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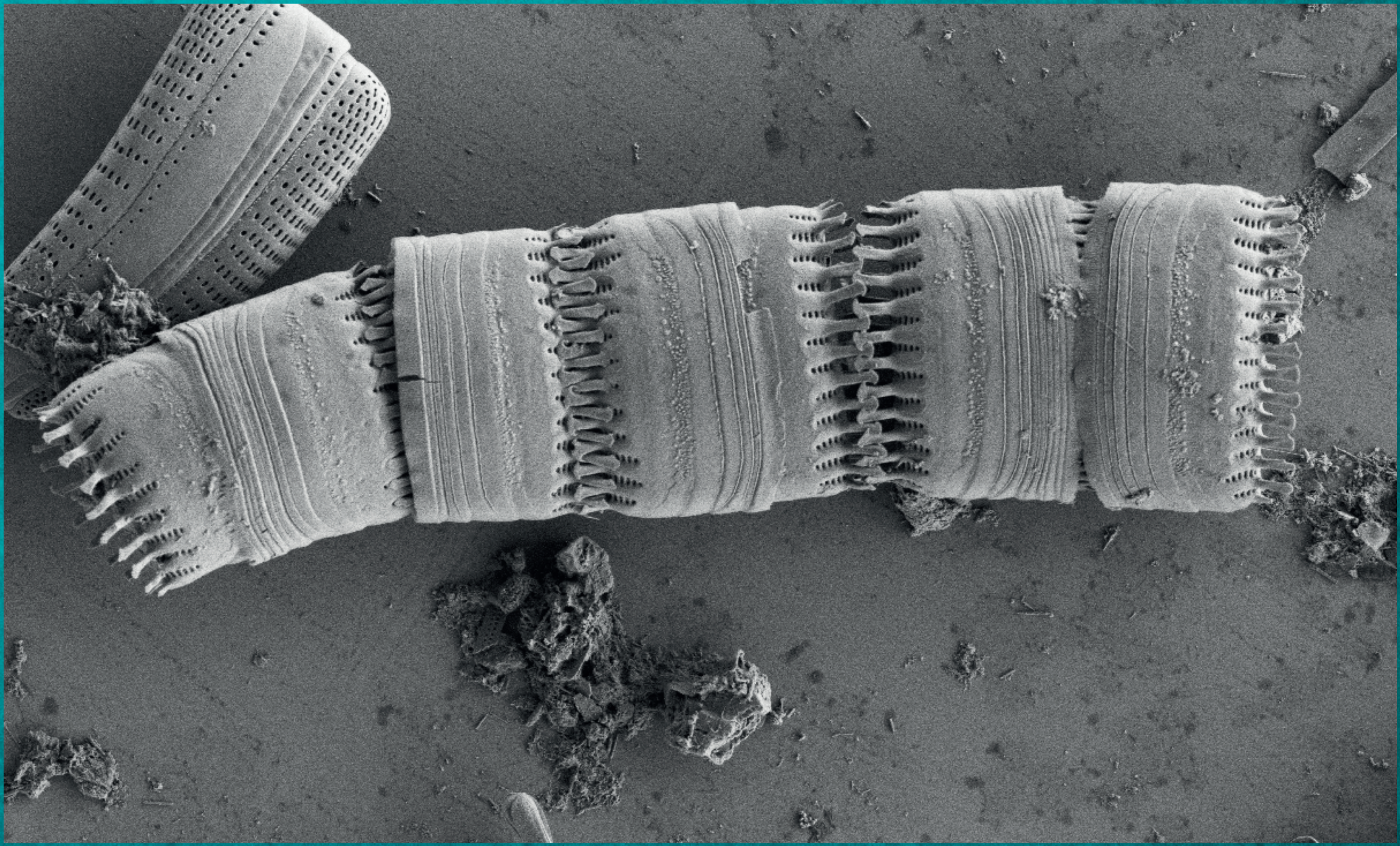

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Sargassum spp. (Phylum: Ochrophyta, Class: Phaeophyceae): Elemental analysis and spatial distribution approximation

Sargassum spp. (Phylum: Ochrophyta, Clase: Phaeophyceae): Análisis elemental y una aproximación a la distribución espacial

María Teresa de Jesús Rodríguez Salazar[1*], José Luz González Chávez[1], Caterin Gutiérrez Sánchez[2], Analaura Skladal Méndez[2], Ariana Janai Morales Velázquez[2], Esperanza Elizabeth Mendoza Solís[2], Rafael Ibarra Contreras[2], Norma Ruth López Santiago[1]

[1]Departamento de Química Analítica, Facultad de Química, Universidad Nacional Autónoma de México (UNAM), Cd. Universitaria, CDMX, México, CP 04510.
[2]Facultad de Química, UNAM.
[*]Email: mtjrs@quimica.unam.mx/mtjrs.papime2020@gmail.com

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ABSTRACT

This review for the 1984-2023 period, includes a sampling sites information employing the Google Earth platform: https://earth.google.com/earth/d/1Qd72z9YXRpNVqmv5jqqJQftJlfS4JLgS?usp=sharing. The seaweed species mainly analyzed were: *Sargassum polycistum*, *S. wightii*, *S. fluitans*, *S. natans,* and *S. muticum*. The most common chemical analytes determined were: Cu, Mn, Zn (micromineral), Ca, K, Fe, Mg, Na, P (macromineral), As, Cd, Cr, Ni, Pb (PTE), C, H, N, S, O (organic elemental analysis). There were a few isotopic data for ^{210}Po and ^{210}Pb (radioactive) and ^{13}C and ^{15}N (light stable). The contamination risk evaluation was preliminary estimated through the indexes *CF*, C_d, *PLI*, E_r^i, *and PERI* using As, Pb, Cd, and Zn global reported concentration data for Mexico´s sampling sites and guideline limits available. In Europe there is regulation for macroalgae but not yet in Mexico. The preliminary indexes values obtained are higher considering the European Regulation is more severe than the Mexican Standards not specific to the use of biomass (NOM 187, NOM 242, and NOM 247). Thus, the analyzed *Sargassum* spp. seaweed could be classified as “high” risk for As and Cd content, and “moderate” for Pb and Zn.

Keywords: Chemical composition, contamination risk evaluation, Sargassum, spatial distribution.

RESUMEN

El presente trabajo de revisión para el período 1984-2023, incluye la información de los sitios de muestreo utilizando la plataforma Google Earth: https://earth.google.com/earth/d/1Qd72z9YXRpNVqmv5jqqJQftJlfS4JLgS?usp=sharing. Las especies de macroalgas más analizadas son: *Sargassum polycistum*, *S. wightii*, *S. fluitans*, *S. natans* y *S. muticum*. Los analitos mayoritariamente determinados son: Cu, Mn, Zn (microminerales), Ca, K, Fe, Mg, Na, P (macrominerales), As, Cd, Cr, Ni, Pb (PTE), C, H, N, S, O (análisis elemental orgánico). Se reportan datos isotópicos: ^{210}Po y ^{210}Pb (radiactivos) y ^{13}C y ^{15}N (ligeramente estables). La evaluación del riesgo de contaminación se estimó preliminarmente a través de los índices *CF*, C_d, *PLI*, E_r^i y *PERI* utilizando datos globales reportados en sitios de muestreo de México para el contenido de As, Pb, Cd y Zn, y los valores permisibles disponibles. En Europa sí existe regulación para el consumo de macroalgas, pero no en México. Los valores preliminares obtenidos para los índices son mayores para la Regulación Europea que las Normas Mexicanas existentes NOM 187, NOM 242 y NOM 247 (no específicas para el uso de la biomasa). Las especies de la macroalga *Sargassum* spp. analizadas podrían clasificarse como riesgo "alto" para los contenidos de metaloides como As y Cd, y "moderado" para los metales Pb y Zn.

Palabras clave: Composición química, evaluación del riesgo de contaminación, sargazo, distribución espacial.

Abstract Abbreviations: *S.*: *Sargassum; CF*: Contamination Factor; E_r^i: Individual Potential Risk Factor; *NOM 187*: NOM-187-SSA1/SCFI-2002 Standard; *PTE*: Potentially Toxic Elements; C_d: Degree of Contamination; *PERI:* Total Ecological Risk Index; *PLI:* Pollution Load Index; *NOM 242*: NOM 242-SSA1-2009 Standard; *NOM 247*: NOM-247-SSA1-2008 Standard.

