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# Characterization of sargassum accumulated on Dominican beaches in 2021: analysis of heavy, alkaline and alkaline-earth metals, proteins and fats

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#### Abstract

This paper presents the characterization of sargassum that reached the shores of eigth Dominican beaches in 2021. The analysis of heavy, alkaline and alkaline-earth metals was performed by ICP-OES. Twelve heavy metals were studied, with the highest concentrations corresponding to Fe, As, and Zn. Regarding the alkaline and alkaline-earth metals, the highest concentrations were detected for Ca, K, Na and Mg. The high values of arsenic and alkali and alkaline-earth metal salts do not suggest using these algae in agriculture. It is recommended to carry out arsenic speciation studies to assess whether the form in which it is found is bioavailable for plants and animals. The heavy metal contamination index was determined, which ranged between 0.318 and 3.279. Finally, for the first time in the country, the organic fraction of sargassum was analyzed.

Keywords: algae, Sargassum, heavy metals, proteins, fats, Dominican Republic

#### 1. Introduction

Since 2011, the massive arrival of brown algae has been reported on the Caribbean coast from the arc of the Lesser Antilles to Mexico. This phenomenon has caused negative impacts on coastal and marine ecosystems and essential economic sectors for the geographical area, such as fishing and tourism (Milledge and Harvey, 2016; van Tussenbroek et al., 2017; Chávez et al., 2020; Desrochers et al., 2020; Oxenford et al., 2021; UNEP-CEP, 2021).

Today, it is known that the origins of these algae come mainly from the North-Equatorial Recirculation Region (NERR), with the mouth of the Amazon River on the Atlantic coast of South America being especially important (Gower et al., 2013; Johnson et al. al., 2013; Sissini et al., 2017; Mendez-Tejeda and Rosado Jiménez, 2019; Wang et al., 2019; Johns et al., 2020; Trinanes et al., 2021; Skliris et al., 2022). The contributions of nutrients from the rivers of South America and West Africa seem to be one of the causes of the excessive growth of sargassum, as well as of the metal content in the algae, according to a recent study by Dassié et al. (2022). They characterized sargassum at seven points in the Atlantic between the two continents.

Generally, two to three varieties of sargassum *S. fluitans III, S. natans I* and *S. natans VIII* coexist (Fernandez et al., 2017; Milledge et al., 2020; Davis et al., 2021; Tejada et al., 2021; Vázquez-Delfín et al., 2021; Dassié et al., 2022; Machado et al., 2022), only the arrival at Cienfuegos Bay (Cuba) in 2012 was monospecific (Moreira et al., 2013).

Given the detrimental effects that the massive accumulation of these species on the coastline has on the economy, it has been suggested that benefits could be obtained from such events (Oyesiku and Egunyomi

2014; Milledge and Harvey, 2016; Desrochers et. al., 2020; Liranzo-Gómez et al., 2021; Oxenford et al., 2021;). Brown algae have been used, among other applications, as agricultural fertilizers (López-Mosquera et al., 2011; Desrochers et. al., 2020; Oxenford et al., 2021;) and in animal feed (Moreira et al., 2012; Desrochers et al., 2020; Oxenford et al., 2021;). However, the possible applications of these algae must be analyzed with caution due to their ability to accumulate toxic metals (Davis et al., 2003; Abkenar et al., 2021; Anbazhagan et al., 2021; Dassié et al., 2022; Peng et al., 2022; Siddique et al., 2022). Therefore, they are used as bioindicators for contamination by these metals (Davis et al., 2003; Pan et al., 2018; Al-Homaidan et al., 2021).

In the case of the Dominican Republic, two papers have been published on the content of heavy, alkaline, alkaline-earth metals, and rare earth metals in samples of Sargassum fluitans and Sargassum natans (Fernández et al., 2017; Tejada-Tejada et al., 2021). Fernandez et al. (2017) studied sargassum on three beaches in 2015: Guayacanes (province of San Pedro de Macorís), Boca Chica (Santo Domingo) and Ojeda (Barahona). For their part, Tejada-Tejada et al. (2021) analyzed the sargassum that reached five beaches in 2019: Juan Dolio (province of San Pedro de Macorís), Caleta and San Andrés (Santo Domingo), Monte Río (Azua) and La Saladilla (Barahona). Fernandez et al. (2017) did not detect worrying levels of any of the elements studied in the algal biomass but suggested that the possible use of sargassum as an agricultural fertilizer or in animal feed could increase the concentrations of toxic metals and salts in the soil and/or incorporating them into the food chain. Likewise, when comparing the concentrations of rare earth in seawater and algal biomass, they detected significant anomalies of several orders of magnitude, confirming that sargassum can bioaccumulate these metals (Fernandez et al., 2017). Regarding the work of Tejada-Tejada and collaborators (2021), they showed that the concentrations of the metals analyzed in the studied species of sargassum are below the acceptable limits to be used as fertilizer and in animal feed without risk of intoxication but state that additional analyzes are required for other toxic elements such as Hg and As, in order to guarantee the safety of the use of these algae for agricultural purposes. Regarding the comparison of the biosorption capacity of the two species studied (S. fluitans and S. natans), the statistical analysis of the data allowed us to conclude that there is no significant difference in the concentrations of the metals studied, which could be an indicator that the mechanisms and biochemical factors involved in biosorption capacity are similar between both species (Tejada-Tejada et al., 2021).

