



Addressing the Problem of Harmful Algal Blooms in Latin America and the Caribbean- A Regional Network for Early Warning and Response

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Harmful algal blooms (HABs) constitute a worldwide problem, affecting aquatic ecosystems, public health and local economies. Supported by the International Atomic Energy Agency since 2009, Latin America and the Caribbean (LAC) countries, including Brazil, Chile, Colombia, Costa Rica, Cuba, Dominican Republic, El Salvador, Guatemala, Haiti, Mexico, Nicaragua, Panama, Uruguay and Venezuela, have integrated a regional network for early warning of HABs and biotoxins in seafood. Technical capacities have been developed at regional level to identify toxic species, evaluate biota toxicity, and to perform retrospective analysis of HAB occurrence. This network involves 58% of the coastal LAC countries, two regional reference centers (in El Salvador and Cuba), 14 well equipped institutions, and 177 professionals trained to contribute to the operation of HAB and biotoxin monitoring programs. All countries from the network have reported planktonic and benthic toxic species, and in selected cases, associated with toxin in biota. Dinocyst abundance analysis in ²¹⁰Pb-dated sediment cores have shown that some harmful species have been present in the region for at least 100 years ago, and that both coastal water pollution and climate change are important drivers for HAB occurrence. Efforts must be made to enrich the data base records on HAB events occurred in LAC, better understand key environmental variables that control HABs and expand coverage of HAB monitoring to all coastal countries in LAC to promote sustainable development of the region.

Keywords: HAB, biotoxin, nuclear techniques, laboratory network, IAEA-technical cooperation

INTRODUCTION

Harmful effects of phytoplankton blooms (abundance increment over background levels) may be associated with oxygen depletion, production of phycotoxins, mucilage, reactive oxygen species and polyunsaturated fatty acids, and physical damage to fish gill tissue; high mortalities of marine organisms including fish, marine mammals and sea turtles have been associated with algal blooms (Anderson, 2017). Worldwide, harmful algal blooms (HABs) appear to have increased in frequency, geographic extent and intensity, due to the increase in nutrient discharges to aquatic ecosystems, and climate variability (Heisler et al., 2008) as well as the introduction of exotic species (van den Bergh et al., 2002). Most of the HAB forming species are dinoflagellates, accounting for as much as 100 taxa in the marine environment (Moestrup et al., 2009).

In Latin America and the Caribbean region (LAC), between 1970 and 2007, ~7800 human intoxications, including 119 human fatalities, were mainly associated with Paralytic Shellfish Poisoning (PSP) in the Pacific and Atlantic coasts, and Ciguatera Fish Poisoning (CFP) in the Caribbean zone (Table 1 and Supplementary Table S1). There is no exact measure of the economic impacts of HABs in LAC; however, analysis of specific cases suggest that they are severe. In Mexico, 61 HAB events caused ~2,500 days of sanitary closures between 2003 and 2014 (Comisión Federal de Protección contra Riesgos Sanitarios [COFEPRIS], 2018), affecting local economies due to bans of shellfish harvesting or extracting; and in 2002, in Bahía de Todos Santos (Pacific coast), a bloom of *Ceratium furca* caused a 15-million dollar loss in a mass mortality episode of farmed tuna (Orellana-Cepeda et al., 2004). In Chile, in 2016, a bloom of *Pseudochattonella cf. verruculosa* affected the salmon farming, with losses over 500 million dollars (Clément et al., 2016). Besides the multiple impacts of HABs in LAC, the scarcity of qualified personnel and properly equipped laboratories acted against the establishment of prevention and mitigation measures. Surveillance of toxic HAB occurrence and/or control of biotoxins in seafood has been unequally developed in the region, and only a few countries have regular monitoring programs for local or export trades.

Over the last decade, countries of LAC have developed capacities to better manage HABs, and strengthen cooperation within the region, through the creation of the “Regional Monitoring and Response Network for Marine Resources and Coastal Environments in Latin America and Greater Caribbean.” This effort was supported by the International Atomic Energy Agency (IAEA) Technical Cooperation Program (projects RLA/7/012, RLA/7/014, RLA/7/020 and RLA/7/022). This work describes selected network achievements in terms of capacity building and establishment/improvement of monitoring programs; and presents examples of the information generated, including HAB events, toxic species and biotoxins identification, and historical reconstruction of HABs.