INTRODUCTION

The origin and location of the tropical floating *Sargassum* spp. are related to the North-Equatorial Recirculation Region (NEER), the Sargasso Sea (place with greatest abundance of pelagic species), and the main currents in the Central Atlantic (Baker *et al*. 2018; DCNA 2019; Fernández *et al*. 2017; Hinds *et al*. 2016). The Langmuir Ocean Circulations allow the grouping of the macroalgae mats (Baker *et al*. 2018; Barstow 1983) as the case of the macroalgae arrivals to the Mexican Caribbean coasts. INECC (2021) reported the abundance of the *Sargassum natans* and *S. fluitans* at the coast, as an environmental problem (related to tourism, economic and health sector) due to a combination of eutrophication from human pollution and oceanographic conditions changes (as temperature).

Sargassum spp. (brown macroalgae) is classified according to Puspita (2017) in Phylum: Ochrophyta, Class: Phaeophyceae, Order: Fucales, Family: Sargassaceae. Hinds *et al*. (2016) highlight the ecological value of floating seaweed as a habitat, shelter, and food for many marine species. Fleurence & Levine (2016) report the potential applications of documented marine macroalgae in the world. For example, a) Years 13000 B.C. for nutrition and health in Chile country, b) Years 0 – 300 A.C. for medicinal use in Greece, as fertilizer in Rome, and as food supplement in Japan. Other reported applications point towards pharmacology and cosmetics of various bioactive compounds of *Sargassum* spp. (Hinds *et al*. 2016; Puspita 2017), biogas (Hernández López 2014; Hinds *et al*. 2016), hydrocarbon pollution bioindicators of petroleum (Lourenço *et al*. 2019), heavy metal biosorbent in contaminated water (DCNA 2019; Hinds *et al*. 2016), pest control, feed supplements, fish and livestock feed, agglomerated material for construction (Hinds *et al*. 2016). Milledge & Harvey (2016) highlight the therapeutic use of bioactive compounds in diabetes, cancer, AIDS, vascular diseases, antioxidants, and anti-inflammatory treatments. Rushdi *et al*. (2020) look over the reported bioactive compounds and the biological activities for clinical applications of *Sargassum* species from the Red Sea.

Diverse methods and techniques have been used to analyze the chemical composition of *Sargassum* samples collected in multiple geographical locations, including volumetry, colorimetry, potentiometry, gravimetry, atomic and molecular spectrometry, and chromatography (Addico & deGraft-Johnson 2016; Baker *et al*. 2018; Fernández *et al*. 2017; Hernández López 2014; Lourenço 2019; Puspita 2017; Rohani-Ghadlkolact & Abdulalian 2012; Solarin *et al*. 2014). Several investigations (Addico & deGraft-Johnson 2016; Fernández *et al*. 2017; Hernández López 2014; Lourenço 2019; Milledge & Harvey 2016; Puspita 2017; Rohani Ghadlkolact & Abdulalian 2012; Solarin *et al*. 2014) provide data on the chemical composition determined: a) As, Cd, Cu, Cu, Mo, Ni, Pb, Se, Zn, Be, Cr, Co, Hg, Na, K, Mg, Ca, Ag, Al. B, Bi, Cs, Fe, Ga, Ge, In, Cl, Mn, P, B; b) Rare earths: Ce, Dy, Er, Eu, Gd, Ho, La, Lu, Nd, Pr, Sc, Sm, Tb, Tm, Y, Yb; c) Nitrate and phosphate anions; d) C, H, O, N, S; e) Proteins, lipids, polyphenols, carbohydrates, pigments, polycyclic aliphatic and aromatic hydrocarbons; f) Percentage (%) of Organic matter, and total ash. Solarin *et al*. (2014) mention that AOAC (Association of Official Analytical Chemists) methods to analyze the samples were used.

This paper aims to present a map review with the chemical composition (elements, isotopic analysis, not speciation studies) reported in *Sargassum* spp. samples, and data analysis of recent years (2019-

2023), specifying the information reported from studies in Mexico (1995-2022). Also, the global data was evaluated concerning to As, Cd, Pb, and Zn guideline limits from regulation available, using the preliminary estimation of the indexes Metal Pollution Index (*MPI*), Contamination Factor (CF), Degree of Contamination (C_d), Pollution Load Index (*PLI*), Individual Potential Risk Factor (E_r^i) and Total Ecological Risk Index (*ERI* or *PERI*). This document attempts to contribute as a tool for the integral management (collection, analysis, application, and disposal) of *Sargassum* seaweed as a raw material, knowing the main inorganic content data reported.

METHODOLOGY

The specialized documentary research was carried out by using the www.bidi.unam.mx platform, the National Autonomous University of Mexico Digital Library (DGB 2023), getting eighty-six references covering the period 1984-2023, highlighting: a) Ten articles from the *Journal of Applied Phycology*, b) Six publications from the *Science of the Total Environment Journal*, c) Four papers from the *Algal Research Journal* and the same founds from *Marine Pollution Bulletin*. The main database was ScienceDirect, except for the *Journal of Applied Phycology*. Three doctoral and four master's theses were also recopilated.

RESULTS AND DISCUSSION

Sargassum species and mostly elemental analytes determined.

During 2020 and 2021 an Excel database (two files) that can be seen in the AMyD (Rodríguez Salazar *et al*. 2023; SPI 2023) free website from Facultad de Química, UNAM was elaborated (https://amyd.quimica.unam.mx/course/view.php?id=662&section=5). Figure 1a shows that Asia leads the research about *Sargassum* spp. marine macroalgae, with 45% of the chemical composition references found; the second place corresponds to 27% for America; next, 17% for Europe; and, finally, 11% corresponding to Africa. The *Sargassum* spp. species reported are shown in Figure 1b. The most studied corresponds to *S. fluitans* and *S. natans* in America, *S. muticum* in Europe, *S. polycistum* and *S. wightii* in Asia, and *S. elegans* in Africa. There was also found research in Africa for the *Sargassum* spp. composed of *S. fluitans* and *S. natans*.

With the above database information mentioned, and updated references during 1984-2023 period, there were mapped (Figure 2a) the sampling sites employing the platform Google Earth Version 9.185.0.0 (Google Earth 2024), with the next electronic link allowing the visualization https://earth.google.com/earth/d/1Qd72z9YXRpN-Vqmv5jqqJQftJlfS4JLgS?usp=sharing (free website). Figure 2b displays the content registered for each point mapped: a) Sampling site (the cited references contain the precise geographical coordinates from the sampling location), b) Analytes, c) DOI (Digital Object Identifier); and d) Certified Reference Material (CRM), if it was used for the analytical methodology as quality assurance.

In Figure 3a, it is observed that most determined analytes by the called group are:

a) Organic elemental analysis: C, H, N, S, O.
b) Potentially toxic elements (PTE): As, Cd, Cr, Ni, Pb.
c) Macromineral elements: Ca, K, Fe, Mg, Na, P.
d) Micromineral elements: Cu, Mn, Zn.
e) Special isotopic analysis: ^{210}Po and ^{210}Pb (radioactive) in *Sargassum boveanum* and *Sargassum oligocystum* from Kuwait (Uddin *et al*. 2019), ^{13}C and ^{15}N (light stable isotopes) in *S. fluitans* and *S. natans* from Mexico (Martínez-Rodríguez 2020; Vázquez-Delfín *et al*. 2021). Figure 3b displays the preliminary concentration average and maximum concentration levels found. According to the average concentration values (mg/kg) the lowest are for Cd, Ni, and Pb; while the highest are for K, Na, and C.

Table 1 specifies information about recent research published corresponding to the 2019-2023 period, which contains the *Sargassum* species analyzed, the sampling site, an analytical technique for the elements determined, and the main findings of the reported results for the authors of the reference cited. The highlighted location of several *Sargassum* species can be observed:

a) Africa: *Sargassum obovatum*, *S.* cf. *portierianum*, *S. robillard*, *S. pfeifferae*, *S. elegans*, *S. vulgare*, *S. cinereum* (Bekah *et al*. 2023; Madkour *et al.* 2019; Magura *et al*. 2019; Mahmoud *et al*. 2019).
b) Iberian Peninsula: *S. muticum* (Álvarez-Viñas *et al*. 2019; Rodrigues *et al*. 2019; Torres *et al*. 2021).
c) Caribbean: *S. fluitans*, *S. natans*, *S. polyceratium* (Alzate-Gaviria *et al*. 2021; Davis *et al*. 2021; Gobert *et al*. 2022; Martínez-Rodríguez 2020; Ortega-Flores *et al*. 2022; Ramírez-Cruz 2021; Rodríguez-Martínez *et al*. 2020; Thompson *et al*. 2020; Vázquez-Delfín *et al*. 2021) and also *S. vulgare* (Martínez-Rodríguez 2020).
d) Asia: *S. horneri* (Huang et al. 2022; Tamura *et al*. 2022), *S. fusiforme* (Huang *et al*. 2022; Su *et al*. 2021), *S. wightii* (Ajith *et al*. 2019; Thadhani *et al*. 2019; Yoganandham *et al*. 2019), *S. polycystum* (Corales-Ultra *et al.* 2019; Sumandiarsa *et al*. 2020; Thadhani *et al*. 2019), *S. ilicifolium* (Kordjazi *et al*. 2019; Siddique *et al*. 2022).