Taking into account the temporal variations in the sargassum content, it is necessary to systematically monitor the composition of the sargassum in order to decide on its possible applications. The present work aims to determine the content of heavy, alkaline, and alkaline-earth metals and part of the organic fraction (proteins and total fats) in sargassum that reached Dominican beaches in 2021, to analyze the possibility of its use in agriculture.

# 2. Materials and methods

# 2.1. Sampling and preparation of sargassum samples

Sampling was carried out between March and August 2021 on eight beaches in the Dominican Republic: Bávaro and Punta Cana in the province of Altagracia, Juan Dolio and Guayacanes in the province of San Pedro de Macorís, San Andrés in the province of Santo Domingo, Nigua in San Cristóbal, Enriquillo in Barahona and Juancho in Pedernales. Figure 1 shows the location of the beaches and indicates the exact coordinates of the sampling points.



 Figure 1. Sargassum sampling sites and images of the studied beaches. 1-Bávaro(19Q-562177.97mE;2065327.96mN); 2-Punta Cana(19Q-568159.13mE; 2050047.81mN); 3-Juan Dolio(19Q-456104.77mE;2037360.29mN); 4-Guayacanes. (19Q-452335.37mE;2036758.09mN); 5-SanAndrés (19Q-433210.67mE;2038902.24mN); 6-Nigua(19Q-89345.54mE;2032036.23mN); 7-Enriquill(19Q-261261.00mE;1978579.11mN); 8-Juancho(19Q-255785.78mE; 97224.52mN)

The samples were composed, taken in at least five points on each beach, in the sea before the sargassum reached the sand, and the water was allowed to drain through a 2-mm mesh for 5 minutes before packaging. The presence of the three species of sargassum that reach the Caribbean, *S. fluitans III, S. natans I* and *S. natans VIII*, with a predominance of *Sargassum fluitans III*, was verified. The species were identified based on their morphological differences at the level of the phyllodes and aerocysts as reported in the specialized literature (Morerira et al. 2013, Oyesiku et al., 2014; Schell et al., 2015; Amaral-Zettler et al., 2017, Littler and Littler, 2000; Parr, 1939).

The samples were transported in plastic bags to the laboratory, where they were immediately washed with drinking water and distilled water and dried in the sun for 10 hours a day for seven days, after which they were dried in a laboratory oven (Thelco) for 24 hours at 100°C. The water loss in the samples due to weight difference ranged between 85 and 88%, values in the range of 81-88% reported by other authors (Desrochers et. al., 2020; Milledge et al., 2020; Amador-Castro et al., 2021). Subsequently, the samples were pulverized and sieved, guaranteeing a homogeneous grain size between 0.5 and 1.5 mm.

# 2.2. Procedure for the determination of metals by ICP-OES

For the determination of heavy, alkaline, and alkaline-earth metals, the digestion of sargassum was carried out by preparing 500 mg of each sample in duplicate, which was poured into a 75 mL Teflon vial, to which 7 mL of 69% HNO<sub>3</sub>; 1.5 mL of  $H_2O_2$ ; 0.5 mL of HF (40-45%) and 2 mL of deionized water. For digestion, a PerkinElmer Titan MPS model digester was used. Residual HF was treated with  $H_3BO_3$ , and the vials were placed back in the sample preparation system. After filtering, the content of the samples was poured into 50

mL volumetric flasks, completed with deionized water up to the volume line. Subsequently, five milliliters of them were taken, diluted in a flask, and calibrated in 50 mL with a 2% w/v solution of nitric acid prepared with deionized water. A PerkinElmer Optima 8000 inductively coupled plasma optical emission spectrometer (ICP-OES) was used to quantify heavy, alkaline and alkaline-earth metals. A calibration curve was established for each element with their respective spectra, and the analysis was performed in duplicate. Table S1 shows specifications and the calibration parameters for metals determination by ICP-OES.

