex Aromatic “Lignin” in *Sargassum* spp. from Mexican Caribbean. *Journal of Marine Science and Engineering*, *9*(1), 6. `https://doi.org/10.3390/jmse9010006`

Antonio-Martínez, F., Henaut, Y., Vega-Zepeda, A., Cerón-Flores, A. I., Raigoza-Figueras, R., Cetz-Navarro, N. P., & Espinoza-Avalos, J. (2020).

Leachate effects of pelagic Sargassum spp. on larval swimming behavior of the coral Acropora palmata. 1–13. https://doi.org/10.1038/s41598-020-60864-z

Antúnez-Argüelles, E., Herrera-Bulnes, M., Torres-Ariño, A., Mirón-Enríquez, C., Soriano-García, M., & Robles-Gómez, E. (2020). Enzymatic-assisted polymerization of the lignin obtained from a macroalgae consortium, using an extracellular laccase-like enzyme (Tg-laccase) from Tetraselmis gracilis. *Journal of Environmental Science and Health - Part A Toxic/Hazardous Substances and Environmental Engineering*, *55*(6), 739–747. https://doi.org/10.1080/10934529.2020.1738171

Ardalan, Y., Jazini, M., & Karimi, K. (2018). Sargassum angustifolium brown macroalga as a high potential substrate for alginate and ethanol production with minimal nutrient requirement. *Algal Research*, *36*(February), 29–36. https://doi.org/10.1016/j.algal.2018.10.010

Bikker, P., van Krimpen, M. M., van Wikselaar, P., Houweling-Tan, B., Scaccia, N., van Hal, J. W., Huijgen, W. J. J., Cone, J. W., & López-Contreras, A. M. (2016). Biorefinery of the green seaweed Ulva lactuca to produce animal feed, chemicals and biofuels. *Journal of Applied Phycology*, *28*(6), 3511–3525. https://doi.org/10.1007/s10811-016-0842-3

Biswas, B., Singh, R., Krishna, B. B., Kumar, J., & Bhaskar, T. (2017). Pyrolysis of azolla, *Sargassum tenerrimum* and water hyacinth for production of bio-oil. *Bioresource Technology*, *242*, 139–145. https://doi.org/10.1016/j.biortech.2017.03.044

Bolger, A. M., Lohse, M., & Usadel, B. (2014). Trimmomatic: A flexible trimmer for Illumina sequence data. *Bioinformatics*, *30*(15), 2114–2120. https://doi.org/10.1093/bioinformatics/btu170

Borines, M. G., de Leon, R. L., & Cuello, J. L. (2013). Bioethanol production from the macroalgae *Sargassum* spp. *Bioresource Technology*, *138*, 22–29. https://doi.org/10.1016/j.biortech.2013.03.108

Chávez, V., Uribe-Martínez, A., Cuevas, E., Rodríguez-Martínez, R. E., van Tussenbroek, B. I., Francisco, V., Estévez, M., Celis, L. B., Monroy-Velázquez, L. V., Leal-Bautista, R., Álvarez-Filip, L., García-Sánchez, M., Masia, L., & Silva, R. (2020). Massive influx of pelagic *Sargassum* spp. On the coasts of the Mexican Caribbean 2014–2020: Challenges and opportunities. *Water (Switzerland)*, *12*(10), 1–24. https://doi.org/10.3390/w12102908

Devault, D. A., Pierre, R., Marfaing, H., Dolique, F., & Lopez, P. J. (2021). Sargassum contamination and consequences for downstream uses: a review. *Journal of Applied Phycology*, *33*(1), 567–602. https://doi.org/10.1007/s10811-020-02250-w

Egan, S., Kumar, V., Nappi, J., & Gardiner, M. (2017). Microbial Diversity and Symbiotic Interactions with Macroalgae. In and L. M. Martin Grube, Joseph Seckbach (Ed.), *Algal and Cyanobacteria Symbioses* (pp. 493–546). World Scientific.Estrada Medina, 2019. (2019). *El karst de Yucatán : su origen , morfología y biología The karst of Yucatan : its origin , morphology and biology Resumen*. 1–18.