To determine the analyte´s concentration level, the most employed analytical techniques, according to

Table 1 were:

a) Organic Elemental Analyzer (OEA) by combustion using infrared detectors and thermal conductivity for quantification of C, H, N, S (and oxygen by difference).

b) Inductively Coupled Plasma- Mass Spectrometry (ICP-MS) for PTE.

c) Atomic Absorption Spectrometry (AAS), Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-AES) for macromineral and micromineral elements, and also ICP-MS for elements at trace concentration levels.

d) Specific volumetric method to determine nitrogen (Kjeldahl) and Ultraviolet-Visible Spectrophotometry for phosphorous.

e) Special isotopic analysis as Ratio Mass Spectrometry (IRMS) to determine ^{13}C and ^{15}N, and Alpha Spectrometry (AS) for ^{210}Po and ^{210}Pb (radioactive).

f) Anodic Stripping Voltammetry (ASV) to determine arsenic.

Among the overall findings obtained through the *Sargassum* spp. sample analysis were as a natural source of bioactive compounds such as vitamins (A, C, B complex), polyphenols, polysaccharides (fucoidan, alginic acid), functional metabolites (alanine, guanidinoacetate, ethylene glycol), fatty acids (palmitic acid, oleic acid), phytosterols (b-sitosterol, fucosterol), phaeophytin and minerals nutrients as Mg, Ca, K, P, N, Fe, Mn, Zn (Álvarez-Viñas *et al*. 2019; Alzate-Gaviria *et al*. 2021; Bekah *et al*. 2023; Choi *et al*. 2020; Circuncisão *et al*. 2018; Dewi *et al*. 2019; Kordjazi *et al*. 2019; Magura *et al*. 2019; Rodrigues *et al*. 2019; Sumandiarsa *et al*. 2020; Tamura *et al*. 2022; Torres *et al*. 2021; Thadhani *et al*. 2019; Yoganandham *et al*. 2019).

According to the valuable chemical composition of the seaweed, the potential applications studied reported were: a) Environmental: production of bioethanol and bioplastics, carbon capture, bioaccumulation of heavy metals (Ni, Cu, Pb), arsenic remover, pollution biomonitoring, bioremediation for polluted water and soil, b) Biomedical: antioxidant activity and action against cancer, antidiabetic treatment, nutraceutical products, antihypertensive properties, c) Agro-Food Industry: alginate extract, fertilizer, natural plant growth stimulant, ruminant feed (Ajith *et al*. 2019; Álvarez-Viñas *et al*. 2019; Choi *et al*. 2020; Corales-Ultra *et al*. 2019; Davis *et al*. 2021; Delgadillo Mendoza 2022; Dewi *et al*. 2019; Gouvêa *et al*. 2020; Gutiérrez Sánchez 2023; Huang *et al*. 2022; Kordjazi *et al*. 2019; Leyvas Acosta *et al*. 2023; Madkour *et al*. 2019; Mahmoud *et al*. 2019; Rakib *et al*. 2021; Ramírez-Cruz 2021; Sumandiarsa *et al*. 2020; Tamura *et al*. 2022; Thadhani *et al*. 2019; Torres *et al*. 2021; Yoganandham *et al*. 2019).

This research paper details information reported for Mexico's *Sargassum* spp. sampling sites in Table 2. The data registered are *Sargassum* species analyzed, sampling site, analytes (including isotopic data), and concentration level (keeping the unities reported for the authors). The sampling sites correspond to Mexico´s Caribbean and Gulf and Baja California Peninsula. The main species reported are *S. fluitans* and *S. natans* for the Caribbean and *S. sinicola* for the BC Peninsula. On the whole, it can be observed the concentration level for the elements determined in the real samples (Fig. 3b): a) Trace: Al, As, Cd, Cu, Mn, Ni, Pb, Zn, b) Minor: Fe, N, P, and c) Major: C, Ca, K, Mg, Na. About the isotopes ^{13}C and ^{15}N, the numerical value indicates an enrichment or depletion of the heavier isotope (^{13}C, ^{15}N) relative to the lighter (^{12}C, ^{14}N). Through the isotopic characterization, the next findings were observed: the pelagic *Sargassum* species acquire carbon as HCO_3^- and are associated with the biological fixation of atmospheric nitrogen (Martínez Rodríguez 2020).

Preliminary estimation of the contamination risk evaluation, through available regulation.

Related to the previous overall findings paragraph, Table 3 presents Mexican regulation data available that may be useful to take advantage of the *Sargassum* spp. as a natural resource and not as a growing pollution problem. The allowance concentration levels for As, Ba, Be, Ca, Cd, Cr, Cu, F, Fe, Hg, I, Mg, Mn, N, Ni, P, Pb, Se, Sn, Tl, V, Zn, and also Ag (not analyzed yet in the reported studies of this review) are recommended by the Official Mexican Standards (NOM). The NOM-051-SCFI/SSA1-2010 (SE & SSA 2010), NOM-247-SSA1-2008 (SSA 2009), NOM 242-SSA1-2009 (SSA 2010), and NOM-187-SSA1/SCFI-2002 (SSA & SE 2003) are employed for Foodstuff applications. While for soil use the norms are NOM-021-RECNAT-2000 (SEMARNAT 2002), NOM-147-SEMARNAT/SSA1-2004 (SEMARNAT & SS 2007), and NOM-004-SEMARNAT-2002 (SEMARNAT 2003). The NOM-127-SSA1-2021 (SS 2022) supplies the radioactivity specifications for drinking water (alpha 0.5 Bq/L, beta 1 Bq/L).

Data from Table 2, were evaluated for possible pollution risks and contamination degree, using different indexes taking into account as background concentration values the guideline limits established by the regulation available (in mg/kg) for PTE (As, Cd, Pb) by the European Regulation shown in Table 4 (CEVA 2020; Timoner *et al*. 2020) and by the mentioned above Mexican Standards (NOM 242 for fishery products, NOM 187 for corn products and NOM 247 for other cereal products). The essential element Zn is regulated by NOM 247. The European Commission is applied to edible algae consumed in the continent for example, the called Hiziki (*S. fusiforme*).

The *MPI* (Rahhou *et al*. 2023; Rajaram *et al*. 2020; Rakib *et al*. 2021;) to evaluate the pollution between species, was applied only as an example for *S. fluitans* y *S. natans* data from Ortega Flores *et al*. (2022) and Vázquez Delfín *et al*. (2021), because both studies determined the concentration for the five elements mentioned above. The MPI calculated using the maximum concentration values were: 1) Based on PTE and Zn: 3.7 and 30.5 for VD and OF, respectively, and 2) Based only on PTE: 2.9 and 23.9 (VD and OF, respectively). The data from Ortega Flores *et al*. (2022) represents a major source for elements evaluated.

Using the average values, the indexes *CF* and *PLI* (reported by Shams El-Din *et al*. 2014; Tyovenda *et al*. 2019) were calculated for seaweeds as bioindicators due to potential bioaccumulation for heavy metals (and also Zheng *et al*. 2023 for mangrove ecosystems). Other indexes were calculated in analogy for the *Sargassum spp.* samples: C_d, E_r^i, *and PERI* (described by Alkan *et al*. 2020; Hankanson 1980; Mahammad Diganta *et al*. 2023), using the average and maximum concentration data reported.

The results for *CF* and E_r^i calculated are presented in Figure 4a and 4b, where it can be seen that the European Regulation and NOM 187 establish more severe guideline limits than the other Mexican standards mentioned previously. The PTE As and Cd contribute in a high degree to the index value, exceeding the limits. In accordance with indexes scale values (Alkan *et al*. 2020; Karimian *et al*. 2021; Mahammad Diganta *et al*. 2023; Tyovenda *et al*. 2019), the contamination level could be classified as "high" with respect to As and Cd, and "moderated" for Pb and Zn content, using the global data reported for Sargassum spp. macroalgae evaluated from Table 3. But if NOM 242 and 247 are applied as criteria for Pb, then it surpasses the guideline values. The micronutrient Zn does not represent ecological risk according to the NOM 247 standard, because the E_r^i has the value <40 (Karimian *et al.* 2021; Mahammad Diganta *et al.* 2023).