# 2.3. Procedure for the determination of total proteins and fats

To determine total proteins and fats, the fresh samples were frozen at -20 °C and lyophilized for 72 hours at a temperature of -55 °C in a Yamato-DC401 lyophilizer. Proteins were determined by the method of Lowry (1951) and total fats by the procedure of Bligh and Dyer (1959).

# 2.4. Heavy metal pollution index

The heavy metal pollution index (HMPI) is determined by equation 1 using the procedure proposed by Giusti (2001) for the determination of the metal pollution index in algae and by Usero et al. (1996) for the determination of the rate of contamination by metals in mollusks.

$$HMPI = (M_1 \times M_2 \times M_3 \times \dots \times M_n)^{1/n}$$
 (1)

where  $M_n$  is the concentration of metal n in the seaweed expressed in mg/kg of dry seaweed.

# 2.5 Statistical analysis

All data analyses were conducted using SPSS 25 and Excel 2019. First, we proceeded to determine the different correlations between studied variables using Pearson's correlation matrix. Then, principal components analysis (PCA) was undertaken to identify clusters of related parameters (as components).

# 3. Results and discussion

Table 1 shows the results of the content of heavy metals in the sargassum on the eight studied beaches. Eleven heavy metals were analyzed, with the highest concentrations corresponding to Fe (116-998 mg/kg), As (35-101 mg/kg) and Zn (4-11 mg/kg). The presence of molybdenum and selenium was not detected in any of the studied beaches, while cadmium was detected in 3 of the 8 studied beaches (37.5%), nickel in 5 (62.5%), and lead in 6 (75%). The remaining elements were present in the samples from all the beaches. From the means of heavy metal content, the order of presence of these elements from highest to lowest is: Fe < As < Zn < Cu < Pb < Ni < Cr < Co < Cd.

Table 1- Heavy metal content (mg/kg) in the sargassum on eight Dominican beaches. \*Range of maximum permitted levels of heavy metals for use in agricultural soils and animal feed in different countries in mg/kg (Kovalsky, 1974; El Bassam and Tietjen, 1977; Linzon, 1978; Kloke, 1979; Kitagishi, 1981; FAO, 1984; McDowell, 1992; Kabata-Pendias, 2001; NOM-147-SEMARNAT-SSA1-2004; Aganga et al., 2005; EU, 2006; Galán and Romero, 2008; Belmonte et al. 2010; Singh et al., 2011; Elliott, Frio and Jarman, 2017; EU, 2019; Kfle et al., 2020).

	As	Cd	Со	Cr	Cu	Fe	Ni	Pb	Zn	HMPI
1 Dávono	67.75±	< 0.001	$0.47\pm$	$0.5\pm$	2.632±	309.13±	0.201±	< 0.01	11.46±	0 875
1- Davaro	1.54	< 0.001	0.01	0.01	0.03	24.30	0.01	< 0.01	0.19	0.875
2 Dunto Cono	35.19±	< 0.001	$0.54\pm$	$0.54\pm$	2.302±	271.63±	< 0.001	0.35±	9.32±	0.651
2- Fund Cana	0.24	< 0.001	0.01	0.01	0.03	33.78	< 0.001	0.01	0.11	0.031
3 Juan Dalia	71.89±	< 0.001	0.36±	$0.15\pm$	$1.85\pm$	121.09±	< 0.001	< 0.01	3.83±	0.219
5- Juan Dono	1.36	< 0.001	0.01	0.01	0.01	20.26	< 0.001	< 0.01	0.01	0.516
1 Cueveene	73.99±	0.11±	0.31±	0.91±	2.364±	727.04±	$1.526 \pm$	0.74±	9.84±	2 270
4- Guayacalles	0.96	0.01	0.01	0.03	0.01	146.76	0.14	0.01	0.07	5.219
5 San Andréa	$80.52 \pm$	< 0.001	0.67±	$0.98\pm$	3.021±	517.54±	$0.506 \pm$	1.34±	10.84±	2 030
5- San Anures	1.13	< 0.001	0.02	0.07	0.02	79.29	0.01	0.08	0.10	2.039