Fenner, M. W. (2008). Genome sequence of Thermofilum pendens reveals an exceptional loss of biosynthetic pathways without genome reduction. *Lawrence Berkeley National Laboratory.*

García-Sánchez, M., Graham, C., Vera, E., Escalante-Mancera, E., Álvarez-Filip, L., & van Tussenbroek, B. I. (2020). Temporal changes in the composition and biomass of beached pelagic sargassum species in the Mexican Caribbean. *Aquatic Botany*, *167*. https://doi.org/10.1016/j.aquabot.2020.103275

Goecke, F., Labes, A., Wiese, J., & Imhoff, J. F. (2010). *Chemical interactions between marine macroalgae and bacteria*. *409*, 267–299. https://doi.org/10.3354/meps08607

Hervé, V., Lambourdière, J., René-Trouillefou, M., Devault, D. A., & Lopez, P. J. (2021). Sargassum differentially shapes the microbiota composition and diversity at coastal tide sites and inland storage sites on Caribbean Islands. *Frontiers in Microbiology*, *12*(October), 1–14. https://doi.org/10.3389/fmicb.2021.701155

Leal-Bautista, R. M., Rodríguez-García, J. C., Acosta-González, G., Chablé-Villacís, R., Tapia-Tussell, R., Bautista-García, J. E., Olguín-Maciel, E., Alzate-Gaviria, L., & González-López, G. (2024). Evaluation of leachates generated by *Sargassum spp*. in the Mexican Caribbean: Part 1 Spatial variations. *Water*, 16(9), 1251. https://doi.org/10.3390/w16091251

Lim, S. J., & Wan Aida, W. M. (2017). Extraction of Sulfated Polysaccharides (Fucoidan) From Brown Seaweed. In *Seaweed Polysaccharides: Isolation, Biological and*

Biomedical Applications. Elsevier Inc. https://doi.org/10.1016/B978-0-12-809816-5.00003-7

López-Contreras, A. M., Van Der Geest, M., Deetman, B., Van Den Burg, S., & Brust, H. (2021). Opportunities for valorisation of pelagic sargassum in the Dutch Caribbean. In *Wageningen Food & Biobased Research*. www.wur.eu/wfbr

Mei, X., Wu, C., Zhao, J., Yan, T., Jiang, P., & Walsh, D. A. (2019). *Community Structure of Bacteria Associated With Drifting Sargassum horneri, the Causative Species of Golden Tide in the Yellow Sea*. *10*(May). https://doi.org/10.3389/fmicb.2019.01192

Méndez, M. A. P., Hernández, R. S., Palma, D. J., & García, S. S. (2011). Caracterización química del compostaje de residuos de caña de azúcar en el sureste de México. *Interciencia*, *36*, 45–52.

Milledge, John J., & Harvey, P. J. (2016a). Ensilage and anaerobic digestion of *Sargassum muticum*. *Journal of Applied Phycology*, *28*(5), 3021–3030. https://doi.org/10.1007/s10811-016-0804-9

Milledge, John J., & Harvey, P. J. (2016b). Golden Tides: Problem or golden opportunity? The valorisation of sargassum from beach inundations. *Journal of Marine Science and Engineering*, *4*(3). https://doi.org/10.3390/jmse4030060

Milledge, John James, Maneein, S., Arribas, E., & Bartlett, D. (2020). *Sargassum Inundations in Turks and Caicos : Methane*.

Morrissey, K. L., Lopez, P. J., Hervé, V., Lambourdière, J., & René-trouillefou, M. (2021). *Sargassum Differentially Shapes the Microbiota Composition and Diversity at Coastal Tide Sites and Inland Storage Sites on Caribbean Islands*. *12*(October), 1–14. https://doi.org/10.3389/fmicb.2021.701155

Nielsen, B. V., Milledge, J. J., Hertler, H., Maneein, S., Al Farid, M. M., & Bartlett, D. (2021). Chemical characterisation of sargassum inundation from the Turks and Caicos: Seasonal and post stranding changes. *Phycology*, *1*(2), 143–162. https://doi.org/10.3390/phycology1020011

Olguin, E., Rosa, M., Leal, M., Liliana, B., Gaviria, A., & Domínguez, J. (2022). *Environmental impact of Sargassum spp . landings : an evaluation of leachate released from natural decomposition at Mexican Caribbean coast*. *0123456789*. https://doi.org/10.1007/s11356-022-22123-8

Oliveira, J. V., Alves, M. M., & Costa, J. C. (2015). Optimization of biogas production from *Sargassum* sp. using a design of experiments to assess the co-digestion with glycerol and waste frying oil. *Bioresource Technology*, *175*, 480–485. https://doi.org/10.1016/j.biortech.2014.10.121

Ponnusamy, V. K., Nguyen, D. D., Dharmaraja, J., Shobana, S., Banu, J. R., Saratale, R. G., Chang, S. W., & Kumar, G. (2019). A review on lignin structure, pretreatments, fermentation reactions and biorefinery potential. *Bioresource Technology*, *271*, 462–472. https://doi.org/10.1016/J.BIORTECH.2018.09.070

Rabemanolontsoa, H., & Saka, S. (2013). Comparative study on chemical composition of various biomass species. *RSC Advances*, *3*(12), 3946–3956. https://doi.org/10.1039/c3ra22958k