Figures 5a, 5b, and 5c displayed the C_d, *PERI,* and *PLI*, considering the global data reported for As, Cd, Pb, and Zn. The graphs show the higher index values calculated using maximum concentration values found than the index values obtained with the average concentrations. In general, the C_d and *PERI* diagrams show the next descendent order in calculated indexes values with respect to the standards criteria for guideline limits: European Regulation > NOM 187 > NOM 247 > NOM 242 (Fishery Products). While for PLI values the order was: NOM 247 > NOM 187 > European Regulation > NOM 242. The indexes scale values available (Alkan *et al*. 2020; Håkanson 1980; Karimian *et al*. 2021; Mahammad *et al*. 2023; Shams El-Din 2014; Zheng 2023) were used also to evaluate the possible contamination risk from the content of PTE (As, Cd, Pb) and Zn, identifying the "moderate" classification of the organisms according to the less severe NOM 242 for Fishery products, and "high" contamination risk applying the European Regulation (macroalgae consumption) and Mexican Standards NOM 187 and 247 (Corn and Cereal Products), especially concern to As and Cd. This paper advises about the contamination risk, instead of polluted in agreement with the definition of both terms given by Chapman (2007), considering that the index values calculation is based on concentration levels and their dispersion, the elements included, and the guideline limits criteria.

This review also remarks that the data about the analytical quality assurance are not ordinarily reported. Table 5 displays the certified reference material (CRM) for analytical methodology validation applied to the macroalgae analysis reported in this review. The CRM based on seaweed matrix are for elemental analysis: BCR-279 (*Ulva lactuca*, not yet available), NIES-03 (microalgae *Chlorella* spp.), ERM-CD200 (Bladderwrack seaweed, *Fucus vesiculosus*), GBW10023 (Laver algae) NMIJ CRM 7405-b (*S. fusiforme*, also for arsenic speciation), NIES No. 9 (*Sargassum fulvellum*). The CRM IAEA 446 (Baltic Sea Seaweed) was applied for radionuclides analytical methodology. For isotopic analysis, the sample matrix types are, for instance, caffeine (IAEA-600, USGS-61) and human hair (USGS-42, USGS-43), and there is no reported seaweed matrix yet. This review agrees with the observation about the opportunity area to characterize the *Sargassum* species to know the chemical constituents and its properties associated with benefits that could be obtained from this biomass instead of considering it as a waste. And advice about the requirement to evaluate environmental indexes and establish regulation about the well-known chemical species with human and animal health risks previous to the application of the macroalgae (like a raw material). And also considering the effects of heavy metals in morphology, growth, photosynthesis and metabolic process on seaweeds, a was documented by Chung & Lee (1989).

Complementary environmental findings. The research about *Sargassum* spp. seaweed draws attention to its properties as a call to take advantage of the observed following information:

Inorganic components.

The data obtained by Madkour *et al*. (2019) indicated the accumulation of Fe, Mn, and Zn in the studied macroalgae species; hence, they can be used as a good target for monitoring metal pollution in marine waters.

Corales-Ultra *et al*. (2019) found that *S. polycystum* bioaccumulates heavy metals, following the metal uptake descending order: Ni »Cu > Pb.

Siddique *et al*. (2022) reported overall mean values <1 for Hazard Index (HI) for metals (Pb, Cd, Zn, Cu, Ni, Mn, Cr, Fe) and Hazard Quotient (HQ) for bioaccumulation of carcinogenic elements (Pb, Cd, Cr, Ni).

Low total concentration levels of Cu, Mo, Zn, Mn, and Pb were found by Rodríguez Martínez *et al*. (2020). However high total concentration levels found for As exceeded the allowable limits; and the availability for application as animal fodder and agricultural soil according to European regulations can be restricted.

Thompson *et al*. (2020) reported that high Hg and As content could limit the exploitation of *Sargassum* species as biogas. Through the hydrothermal pretreatment of the biomass, it was possible to reduce the H_2S concentration from 3 to 1% in the biogas obtained.

Differences in arsenic concentration (μg / g as dry weight, dw) levels between the coastal and oceanic area *Sargassum* species collected were observed experimentally by Gobert *et al*. (2022): 30–45 and 120–240. The above is because of the depuration performed by the competitive exchange with high concentrations of terrigenous metals (Al, Fe, Mn) in the coast.

Inorganic arsenic (iAs: As^{III} + As^{V}) analysis was also performed by Huang *et al*. (2022) reported a high iAs concentration of 15.1 to 83.7 mg/kg, especially in *S. hemiphyllum*and *S. henslowianum*. These values exceed the limit for seaweed as additives for infant food in the National Food Safety Standard of Pollutants in China. The authors also evaluate the water extraction efficiency for As species, obtaining values above 60 %.

Rakib *et al*. (2021) evaluate Pb content in *S. oligocystum*, where the concentration level (10.63 mg/kg), exceeds the maximum international guidance level (5 mg/kg) recommended by the French High Council for Public Health and The Center for the Study and Development of Algae (CEVA), and as leafy vegetable according to Food and Agriculture Organization (FAO, 0.3 mg/kg).

Martínez-Rodríguez (2020) evaluates the carbon and nitrogen biological fixation by the macroalgae using C and N isotopes. The numerical values foundwere δ ($^{o}/_{oo}$)^{13}C -18.26 ± 0.40 in *S. fluitans* III and ^{15}N -1.03 ± 0.73 in *S. natans* I.

Radioactive isotopes were found by Uddin *et al*. (2019) for *S. boveanum* and *S. oligocystum*, the Concentration Factor values observed were:

a) ^{210}Po (•10^{4}): 1.05 and 0.85, respectively for both species. These values are higher than the IAEA (International Atomic Energy Agency) recommended value (1 •10^{3}).

b) ^{210}Pb (•10^{3}): 0.8 and 1.54, respectively. Both values are below the ICRP (International Commission on Radiological Protection) recommended value (2 •10^{3}).

Dewi *et al*. (2019) produced an organic liquid fertilizer from *Sargassum* sp., through the addition of a bread starter with *Saccharomyces cerevisiae* microorganism to accelerate the fermentation process (transforming glucose into ethanol, CO_2, and organic acid as pyruvic acid and lactic acid). Some chemical parameters of the product were evaluated: 2.7 % N, pH 5.43, and 94.36 % total solid content. The tempe starter addition with *Rhizopus oligosporus* microorganism gave a high content of micronutrients such as Fe, Mn, and Zn.

Organic components.

Davis *et al*. (2021) found that *S. fluitans* and *S. natans* pelagic morphotypes (I and VIII) are carbohydrate sources for microbial production of ethanol and bioplastics.

Magura *et al*. (2019) reported that *S. elegans* macroalgae contains bioactive compounds: β-sitosterol, fucosterol, and phaeophytin.

Choi *et al*. (2020) performed the identification and quantification of functional metabolites in *S. fulvellum* useful for rumen fermentation, among them: alanine (1.00 ± 0.06 mM/L), guanidoacetate (41.93 ± 3.36 mM/L) and ethylene glycol (8.21 ± 0.69 mM/L). The study also found high content of: a) minerals (Na and Ca) with nutritional value, b) As (122.05 ± 5.69 mg/kg), and c) F (4.37 ± 0.18 mg/kg). The arsenic concentration is within the acceptable limit for ruminants' feed.

Mahmoud *et al*. (2019) applied an extract from *S. vulgare* to red radish plants as a natural plant growth stimulant. The results showed significant improvement in the next parameters: plant length, number of leaves, the diameter of roots, and leaf pigment content (chlorophyll a, b, a + b, and carotenoids). Also, similar effects were observed for increasing the nutritional content: a) Phytochemicals compounds (total phenolics, flavonoids, and anthocyanins), and b) N, P, K, Fe, Zn, and Mn.

Tamura *et al*. (2022) confirmed the arsenic removal and the antihypertensive effect of fermented pretreatment of *S. horneri* with *Lactiplantibacillus pentosus* SN001, enhanced ACE (angiotensin-I-converting enzyme) inhibitory activity. The above was done by analyzing the liver, kidney, and spleen of the spontaneously hypertensive rats (SHR) model.

Rodrigues *et al*. (2019) performed an enzymatic aqueous extraction with Alcalase for *S. muticum* to concentrate macro and microelements, increasing the nutritional values for K and P. Also, it was confirmed the presence of polysaccharides as fucoidans with prebiotic and antidiabetic potential. And no cytotoxicity against normal mammalian cells was observed.

Torres *et al*. (2021) obtained an ultrasound-assisted hydrolysis of crude fucoidan from *S. muticum*, and it was observed cell growth inhibitory activity against human cervical carcinoma cells (HeLa 229).

Álvarez-Viñas *et al*. (2019) as a result of fractionation of fucoidans obtained from *Sargassum* by hydrolysis treatment through a sequence of progressively lowering molecular weight membranes (different Kilodalton values, kDa), observed the next:

c) 10–30 kDa: fraction showed IC_{50} (concentration inhibiting growth by 50 %) 44.4 mg L, against cervix cancer cells (HeLa 229).

d) 50–100, 5–10, and < 5 kDa: fractions found active against ovarian cancer cells (A2780).