6- Nigua	88.49± 0.93	0.16± 0.01	0.06± 0.01	$\begin{array}{c} 0.83 \pm \\ 0.03 \end{array}$	1.609± 0.01	998.64± 83.64	0.937± 0.04	$0.8\pm$ 0.05	7.6± 0.10	2.651
7- Enriquillo	101.89± 1.57	< 0.001	$\begin{array}{c} 0.53 \pm \\ 0.01 \end{array}$	0.41± 0.05	$1.374 \pm 0.02$	179.25± 42.57	< 0.001	3.41± 0.10	8.96± 0.02	0.819
8- Juancho	95.38± 0.94	$0.4\pm 0.01$	0.23± 0.01	1.62± 0.02	2.642± 0.02	116.70± 12.55	1.403± 0.08	1.19± 0.03	4.86± 0.04	3.204
*Agricultural soils	15-50	1-37	25-50	50-280	60-200	-	100-139	100-1000	70-600	-
*Animal feed	2-40	0.5-15	10-40	100	25-100	-	100	5-100	10-50	-

In order to compare our results with those obtained by other authors, the range of variation of the heavy metal content in sargassum recently published in studies with samples from the Dominican Republic (Fernández et al., 2017; Tejada-Tejada et al., 2021), Jamaica (Davis et al., 2021), Turks and Caicos Islands (Milledge et al., 2020), and in the Atlantic Ocean, near the area now known as the new Sargasso Sea (Dassié et al., 2022) was determined. The results are presented in Figure 2.





Figure 2. Comparison of heavy metal concentrations ranges in different Dominican, Caribbean beaches, and in the Atlantic Ocean in 2015-2021. The scales of each figure vary according to the metal. RD2015: the Dominican Republic 2015 (Fernández et al., 2017); RD2019: the Dominican Republic 2019 (Tejada-Tejada et al., 2021); RD2021: the Dominican Republic 2021 (this work); J2019: Jamaica 2019 (Davis et al., 2021); Mexico 2018 and 2019 (Vázquez-Delfín et al., 2021); TC2019: Turks and Caicos Islands 2019 (Milledge et al., 2020); O2018: Atlantic Ocean 2018 (Dassié et al., 2022).

As can be seen in figure 2, the metal concentrations in sargassum vary on both spatial and temporal scales, as reported in previous studies (Soerensen et al. 2014; Rodríguez-Martínez et al. 2020). On the one hand, it is known that pelagic algae are capable of concentrating these metals up to six orders of magnitude, compared to their concentration in seawater, but they do not accumulate each element in the same way (Abirhire and Kadiri 2011; Sadeghi et al. 2014; Fernández et al., 2017; Chen et al. 2018). On the other hand, recent studies show that these algae have proliferated in waters richer in these metals near the mouth of large rivers such as the Amazon, as a consequence of the contamination of their waters by heavy metals (Fernández et al., 2017; Dassié et al., 2022). The heavy metals can also enter the ocean by atmospheric inputs (Mart et al. 1982). Them, the heterogeneity in heavy metal concentrations observed between this study and by other authors might be linked to the origin of the Sargassum spp. and their trajectory along different oceanic currents and/or to regions contaminated with heavy metals as shown by Dassié et al. (2022).

When comparing with previous results of heavy metal determination carried out in 2015 (Fernández et al., 2017) and in 2019 (Tejada et al., 2021) on Dominican beaches, the values reported in our work for Co, Cr, Cd, Ni, and Zn are lower; for Cu and Pb they are in the same range of values and for As they are up to two times higher than the values found in samples from 2015 and 2019.

When studying the most recent works of sargassum analysis, the researchers' most significant concern focuses on the high values of arsenic. The arsenic values obtained in this study were higher than those reported in the Dominican Republic in samples from 2015. they are in the range detected in other Caribbean countries and are lower than those found in the Atlantic by Dassié et al. (2022). The high values of As found in the Atlantic Ocean show that the contamination problem with the metal comes from the origin of the formation of floating sargassum rafts. The content of this metal decreases as it moves towards the Caribbean, presumably, due to the reproduction of the algae and the less quantity of this element in our waters. Several authors have drawn attention to the consequences of the high content of arsenic in sargassum for applications related to agriculture due to the possible effects on soils, crops, and animals (Desrochers et. al., 2020; Rodríguez-Martínez et al., 2020; Milledge et al., 2020; Tejada-Tejada et al., 2021). When comparing our results with the range of maximum values established for arsenic in use in agricultural soils and animal feed (table 1), it is observed that in all cases, the arsenic content in the samples studied is in that range or exceeds it, which constitutes a limitation for purposes of agricultural use. The value of 40 mg/kg is established precisely for seaweed meal and raw materials for the feed from seaweed (EU, 2019).