METHODOLOGY

Microalgae Species

Harmful algal bloom monitoring programs within the network followed the methods in Reguera et al. (2011, 2016). Phytoplankton was sampled with nets and Niskin bottles and quantified through the Utermöhl method using inverted microscopes; and benthic dinoflagellates were collected from seagrass and macroalgae, and counted using Sedgewick-Rafter chambers using compound microscopes.

Toxins

Paralytic shellfish toxins (PSTs) and ciguatoxins were analyzed in marine biota using the receptor binding assay (RBA; IAEA, 2013), that quantifies the toxin potency, by determining, through a scintillation counter, the concentration of tritiated toxin standards, which compete with the toxin from sample extracts, for binding to voltage-gated sodium channels in a rat brain membrane preparation (Van Dolah et al., 2012).

Dinocysts

The analysis of dinocysts (cyst/g, dry weight) in ^{210}Pb dated sediment cores was performed following de Vernal et al. (2010). Briefly, sediment samples, added with *Lycopodium* spores as marker for quantitative analysis, are treated with a mixture of strong acids to clean the sample, which is used to prepare permanent slides to be counted in optic microscopes. The relation of environmental records (e.g., temperature or rainfall) and dinocysts abundances was examined using multivariate analysis (Cuellar-Martinez et al., 2018).

RESULTS AND DISCUSSION

The strategy followed by the network included: (i) capacity building (e.g., improvement of analytical infrastructure and formation of human resources through specialized courses and hands-on trainings); and (ii) standardization of sampling and analytical methods for microalgae and toxin quantification, and the retrospective analysis of HAB event occurrence. The most important activities and findings obtained are described below.

Capacity Building

As of 2018, the network involves laboratories in 14 countries (Supplementary Table S2), including Brazil, Chile, Colombia, Costa Rica, Cuba, Dominican Republic, El Salvador, Guatemala, Haiti, Mexico, Nicaragua, Panama, Uruguay and Venezuela, representing 58% of the Member States along the Atlantic and Pacific coasts of Latin America and 14% of the Caribbean coasts. As part of the network activities since 2009, the countries (except Chile and Brazil, who recently joined the network) developed pilot HAB monitoring programs in sampling sites off their coastline (Figure 1); and through 17 training courses, 177 persons were trained on diverse topics, including taxonomy and identification of harmful microalgae, dinocysts and foraminifera; analysis of phycotoxins (including RBA); data analysis (submission of data to the

TABLE 1 | HAB records from countries of the Latin America and Caribbean network in the Harmful Algae Event Database (Harmful Algae Event Database [HAEDAT], 2018).

Country	Years	HAB events	Species frequently reported ¹ (number of HAB events)	Harmful event impact
Brazil*	1990–2016	7	<i>Dinophysis</i> sp. ² (2)	Massive shellfish mortalities and human intoxication.
Chile	1972–2018	39	<i>Alexandrium catenella</i> ³ (17), <i>Chatonella verruculosa</i> (5), <i>Leptocylidrus minimus</i> (4)	113 human intoxication and 15 fatalities, harvest and shellfish farms closures, salmon mortalities.
Colombia	1994–2017	27	Species not identified (8), <i>Synechocystis</i> sp. (6), <i>Mesodinium</i> sp. (3)	Mass fish deaths.
Costa Rica*	1989–2003	3	<i>Pyrodinium bahamense</i> ³ (2)	20 human intoxications and 1 fatality, harvest sites closure.
Cuba	2003–2015	7	<i>Cochlodinium polykrikoides</i> (3)	Not reported.
Dominican Republic*	2015	1	Not identified	Not reported.
El Salvador	2001–2018	13	<i>P. bahamense</i> ³ (4), <i>C. polykrikoides</i> (3)	7 human intoxications and 1 fatality, sea turtle deaths.
Guatemala	1987–2004	7	<i>P. bahamense</i> ³ (4)	26 human fatalities.
Haiti	no data	–	–	–
Mexico*	1979–2014	49	<i>Gymnodinium catenatum</i> ³ (10), CFP (9), <i>Gambierdiscus</i> spp. ⁴ (9), <i>P. bahamense</i> ³ (5)	High toxin concentrations, 25 human intoxications by CFP.
Nicaragua*	2005–2018	4	<i>Phaeocystis</i> sp. (2)	Presence of mucilage, foam, bad smell, 50 human intoxications.
Panama*	2001	1	CFP	Not reported.
Uruguay	1980–2016	53	<i>G. catenatum</i> ³ (13), <i>D. acuminata</i> ² (12), <i>Dinophysis</i> spp. ² (5), <i>D. ovum</i> ² (4), <i>A. tamarensis</i> ³ (3), <i>Akashiwo sanguinea</i> (3), <i>M. rubrum</i> (3)	Shellfish closure and harvesting ban
Venezuela*	1956–2010	12	Diverse species (one per event)	Not reported.