There have been carried out estimations of the above-ground biomass (AGB), which represents an ecological practical parameter to study the carbon cycle taking into account the function of the forests in carbon storage and climate change (Li *et al*. 2023). Gouvêa *et al*. (2020) obtained the next information as a combination of predictors to the distribution for Sargassum species (floating and benthic):

a) Iron, temperature, salinity, and phosphate: $305.95 \cdot 10^4$ km^2 Pelagic (floating).

b) Light at the bottom, nitrate, salinity, and temperature: $139.59 \cdot 10^4$ km^2 (benthic).

For both species, it was obtained a total area of $445.54 \cdot 10^4$ km^2, 84.05 Gg/km^2 value for AGB, and predicted 13.1 Pg C (petagrams of carbon per year) value for carbon stock in AGB globally. These authors point out that *"...Sargassum aquaculture, natural stock management, and restoration can represent important allies in the urgent need for CO_2 mitigation..."*.

CONCLUSIONS

A map with the sampling site of reported studies was developed using the Google Earth platform and it could be seen that most of the research has been conducted in Asia, analyzing mainly *S. polycistum* and *S. wightii* species, followed by America with *S. fluitans* and *S. natans* (for example at Mexican Caribbean).

Through the review data on chemical composition (elements and isotopic, not speciation) was observed that most of the determined analytes correspond to Potentially toxic elements (As, Cd, Cr, Ni, Pb), macromineral (major) elements (Ca, K, Mg, Na) and micromineral (trace) elements (Cu, Mn, Zn); from the human health approach. According to the concentration level of the above elements, analytical techniques such as ICP-MS (trace), ICP-AES (minor and major), and the AAS methods (Flame, Graphite Furnace, and Hydride Generation) were employed. Stable and radioactive isotopic analysis were also found to be scarce: a) ^{13}C, ^{15}N in Mexico, and b) ^{210}Po and ^{210}Pb in Kuwait. The appropriate quality assurance approach to worldwide analysis results necessary. The use of available CRM reported for algae matrix: NIES-03, ERM-CD200, GBW10023, NMIJ CRM 7405-b, and NIES No. 9 is recommended.

The preliminary estimation for *CF*, C_d, *PLI*, E_r^i and *PERI* indexes allows to advise about a "moderated" to "high" risk contamination due to the As, Cd, Pb and Zn concentration global data reported for the *Sargassum* spp. samples analyzed at Mexico sites, especially when it follows severe guideline limits as European Regulation or Mexican NOM 187 or NOM 247. Although this is in agreement with that, the biomass is viable as a biomonitoring and bioremediation material.

Nowadays, in Mexico there is no specific regulation for the emerging exploitation of *Sargassum* spp. biomass, although it is suggested to look over the standards catalog to establish its necessity, considering potential applications as: bioremediation for polluted water and soil, human health benefits (products with antioxidant activity) and farming and cattle advantages (natural plant growth stimulant and livestock feed). This paper provides a *Sargassum* spp. biomass composition (elements, isotopic) baseline data to complement adequate management strategies, possible valorization routes, development of regulatory measures, and biomonitoring programs.

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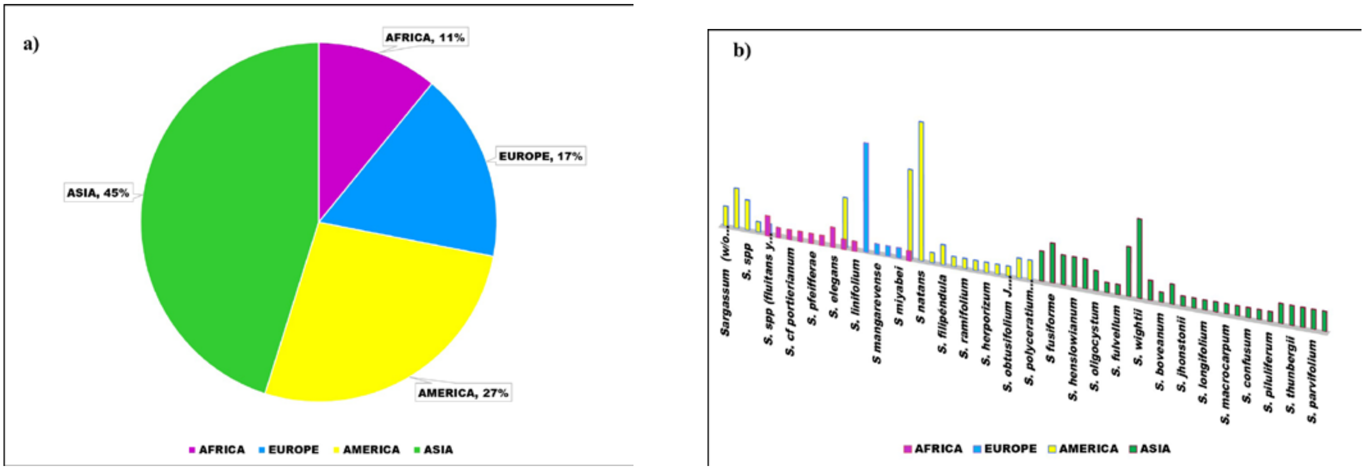

Figure 1. a) Geographic origin from *Sargassum* spp. reported studies found for the documentary research (eighty-two references) and **b)** *Sargassum* species reported from the scientific publications found.

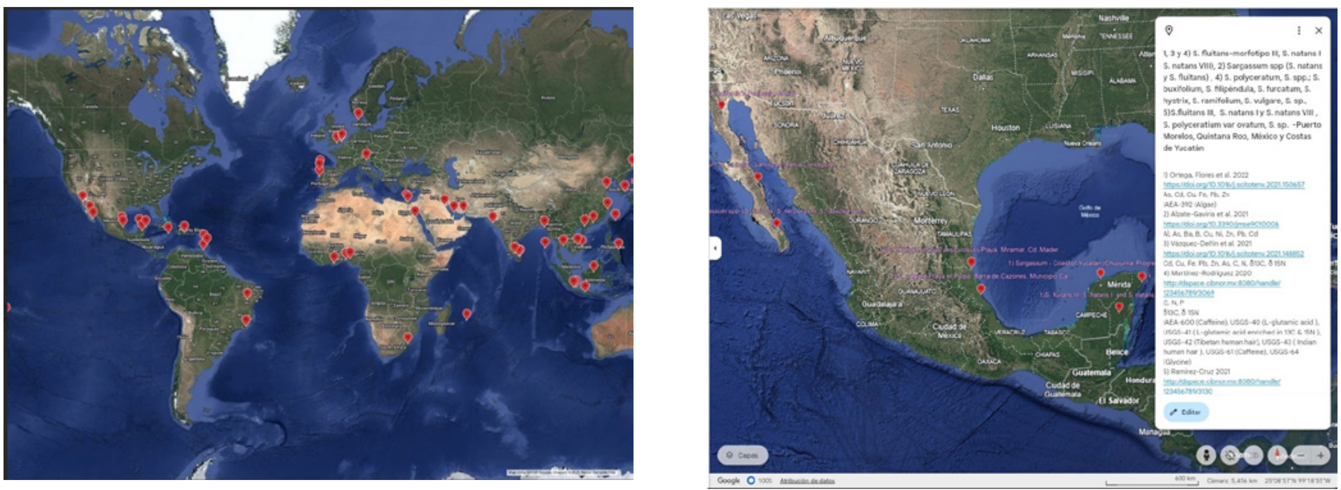

Figure 2. a) Worldwide geographical distribution of *Sargassum* species analyzed (Map created using Google Earth Version 9.185.0.0, Google Earth 2024). Electronic link: https://earth.google.com/earth/d/1Qd72z9YXRpNVqmv5jqqJQftJl-fS4JLgS?usp=sharing, and **b)** Example of the *Sargassum* spp. analysis data registered in the tag for the map sites (Google Earth 2024).