Desrochers et al. (2020) warn that "the high arsenic content, as well as other components detected in several pelagic sargassum samples, can also damage soils in the long term and potentially pass into the food chain

through food crops." They also warn about the implications of the presence of As in developing products of interest for the pharmaceutical, cosmetic, and beauty products industries.

On the other hand, other authors warn about the importance of differentiating between the organic and inorganic fractions of As, where the latter is considered highly toxic (Johnson and Braman 1975; Circuncisão et al., 2018; Tirolien 2019; Milledge et al., 2020). Tejada-Tejada et al., 2021). A study of a sample of sargassum from Martinique found that 70 percent of the total arsenic was inorganic (Tirolien, 2019). In a study conducted in the 1970s on the Sargassum population of the Gulf of Mexico/Sargasso Sea, four samples of pelagic sargassum were collected from different locations (Bermuda, eastern Gulf of Mexico, Florida Strait, and Key West in deep water; Johnson and Braman 1975). In three samples, more than 82 percent of the total arsenic was found to be inorganic, in the form of arsenite (As III) and arsenate (As V), and one sample contained only 29 percent inorganic arsenic. For their part, Devault et al. (2022) confirm, when studying arsenic speciation in samples from Guadeloupe and Martinique, they determined that As(V) was the dominant form of arsenic, previous corroborating findings (Johnson and Braman 1975; Michel, 1985). Another interesting finding is that Devault's team found significantly lower values of total As, As(V), methylarsonic acid (MMA), and arsenobetaine (AsBet) in dry beach samples than in floating algae. For the As(III) and dimethylarsinic acid (DMA) species, no significant differences were observed between one type of sample and the other.

Recently, Ortega-Flores et al. (2022) showed that the main variables associated with the accumulation of arsenic in S. fluitans were the year's season, the content of copper, iron, uronic acids, and sulfates in fucoidan. The arsenic content in sargassum biomass reached maximum in the rainy season.

As for the rest of the heavy metals studied, cobalt and iron were studied for the first time in samples of sargassum in the Dominican Republic, and their values are within the range of the determined content of these elements in other Caribbean countries (Fernández et al., 2017; Tejada-Tejada et al., 2021; Davis et al., 2021; Vázquez-Delfín et al., 2021; Milledge et al., 2020). The other metals studied (Cd, Cr, Cu, Ni, Pb and Zn) are in the range of values studied by other authors for sargassum samples (Davis et al., 2021; Vázquez-Delfín et al., 2020). For all these elements, their content in sargassum is lower than the maximum limits established for their use in agricultural soils and animal feed.

In the present work, the contamination index with heavy metals was determined for the first time, to the best of our knowledge, for sargassum in our geographical area, specifically for the varieties S. fluitans and S. natans. The HMPI values ranged between 0.318 and 3.279. These values are higher than those calculated by Kaviarasan et al. (2018) for four species of algae (Halimeda gracilis, Padina pavonica, Sargassum swartzii and Turbinaria ornata) in the Wandoor area south of Andaman Island in western Thailand (0.025<HMPI<0.225), but much lower than those determined for the brown alga Fucus vesiculosus on the English coastline between the mouths of the Rivers Wear and Tees, with HMPI values ranging from 0.5 to 42 (Giusti, 2001). According to Giusti (2001), low HMPI values are expected in algae since the accumulated heavy metals are derived only from dissolved species present in seawater.

Based on experimental evidence, several authors consider that brown algae, and among them the varieties S. fluitans and S. natans, can be used as bioindicators of seawater contamination with heavy metals (Kaviarasan et al., 2018; Giusti, 2001). For this reason, it is recommended that, in future works, the HMPI index be used as an indicator to compare both water and algae contamination in order to determine its possible applications.

Regarding alkaline and alkaline-earth metals (Table 2), the highest concentrations were detected for Ca (40-68 g/Kg), K (29-46 g/Kg), Na (9-14 g/Kg), and Mg (6-7 g/Kg), in ascending order. These high values presuppose another inconvenience in using sargassum as fertilizers due to the high levels of sodium, calcium, and magnesium present in these algae that could cause salinization of cultivated soils. In this sense, the high

calcium values stand out, higher even in one case than the average geochemical values in the earth's crust, and potassium, higher than the geochemical values in all the samples studied. Likewise, the Ca, K, Mg, and Na values are above the mean concentrations of these salts in seawater.