¹ Species in ≥ 3 events, except countries marked with*; Species associated with DSP², PSP³, and CFP⁴.

international Harmful Algae Event Database [HAEDAT], 2018, and retrospective reconstruction of HABs occurrence through the study of dated sediment cores); ocean acidification; and communication skills to disseminate scientific findings to stakeholders (Supplementary Figure S1).

In order to perform the different monitoring activities, prioritized by the countries based on their needs and capacities, each laboratory of the network received specific equipment and consumables (e.g., Utermöhl and Sedgewick-Rafter counting chambers, Niskin bottles, phytoplankton nets, multiparameter probes for water quality and inverted microscopes), some of them inaccessible within the region. Liquid scintillation counters and minor equipment for toxin extraction and quantification using RBA, were provided to laboratories in El Salvador, Nicaragua, Colombia, Cuba and Costa Rica; and alpha and gamma spectrometry capacities were established in Colombia, Cuba, Mexico, Nicaragua and El Salvador, for retrospective studies on HAB forming species in ²¹⁰Pb dated sediment cores.

Pilot Monitoring Programs

Standardization of Methods Used in the Network

The collaborative work among the network laboratories led to production of manuals and guides, that led to significant findings and numerous scientific documents. The “Guide for

designing and implementing a plan to monitor toxin-producing microalgae,” prepared through a collaboration between scientists from the network (and other countries), the International Oceanographic Commission (IOC) and the IAEA, includes standardized sampling and counting procedures (Reguera et al., 2011, in Spanish); and has been expanded to include methods to study benthic HABs (Reguera et al., 2016, in English).

Microalgae Blooms Observations and Impacts in the Network Countries

In the Colombian Caribbean Coast, three blooms caused by *Cochlodinium* spp. and other three by *Mesodinium* cf. *rubrum* occurred in Santa Marta Bay and nearby areas, between 2010 (Malagón and Perdomo, 2013) and 2017; and in Ciénaga Grande de Santa Marta, about nine episodes of mass fish deaths were attributed to low oxygen concentrations associated with HABs (2014–2017; unpublished data). Additionally, in Chengue Bay, Tayrona National Park, 14 potentially harmful benthic dinoflagellates were identified in seagrass meadows, including *Prorocentrum* and *Ostreopsis* as the most frequent genus (Arbeláez-Merizalde et al., 2017). *Prorocentrum lima* was the most representative species on *Thalassia testudinum* and its maximum abundances were associated with salinities close to 35, high temperatures (> 29°C) and low nutrient (N-P)