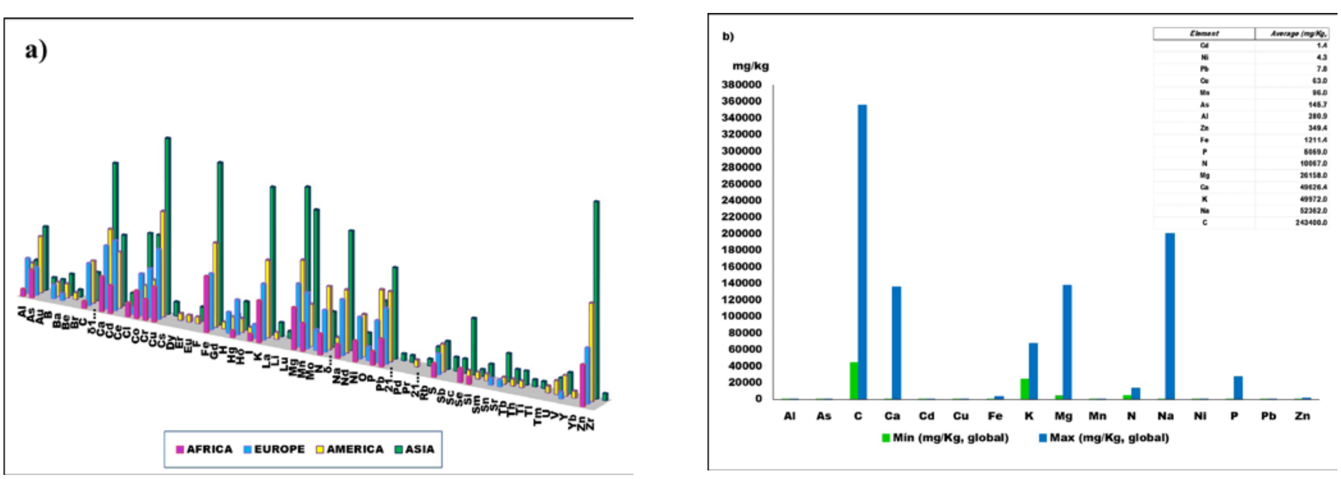

Figure 3. a) Analytes determined in *Sargassum* species from the mapping review publications and **b)** Concentration levels (mg/kg) for elements reported (twice at least) from Mexico´s *Sargassum* spp. data. The average values include the maximum digits reported after the decimal point.

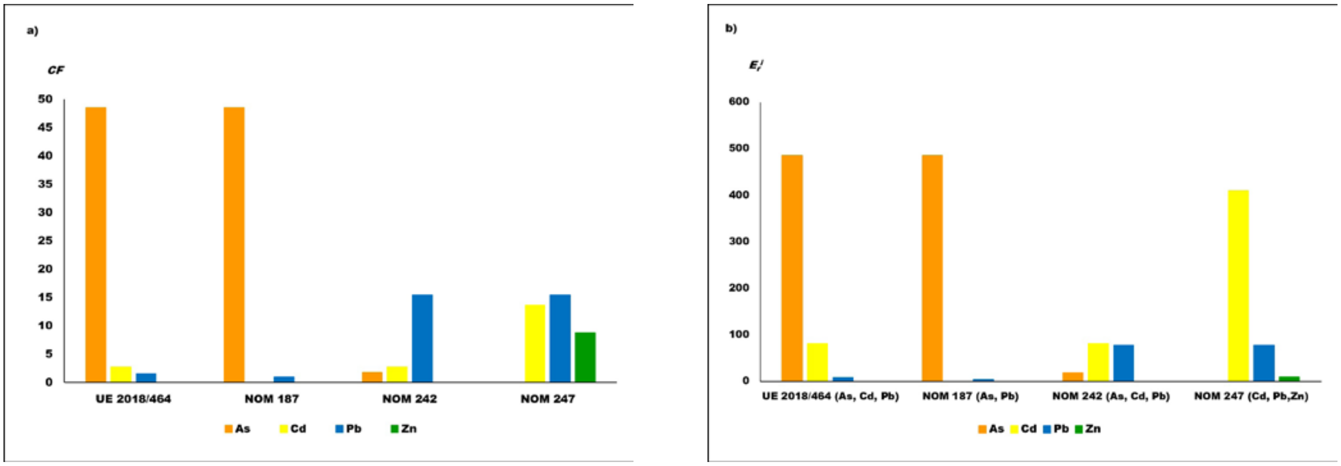

Figure 4. a) Contamination Factor (*CF*) and **b)** Individual Potential Risk Factor (E_r^i) using average values concentration in *Sargassum* spp. samples from Mexico, according to Regulation available for As, Cd, Pb, and Zn.

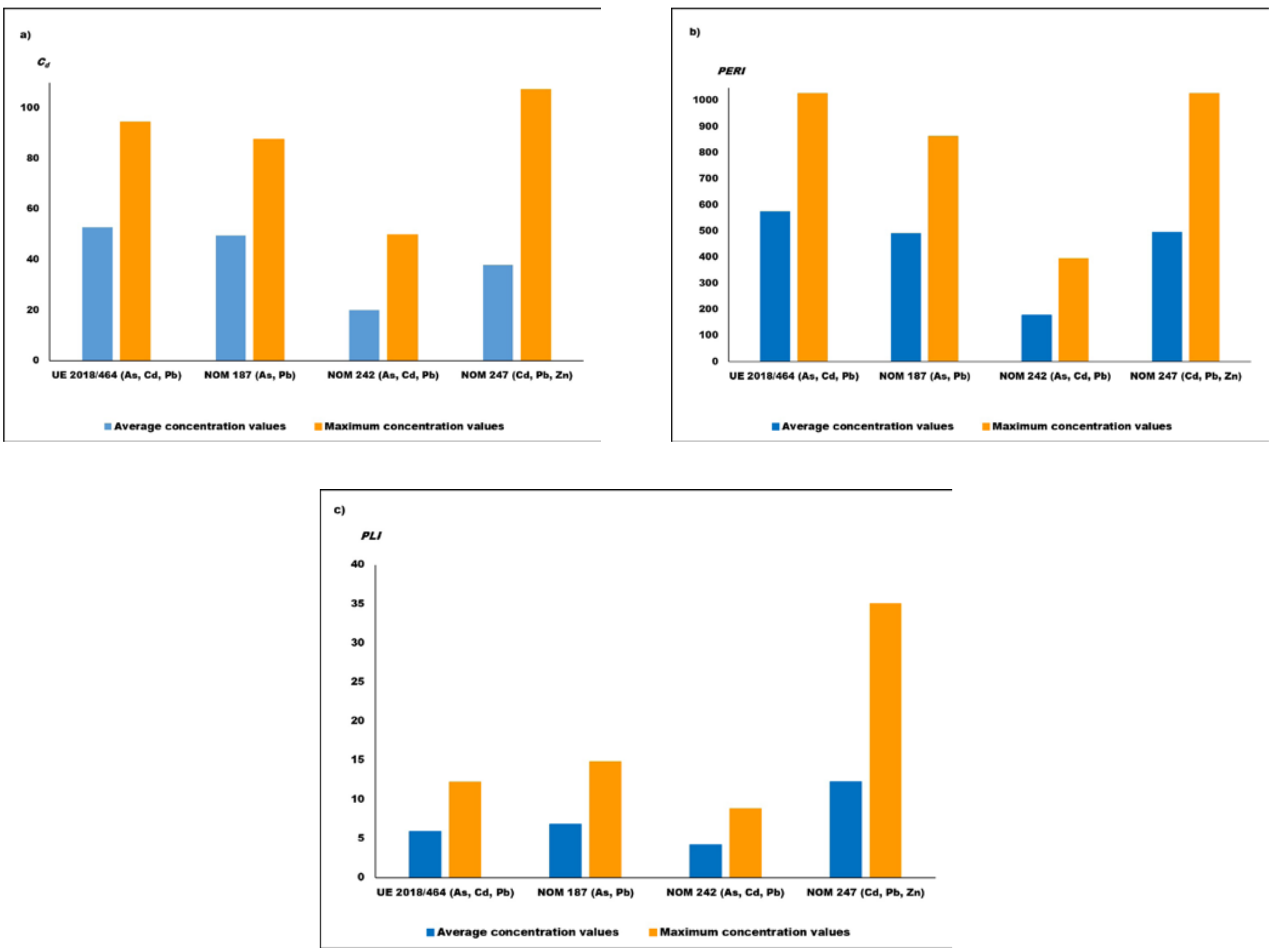

Figure 5. a) Contamination Degree (C_d), **b)** Total Ecological Risk Index (*ERI or PERI*) and **c)** Pollution Load Index (*PLI*) using average and maximum concentration values in *Sargassum* spp. samples from Mexico, according to Regulation available for As, Cd, Pb and Zn.

Table 1. *Sargassum* species worldwide studies with elemental chemical composition reported (2019-2023).