Table 2- Content of alkali and alkaline-earth metals in sargassum in eight Dominican beaches and their standard values in the earth's crust and in the oceans. The concentration of the elements in mg/kg (\*Kovalsky, 1974 Fernández et al., 2017)

	Ве	Са	К	Mg	Na
1- Bávaro	4.13±	40125.7±	39628.53±	6366.98±	10228.04±
	0.11	348.44	148.58	30.13	140.34
2- Punta Cana	4.17±	45168.76±	36358.89±	6485.93±	9284.74±
	0.02	80.80	74.25	83.14	36.94
3- Juan Dolio	4.3±	50594.08±	38766.68±	6208.21±	9892.83±
	0.10	358.81	279.13	66.40	70.74
4-Guayacanes	4.83±	48816.88±	44034.89±	6319.9±	10907.48±
	0.18	283.12	317.41	57.13	127.53
5- San Andrés	4.15±	40470.04±	46026.57±	6710.13±	10498.04±
	0.13	284.19	741.20	30.50	82.71
6- Nigua	4.27±	67658.13±	32037.1±	7403.68±	11318.09±
	0.06	494.08	232.97	38.59	115.17
7- Enriquillo	4.23±	41761.32±	34366.23±	6994.62±	13521.53±
	0.22	517.56	428.21	37.99	73.73
8- Juancho	4.29±	46721.18±	28980.99±	7484.2±	11978.36±
	0.08	496.55	451.96	7.62	70.49
In Earth's crust*	-	52900	9100	32000	23000
In the ocean*	-	400	380	1200	10600

When comparing the behavior of alkali and alkaline-earth metals with previous studies (Figure 3), we can observe that the values obtained for Be and K are higher than those found by Fernández et al. (2017) in sargassum samples collected on beaches. Dominican, the Na values are similar to those observed in 2015, while the content of Ca and Mg are now lower. When comparing our results with others obtained in investigations in our region and the Atlantic Ocean, the concentrations of these salts are in the range reported by other authors (Davis et al., 2021; Vázquez-Delfín et al., 2021; Milledge et al., 2020).

Na, ppm           25000           20000           15000           10000           5000           0           RD2015           RD2015           RD2014           J2019           M2018-2019           TC2019		K, ppm 80000 70000 60000 30000 20000 0 RD2015 RD2021 J2019 M2018-2019 TC2019
6 5 4 3 2 1 0 R02015 RD2021 J2019 M2018-2019 TC2019	20000 18000 16000 12000 10000 8000 6000 6000 6000 6000 6000 6000 6000 6000 6000 6000 6000 6000 6000 6000 7	

Figure 3. Comparison of the intervals of variation of the concentrations of alkali and alkaline-earth metals in different Dominican, Caribbean, and Atlantic Ocean beaches in 2017-2021. The scales of each figure vary according to the metal. RD2015: Dominican Republic 2015 (Fernández et al., 2017); RD2021: Dominican Republic 2021 (this work); J2019: Jamaica 2019 (Davis et al., 2021); Mexico 2018 and 2019 (Vázquez-Delfín et al., 2021); TC2019: Turks and Caicos Islands 2019 (Milledge et al., 2020).

To establish the correlation between the studied metals and their spatial distribution, the different correlations between the metals determined experimentally at all sampling points were determined (Table 3). An important result of this analysis is precisely the spatial relationship between the concentration of metals and the distance traveled in the direction of the current from east to southwest of the Dominican Republic. From the Pearson matrix, it can be seen that there is a statistically significant increase along the coast for arsenic, magnesium, and sodium, which means that as the algae advance through our seas, it enriches in these metals, showing the bioaccumulation capacity of sargassum.

	Beaches	As	Cd	Co	Cr	Cu	Fe	Ni	Pb	Zn	Be	Ca	К	Mg	Na
Beaches	1														
As	.832*	1													
Cd	.649	.442	1		_										
Со	341	325	670	1											
Cr	.599	.356	.856**	322	1										
Cu	249	295	.141	.366	.501	1									
Fe	.059	.073	.053	447	.175	083	1								
Ni	.466	.357	.783*	605	.814*	.272	.479	1							
Pb	.705	.660	.028	.223	.099	399	122	047	1						
Zn	- 378	- 260	- 491	532	- 086	337	335	- 112	.103	1					
Be	.088	.116	233	- 353	.186	- 048	.343	.664	- 027	- 033	1				
Ca	238	157	326	- 843**	064	- 502	659	340	- 213	- 418	168	1			
ĸ	- 509	- 315	- 616	567	- 263	457	211	- 088	- 203	575	285	- 397	1		
Ma	830*	616	708*	.507	644	180	186	304	.203	280	251	411	733*	1	
Ne	.030	.010	.700	470	.044	160	.100	.374	.444	200	251	.411	755	1	1
Na	.819	.8/1	.359	185	.278	450	034	.236	.880	091	.119	.011	434	.615	1