Resiere, D., Mehdaoui, H., Florentin, J., Gueye, P., Lebrun, T., Blateau, A., Viguier, J., Valentino, R., Brouste, Y., Kallel, H., Megarbane, B., Cabie, A., Banydeen, R., & Neviere, R. (2020). Sargassum seaweed health menace in the Caribbean: clinical characteristics of a population exposed to hydrogen sulfide during the 2018 massive stranding. *Clinical Toxicology*, 1–9. https://doi.org/10.1080/15563650.2020.1789162

Robledo, D., & Vázquez-Delfín, Er. (2019). Sargazo, conociendo al “enemigo.” *Revista Avance y Perspectiva*, *5*(3). https://avanceyperspectiva.cinvestav.mx/sargazo-conociendo-al-enemigo/?print-posts=pdf

Rodríguez-Martínez, R. E., Gómez Reali, M. Á., Torres-Conde, E. G., & Bates, M. N. (2024). Temporal and spatial variation in hydrogen sulfide (H_2S) emissions during holopelagic *Sargassum* spp. decomposition on beaches. *Environmental Research*, *247*, 118235. https://doi.org/10.1016/J.ENVRES.2024.118235

Rodríguez-Martínez, R. E., Roy, P. D., Torrescano-Valle, N., Cabanillas-Terán, N., Carrillo-Domínguez, S., Collado-Vides, L., García-Sánchez, M., & van Tussenbroek, B. I. (2020a). Element concentrations in pelagic sargassum along the Mexican Caribbean coast in 2018-2019. *PeerJ*, *2020*(2), 1–19. https://doi.org/10.7717/peerj.8667

Rodríguez-Martínez, R. E., Roy, P. D., Torrescano-Valle, N., Cabanillas-Terán, N., Carrillo-Domínguez, S., Collado-Vides, L., García-Sánchez, M., & van Tussenbroek, B. I. (2020b). Element concentrations in pelagic sargassum along the Mexican Caribbean coast in 2018-2019. *PeerJ*, *8*, e8667. `https://doi.org/10.7717/peerj.8667`

Saldarriaga-Hernandez, S., Melchor-Martínez, E. M., Carrillo-Nieves, D., Parra-Saldívar, R., & Iqbal, H. M. N. (2021). Seasonal characterization and quantification of biomolecules from sargassum collected from Mexican Caribbean coast – A preliminary study as a step forward to blue economy. *Journal of Environmental Management*, *298*(August). `https://doi.org/10.1016/j.jenvman.2021.113507`

Serebryakova, A., Aires, T., Viard, F., Serrão, E. A., & Engelen, A. H. (2018). Summer shifts of bacterial communities associated with the invasive brown seaweed *Sargassum muticum* are location and tissue dependent. *PLoS ONE*, *13*(12), 1–18. `https://doi.org/10.1371/journal.pone.0206734`

Takagi, T., Kuroda, K., & Ueda, M. (2018). Platform construction of molecular breeding for utilization of brown macroalgae. *Journal of Bioscience and Bioengineering*, *125*(1), 1–7. `https://doi.org/10.1016/j.jbiosc.2017.08.005`

Thompson, T.M., Young, B. R., & Baroutian, S. (2020). Pelagic sargassum for energy and fertiliser production in the Caribbean: A case study on Barbados. *Renewable and Sustainable Energy Reviews*, *118*, 109564. `https://doi.org/10.1016/j.rser.2019.109564`

Thompson, Terrell M., Young, B. R., & Baroutian, S. (2019). Advances in the pretreatment of brown macroalgae for biogas production. *Fuel Processing Technology*, *195*. `https://doi.org/10.1016/j.fuproc.2019.106151`

Torralba, M. G., Franks, J. S., Gomez, A., Yooseph, S., Nelson, K. E., & Grimes, D. J. (2017). Effect of Macondo Prospect 252 Oil on microbiota associated with pelagic sargassum in the Northern Gulf of Mexico. *Microbial Ecology*, *73*(1), 91–100. `https://doi.org/10.1007/s00248-016-0857-y`

Vázquez-Delfín, E., Freile-Pelegrín, Y., Salazar-Garibay, A., Serviere-Zaragoza, E., Méndez-Rodríguez, L. C., & Robledo, D. (2021). Species composition and chemical characterization of sargassum influx at six different locations along the Mexican Caribbean coast. *Science of the Total Environment*, *795*. `https://doi.org/10.1016/j.scitotenv.2021.148852`

Wang, M., Hu, C., Barnes, B. B., Mitchum, G., Lapointe, B., & Montoya, J. P. (2019). The great Atlantic sargassum belt. *Science*, *364*(6448), 83–87. `https://doi.org/10.1126/science.aaw7912`

Wood, D. E., & Salzberg, S. L. (2014). Kraken: Ultrafast metagenomic sequence classification using exact alignments. *Genome Biology*, *15*(3). `https://doi.org/10.1186/gb-2014-15-3-r46`