***Sargassum* species**	**Sampling site**	**Analytes**	**Analytical Technique**	**Ref.**
S. fluitans III, *S. natans* I y *S. natans* VIII, *S. polyceratium var ovatum, S. sp.*	Quintana Roo and Yucatán, México	As	HG-AAS [1]	Ramírez Cruz 2021
S. fluitans III, *S. natans* I y VIII, *S. buxifolium, S. filipéndula, S. furcatum, S. hystrix, S. ramifolium, S. vulgare, S. sp., S. polyceratium, S. spp.*		P	HR-ICP- MS [2]	Martínez Rodríguez 2020
		$\delta^{13}C$, $\delta^{15}N$	IRMS [3]	
		C, N	OEA [4]	
S. fluitans, S. natans	Quintana Roo, México	As	GFAAS [5]	Ortega-Flores *et al.* 2022
		Cd, Cu, Fe, Pb, Zn	FAAS [6]	
		Al, As, Ba, B, Cu, Ni, Pb, Zn, Cd	ICP-AES [7]	Alzate Gaviria *et al.* 2021
		As	HG-AAS	Vázquez Delfín *et al.* 2021
		$\delta^{13}C$, $\delta^{15}N$	IRMS	
	Mexican Caribbean	Al, As, Ca, Cl, Cu, Fe, K, Mg, Mn, Mo, P, Pb, Rb, S, Si, Sr, Th, U, V, Zn	EDXRF [10]	Rodríguez Martínez *et al.* 2020
	Port Royal, Jamaica	Na, Mg, Al, K, Ca, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Cd, Ba, Pb, U	ICP-MS	Davis *et al.* 2021
	Consey Bay, Barbados	Na, Mg, Al, P, K, Ca, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Cd,Hg, Pb		Thompson *et al.* 2020
		C, N, O, H, S	OEA	
	Guadaloupe and Martinique Islands, France	As	ASV [11]	Gobert *et al.* 2022
		Al, As, Cd, Co, Cr, Cu, Fe, Mn, Ni, P, Pb, V, Zn	ICP-QMS [12]	
		As, Ca, Cd, Cl, Cu, F, Fe, Mn, P, Pb, Zn	FAAS	
S. fulvellum	Tongyeong, Korea	Co, Cr, Na, Mg, S, Se	ICP-MS[8]	Choi *et al.* 2020
		Hg	FIMS[9]	

Sargassum spp	Yogyakarta, Indonesia	Cu, Pb, Zn	FAAS	Dewi *et al.* 2019
		Cd	GFAAS	
		K	FES [13]	
		P	UV-Vis [14]	
		C	Volumetric Analysis and UV-Vis	
		N	Volumetric Analysis	
S. muticum	Pontevedra, Spain	Na, K	AES [15]	Torres *et al.* 2021
		Ca, Mg, Cu	AAS [16]	
		Cr, Cd, Pb	ICP-MS	
		Ca, K, Na, Mg, Cu, Pb, Hg, Cr, Cd	Not Specified	Álvarez Viñas *et al.* 2019
	Figueira da Foz, Portugal	Mo, B, Zn, P, Cd, Co, Ni, Mn, Fe, Mg, Ca, Cu, Na, Al, K	ICP-AES	Rodrigues *et al.* 2019
		C, H, S	OEA	
		N		
S. polycystum	Sebesi Island, Indonesia	Mn, Ba, Zn, Fe, Cu, Se, Mo	FAAS	Sumandiarsa *et al.* 2020
	Manicani Island, Filipinas	Ni, Cu, Pb	MP-AES [17]	Corales Ultra *et al.* 2019
S. fusiforme	Wenzhou City, China	As, Cr, Cd, Cu, Hg, Pb, Zn	ICP-MS	Su *et al.* 2021
S. boveanum, S. oligocystum	Northern Gulf, Kuwait	^{210}Po, ^{210}Pb	AS [18]	Uddin *et al.* 2019
S. horneri, S. fusiforme, S. hemiphyllum, S. henslowianum	Bohai Bay, Yellow Sea, East China Sea, and South China Sea	As	ICP-MS	Huang *et al.* 2022
S. horneri	Oki Shimane, Japan			Tamura *et al.* 2022
S. wigthii	Tamil Nadu, India	C, H, N, S	OEA	Ajith *et al.* 2019
S. oligocystum	Bay of Bengal, Indian Ocean	As, Pb, Cr, K, Mn, Fe, Co, Cu, Zn, As, Br, Rb, Sr, Zr,	EDXRF	Rakib *et al.* 2021

S. wightii, S. crassifolium, S. polycystum	Mannar, Sri Lanka	K, Na, Ca, Mg, Fe, Mn, Zn, Cu, Ni, Co, Cr, Se, Pd, As, Cd		Thadhani *et al.* 2019
S. wightii, S. swartzii	Mundapam, India	Na, K, Ca, Mg, P, Fe, Cu, Zn, Mn	ICP-MS	Yoganandham *et al.* 2019
S. ilicifolium	Saint´s Martin Island, Bangladesh	Al, As, Be, Cd, Ca, Co, Cr, Cu, Fe, Pb, Li, Mg, Mn, Mo, Ni, Se, Sr, Tl, Ti, V, Zn		Siddique *et al.* 2022
S. ilicifolium, S. angustifolium	Qeshm Island, Persian Gulf, Irán	Ca, Mg, Fe, Mn, Cu, Zn	FAAS	Kordjazi *et al.* 2019
S. boveanum, S. oligocystum	Northern Gulf, Kuwait	^{210}Po, ^{210}Pb	AS [18]	Uddin *et al.* 2019
S. obovatum, S. cf. portierianum, S. robillard, S. pfeifferae	Mauritius Island	As	ICP-MS	Bekah *et al.* 2023
		K, Na, Mg, Si, S, Cl, K, Ca, Al, P, Fe	EDX-SEM [19]	
S. elegans Suhr 1840	Durban, South Africa	As, Ca, Cd, Co, Cr, Cu, Fe, Mg, Mn, Ni, Pb, Se, Zn	ICP-AES	Magura *et al.* 2019
S. vulgare	Monufia Governorate, Egypt	N	Volumetry	Mahmoud *et al.* 2019
		P	UV-Vis	
		K	FES	
		Fe. Mn, Zn	AAS	
S. cinereum C. Agardh	Egypt	Zn, Cd, Cu, Co, Fe, Mn, Ni	FAAS	Madkour *et al.* 2019
Sargassum	Brazilian Coast	C, N	OEA	Gouvêa *et al.* 2020

Note: [1] HG-AAS: Hydride Generation-Atomic Absorption Spectrometry [2] HR-ICP-MS: High Resolution Inductively Coupled Plasma Mass Spectrometry [3] IRMS: Isotope Ratio Mass Spectrometry [4] OEA: Organic Elemental Analyzer [5] GFAAS: Graphite Furnace Atomic Absorption Spectrometry [6] FAAS: Flame Atomic Absorption Spectrometry [7] ICP-AES: Inductively Coupled Plasma-Atomic Emission Spectrometry [8] ICP-MS: Inductively Coupled Plasma -Mass Spectrometry [9] FIMS: Flow Injection Mercury System [10] ED XRF: Energy Dispersive X-Ray Fluorescence Spectrometry [11] ASV: Anodic Stripping Voltammetry [12] ICP-QMS: Inductively Coupled Plasma-Quadrupole Mass Spectrometry [13] FES: Flame Photometry or Flame Emission Spectroscopy [14] UV-Vis: Ultraviolet-Visible Spectrophotometry[15] AES: Atomic Emission Spectrometry [16] AAS: Atomic Absorption Spectrometry [17] MP-AES: Microwave Plasma Atomic Emission Spectrometry [18] AS: Alpha Spectrometry [19] EDX SEM: Scanning Electron Microscopy - Energy Dispersive X-ray spectroscopy [20] FTIR: Fourier Transform Infrared Spectroscopy, XRD: X-Ray Diffraction, SEM: Scanning Electron Microscope

Table 2. Elemental chemical composition reported studies (1995-2022) from *Sargassum* species found in Mexico.

Sampling Site	***Sargassum* species sampled µg/g**	**Concentration values reported[1]** **mg/kg DW**	**ppm**	**mg/100 g**	**mg/g**	**g/100 g**	**%**	**δ Isotope values (o/oo)**		**Ref. 7**
Puerto Morelos, Q. Roo [4], Mexico	*S. fluitans, S.natans*		DW [2] As 9.5-255.2 Cd < 0.02 to 2.6 Cu <0.01 to 2.85 Fe < 0.07 to 78.2 Pb <0.05 to 20.7 Zn < 0.02 to 62.8							Ortega Flores *et al.* 2022
	Sargassum spp (*S. natans, S. fluitans*)		Al 33.81 - 61.88 As 76.49 - 115.66 Ba, 13.73 - 16.7 B 204.36 - 228.83 Cu 3.83 - 4.51 Ni <LOD [3] to 2.5 Zn 30.8 - 80.54 Pb < LOD [3] Cd 0.44 - 0.47							Alzate Gaviria *et al.* 2021
Quintana Roo and Yucatan, Mexico	*S.fluitans* III, *S. natans* I, *S. natans* VIII, *S. polyceratium* var. *ovatum*, *S.* sp.	As 0.7-119.9								Ramírez Cruz 2021
	S. fluitans III, *S. natans* I and VIII, *S. buxifolium*, *S. filipéndula*, *S. furcatum*, *S. hystrix*, *S. ramifolium*, *S. vulgare*, *S.* sp.,, *S. polyceratum*, *S.* spp.							DW [2] C 26.69-35.60 N 0.58-1.36 P 0.013-0.086	$\delta^{13}C$ -19 to-15.24 $\delta^{15}N$ -2.30 to 2.17	Martínez Rodríguez 2020
Mexican Caribbean (Q. Roo: Playa Mirador,-Tulum; Playa Blanca, Akumal; Playa Xcalococo, Playa del Carmen; Puerto Morelos; Playa Coral y Playa Delfines, Cancun)	*S. fluitans, S.natans*		Cd: 0.32-1.36 Cu < 0.20 to 1.09 Fe 24 – 54.6 Pb < 0.20 to 0.29 Zn 3.65 – 7.2 As 29.0 - 65.7					C 26.8 - 33.0 N 0.9 – 1.2	$\delta^{13}C$-17.87to17.09 $\delta^{15}N$-0.55 to 0.19	Vázquez Delfín *et al.* 2021
			DW [2]							
			Fe 277.1							
Yucatan, Mexico (Chuburná, Progreso, Chicxulub)	*Sargasso*		Cu 20.6 Zn 49.7 Co 3.09					DW [2] Mg 0.45		Castellanos Ruelas *et al.* 2010