Table 3- Pearson correlation matrix

\* Significant correlation for = 0.05 (2 tails)

Regarding the associations between elements, positive interactions (one element increases with the increase in the presence of the other) are observed for the pairs As-Na, Cd-Cr, Cd-Ni, Cd-Mg, Cr-Ni, and Pb- Na, while negative interactions (one element decreases its concentration with the increase of the other) were validated for the Co-Ca and K-Mg pairs.

Similarly, a principal components analysis of the entire sampling area was carried out to search for the main combinations of variables that may be associated with certain physical-spatial phenomena (see supporting information Table S2). Principal components analysis allows a representation of a large set of variables from a smaller space of variables that constitute a combination of the remaining variables, which allows their representation in the plane in order to achieve a better identification and interpretation of the phenomena with which they are associated. As can be seen in table S2, 5 main components were obtained, achieving a description of 95.55% of the variability of the variables considered.

The main components obtained adequately explained the variables considered, whereas As was the worst explained variable with 81.00% description. This value that is still considered adequate in this type of analysis. Table S3 shows the weight coefficients of each variable in each main component obtained.

As can be seen in Figure 4, principal component 1 explains 37.39% of the information in the original data table. This component is related to a phenomenon in which the subset of metallic elements (Co, K, Zn, and Cu) are opposed to the rest of the elements studied. This relationship could be fundamentally associated with the direction of the sea current from Punta Cana beach to Juancho beach. The relationship between both subsets can be better explained if the characteristics of the different points located in the path of the ocean current are taken into account.



Figure 4. Principal component analysis. The graph on the left shows the resulting subspace of reduced variables. The graphs on the right show the most important variables considered in the resulting subspace of

reduced variables (1-Bavaro,2-Punta Cana,3-Juan Dolio, 4-Guayacanes,5-San Andres, 6-Nigua,7-Enriquillo, and 8-Juancho).

For its part, principal component 2 explains 20.40% of the information in the original data table. This component is related to a phenomenon in which the subset of metallic elements (Co, Pb, As, Na and Mg) are opposed to the rest of the elements studied. These correlations are manifested fundamentally between the sampling points of the Enriquillo and Guayacanes beaches.

Principal component 3 explains 16.42% of the information in the original data table. This component describes a phenomenon in which the concentration of the subset of metallic elements (Fe and Ca) is opposed to the remaining metallic elements considered in the analysis. This phenomenon is clearly manifested between the Punta Cana, Juan Dolio and Nigua beaches.

Principal component 4 explains 13.18% of the information in the original data table. This component describes a phenomenon in which the concentration of the subset of metallic elements (Co, Cu, Cr, Mg and Cd) is opposed to the remaining metallic elements considered in the analysis. This phenomenon is clearly manifested between the beaches of San Andrés, Guayacanes, Enriquillo, and Nigua.

Finally, principal component 5 explains 8.17% of the information in the original data table. This component describes a phenomenon in which the concentration of the subset of metallic elements (K, Be, Na, As, and Cd) is opposed to the remaining metallic elements considered in the analysis. This phenomenon is clearly manifested between the Enriquillo, Juan Dolio, Guayacanes, and Juancho beaches.

Likewise, a multidimensional scaling analysis was carried out to represent in a plane the main correlations between the variables studied, considering the proximity between them as a comparison criterion (Table S4). The analysis was performed with an appropriate model fit with fit values greater than 98% and a normalized raw stress value. It should be noted that this analysis is much more general and may include a combination of the phenomena previously identified during principal component analysis. As can be seen in Figure 5, dimension 1 is related to a phenomenon in which the subset of metallic elements (Co, K, Zn, As, Na, Mg, Be, Ca, and Cu) are opposed to the rest of the elements studied, clearly defining two fundamental conglomerates of metallic elements. Likewise, similar results (formation of two clusters) were obtained for principal component 1, where the greatest explanation of the information contained in the original variable table was obtained. This phenomenon is clearly manifested among the conglomerates made up of Guayacanes, Nigua, and Juancho beaches, and the remaining beaches are very close to zero (0.01).