Mexican Caribbean (Contoy Island, Puerto Morelos, Cozumel, Mahahual, Chinchorro, Xahuayxol, Xcalak)	*S. fluitans* III, *S. natans* I y *S. natans* VIII			Al <LOD [3] to 500 As 24-172 Ca 23, 273-136,146 Cl 747-53101 Cu <LOD [3] to 540 K 1990 - 46002 Mg< LOD [3] to 13662 Mn 40 - 139 P 228 - 401 Rb 30 - 143 Si 447 - 2922 Th 5-23 U 11-48 V <LOD [3] to 13 Zn <LOD [3] to 17			Rodríguez Martínez *et al.* 2020
Barra de Cazones, Veracruz	*S. vulgare*		Cu 3.251 Cd 1.025 Cr 1.4 Ni 6.001 Pb 8.002 Zn 17.604				Uribe Orozco *et al.* 2018
Ciudad Madero, Tamaulipas	*Sargassum*					N 0.46 C 3.85 - 4.42 S 2.02 - 2.26 P 0.545 Na 3.22-3.43 K 3.91-4.09 Ca 6.86 Mg 1.379	Hernández López 2014

BCS [6]			Cu 1 Zn 1600 Fe 3600	Ca 3.21 P 0.1 Na 20.1	K 5.77 Mg 0.9	Carrillo *et al.* 2012
	S spp		Na 2066.8 K 6800.4 Ca 500.7 Mg 701.4 P 44.9 Mn 5.3 Zn 0.98 Fe 41.20 Cu 0.66 Pb 0.2			Casas Valdés *et al.* 2006 [8]
				Mg 138.3 K 24.4 Na 24.5 Ca 32.7 P 27.9		Gojon Baéz *et al.* 1998
Bahía de la Paz, BCS [6]	*S. herporizum, S. sinicola*	Zn 32 - 50 Cu 47 Fe 419 - 458			Ca 6.74 7.28 P 0.5 - 0.53 Na 3.2 - 3.44 K 3.91 - 5.51 Mg 1.39 - 1.4	Carrillo Do-mínguez *et al.* 2002
	S. sinicola		Fe 3600±0.33 Cu 1.00± 0.00 Zn 1600±0.11		Ca 3.21 ±0.54 P 0.011 ±0.00 Mg 0.90±0.09 Na20.07±0.45 K 5.77 ±0.06	Rodríguez Bernal 1995

Note: [1] Concentration units according to the original publication [2] DW: Dry Weight [3] LOD: Limit of Detection [4] Q. Roo: Quintana Roo Mexican State [5] BC: Peninsula of Baja California located in Mexico [6] BCS: Baja California Sur, Mexican State [7] Ref: Bibliography [8] The standard error values are also reported for each analytical parameter

Table 3. Official Mexican Standards (NOM) for elemental concentration regulation values as a possible legal framework for *Sargassum spp.* applications.

Analyte	**Official Mexican Standards** (NOM)						
	Foodstuff				**Soil**		
	051-SCFI/SSA1-2010 (SE & SSA 2010)	**247-SSA1-2008** (SSA 2009)	**242-SSA1-2009** (SSA 2010)	**187-SSA1/SCFI-2002** (SSA & SE 2003)	**021-RECNAT-2000** (SEMARNAT 2002)	**147-SEMARNAT/SSA1-2004** (SEMARNAT & SSA 2007)	**004-SEMARNAT-2002** (SEMARNAT 2003)
	Concentration units						
	Specified for each value		**mg/kg**		**Specified for each value**	**mg/kg**	
As			80	3		22	41
Ba						5400	
Be						150	
Ca	900 mg [1]	900.0 mg [3]					
Cd		0.1 mg/kg	2.0 / 0.5 [5]		0.35 mg/kg	37	39
Cr	22 µg [1]						1200
Cu	650.0 µg [1]	650.0 µg [3]			> 0.2 mg/kg		
F	2.2 mg [1]	2.2 mg [3]		40			
Fe	17 mg [1]	40 mg/kg, 17.0 mg [3]			> 4.5 mg/kg		
Hg						23	17
I	150 µg [1]						
Mg	248 mg [1]	250.0 mg [3]					
Mn					> 1.0		
N					0.10 - 0.15 %		
Ni					50 mg/kg	1600	420
P	664 mg [2]	664.0 mg [3]			< 250 ppm		
Pb		0.5 mg/kg	0.5 / 1 [6]	8	35 mg/kg	400	300
Se	41 µg [1]					390	

Sn			100			
Tl					5.2	
V					78	
Zn	10 mg [1]	40 mg/kg, 10.0 mg [3]		> 1.0 mg/kg		2800
Ag [4]					390	

Note: [1] Reference Nutritional Value (VNR), Daily Suggested Intake (IDS) [2] Reference Nutritional Value (VNR), Daily Recommended Intake (IDR) [3] Daily Recommended Intake (IDR)
[4] Not determined yet in reported *Sargassum* studies found for the present review [5] Values for fishing products: mollusk/others [6] Values for fishing products: fresh / processed

Table 4. European Regulation for heavy metals in edible algae (CEVA 2020, Timoner *et al.* 2020).

Analyte	mg/kg (dry weight)
As	3
Cd	0.5
Hg	0.1
I	2000
Pb	5
Sn	5

Table 5. Certified reference materials applied for the quality assessment of the methodologies for the *Sargassum spp.* samples analysis.

Certified Reference Material	Certified parameters	Reference
IAEA 446 (Baltic Sea Seaweed)	Radionuclides	Uddin *et al.* 2019
NMIJ CRM 7405-b (Hijiki seaweed)	As speciation	Huang *et al.* 2022
IAEA-600 (Caffeine)	Isotopes ($\delta^{13}C$, $\delta^{15}N$)	Martínez Rodríguez 2020
USGS-40 (L-glutamic acid)		
USGS-41 (L-glutamic acid enriched in ^{13}C & ^{15}N)		
USGS-42 (Tibetan human hair)		
USGS-43 (Indian human hair)		
USGS-61 (Caffeine)		
USGS-64 (Glycine)		
BCR-402 (White clover)	Elements	Magura *et al.* 2019
BCR-279 (*Ulva latuca*)		Gobert *et al.* 2022
DORM-4 (Fish protein)		
ERM-BB422 (Fish muscle)		Marzocchi *et al.* 2016
NIES-03 (Chlorella)		Rodrigues *et al.* 2015
1566a (Oyster tissue)		Khristoforova & Kozhenkova 2002
IAEA-392 (Algae material: *Scenedesmus obliquus*)		Ortega Flores *et al.* 2022, Ramírez Cruz 2021
SRM 1570a (Spinach leaves)		Seepersaud *et al.* 2018
Es-2 (Organic rich argillite)		Rodríguez Martínez *et al.* 2020
Es-4 (Dolostone)		
ERM-CD200 (Bladderwrack seaweed, *Fucus vesiculosus*)		Huang *et al.* 2022
GBW10023 (Laver algae)		
NMIJ CRM 7405-b (*S. fusiforme*, Hijiki seaweed)		Gobert *et al.* 2022, Huang *et al.* 2022
BCSS (Marine sediment)		Kaviarasan *et al.* 2018
MAG-1 (Marine mud)		
NIST 1570a (Spinach) NIST 1571 (Orchard leaf) NIES No. 9 (*Sargassum fulvellum*)		Rakib *et al.* 2021 García-Salgado 2013, Hou 1999, Hou & Yan 1998
NIST-1572 (Citrus leaves)		Hou 1999, Hou & Yan 1998