Figure 5. Multidimensional scaling analysis. The upper graph shows the resulting dimensions obtained from the analysis by variables. The lower graph shows the dimensions resulting from the spatial analysis (1-Bávaro, 2-Punta Cana, 3-Juan Dolio, 4-Guayacanes, 5-San Andrés, 6-Nigua, 7-Enriquillo, and 8-Juancho).

Finally, table 4 shows the analysis of the organic fraction of sargassum, detecting proteins in the range of 4 to 6% of the total weight and total fats in the range of 0.16 to 0.33% of the total mass. In the previous works in the Dominican Republic, there are no reports of the concentrations of proteins and fats in the sargassum samples studied. When comparing our results with other investigations in our geographical area, it can be seen that the protein content is in the range of the values reported by other authors between 2.5-15.4% (Bird et al., 1990; Wang et al., 2008; Chale-Dzul et al., 2017; Devault et al., 2021; Díaz-Vázquez et al. 2015; Machado et al., 2022). Regarding total fats, the values are at the lower limit of what is reported by other authors, 0.3-4.6% (Bird et al., 1990; Wang et al., 2008; Díaz-Vázquez et al. 2015; Saravanan et al., 2018; Milledge et al., 2020; Devault et al., 2021) or less than them. These values allow the evaluation of sargassum for applications in agriculture and animal feed as long as the standards established in the content of micropollutants are met.

Table 4- Content of lipids and proteins in sargassum in eight Dominican beaches.

Sample	Proteins (N x 6.25) %	Lipids %
1-Bávaro	5.64±0.27	0.33±0.02
2-Punta Cana	6.14±0.24	$0.17{\pm}0.01$
3-Juan Dolio	5.68±0.28	$0.17 \pm 0.01$
4-Guayacanes	5.61±0.24	0.22±0.01
5-San Andrés	4.04±0.16	$0.24{\pm}0.01$
6-Nigua	5.72±0.29	0.16±0.01
7-Enriquillo	4.16±0.21	0.19±0.01
8-Juancho	5.73±0.22	0.25±0.01

#### 4. Conclusions

When characterizing the sargassum that arrived at eight Dominican beaches during the year 2021, the presence of the three species of sargassum that reached the Caribbean, S. fluitans III, S. natans I and S. natans VIII, with a predominance of Sargassum fluitans III was verified. The content of eleven heavy metals was determined, the highest concentrations corresponding to Fe (116-998 mg/kg), As (35-101 mg/kg), and Zn (4-11 ppm). The presence of molybdenum and selenium was not detected in any of the beaches studied. Only arsenic was above the values defined in international standards for using sargassum in agriculture or animal feed. However, to reach a conclusive decision, work must be done on the speciation of arsenic to discover its bioavailability and possible transmission in the food chain. The heavy metal contamination index (HMPI) was determined for the first time, to the best of our knowledge, for sargassum in our geographical area, specifically for the varieties S. fluitans and S. natans. The HMPI values ranged between 0.318 and 3.279, being in the range of this indicator determined for other macroalgae in other areas of the planet. Regarding the alkaline and alkaline-earth metals, the highest concentrations were detected for Ca (40-68 g/Kg), K (29-46 g/Kg), Na (9-14 g/Kg) and Mg (6 -7 g/Kg), which presupposes another additional inconvenience for the use of sargassum as fertilizers due to the high levels of sodium, calcium and magnesium present in these algaes that could cause the salinization of cultivated soils. In this sense, the high potassium values stand out, higher than the geochemical values in all the samples studied. When evaluating the spatial relationship between the concentration of metals and the distance traveled by the sargassum in the direction of the marine current from east to southwest of the Dominican Republic, it was observed that there is a statistically significant increase along the coast for arsenic, magnesium and sodium, showing the bioaccumulation capacity of sargassum. Finally, it was shown that the protein and fat content is within the ranges determined for these elements for proteins and slightly lower for lipids. The results of this research confirm the need to maintain constant monitoring of the composition of sargassum to decide on its possible applications.

#### **CRediT** authorship contribution statement

RELG: Methodology, Investigation, Formal analysis, Writing – original draft, Writing – review & editing, Visualization; AMG: Resources, Investigation, Formal analysis, Writing –original draft, Writing – review & editing; BG: Investigation, Formal analysis, Writing –original draft, Writing – review & editing; YGH: Conceptualization, Formal analysis, Writing – review & editing; UJJH: Conceptualization, Methodology, Investigation, Formal analysis, Project Management, Resources, Writing – review & editing, Funding acquisition.

#### **Declaration of competing interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

#### Data availability

All data are available under request. Some information can be found as supporting information.

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