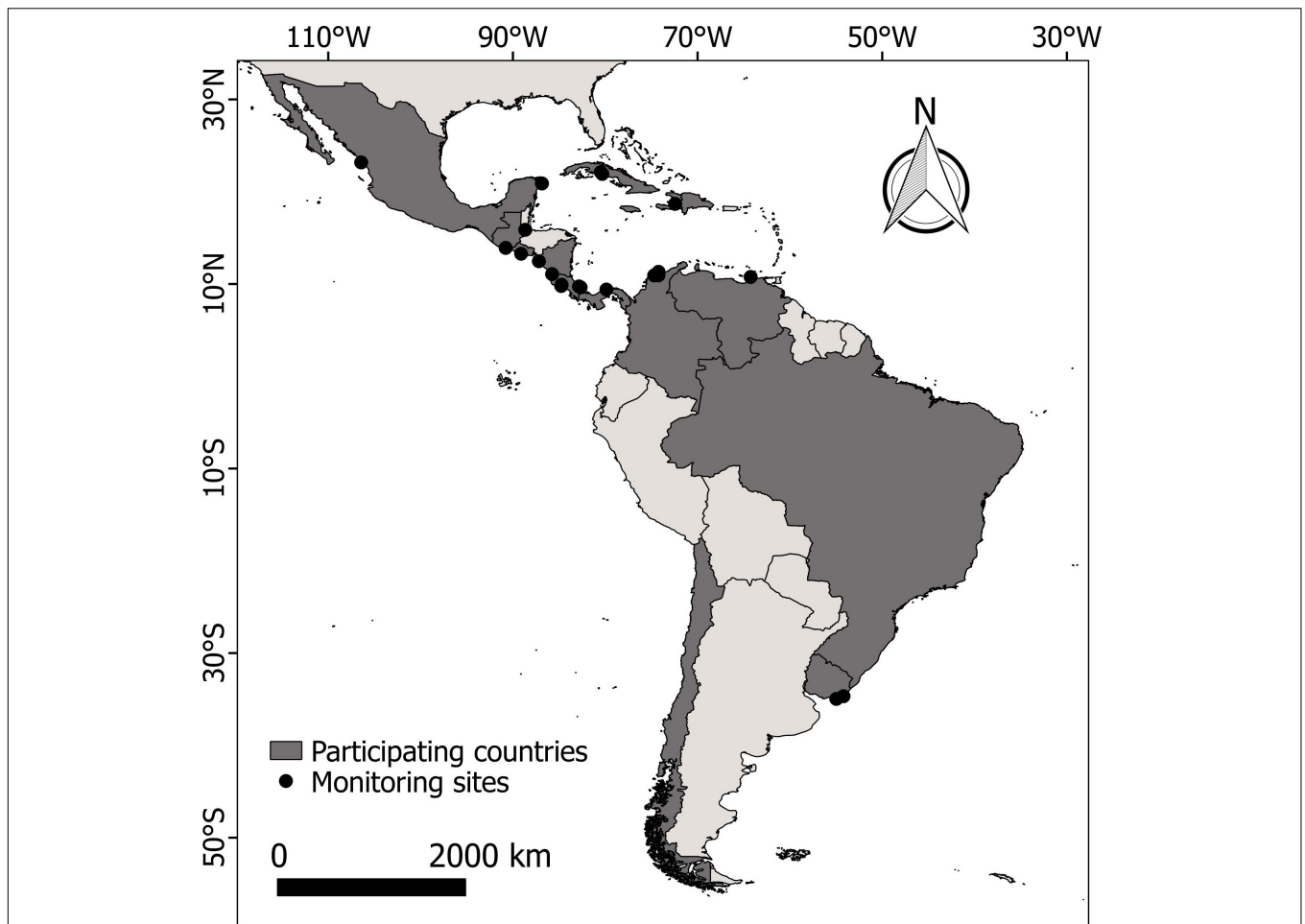


FIGURE 1 | Monitoring sites within the technical cooperation IAEA projects on HABs. Mexico: Mazatlán and Puerto Morelos, Cuba: Cienfuegos Bay and Ensenada de Guajimilco, Dominican Republic: Playa Caleta, Haiti: Port-au-Prince, Guatemala: Puerto Quetzal and Amatique Bay, El Salvador: all the coast, Nicaragua: Port of Corinto and El Ostional Bay, Costa Rica: Cahuita National Park, Gulf of Nicoya, Uvita Island and Puerto Vargas National Park, Panama: Punta Galeta, Colombia: Ciénaga Grande de Santa Marta and Chengue Bay, Venezuela: Cubagua Island, Uruguay: Punta del Este and La Paloma.

concentrations, during an ENSO period (Arbeláez-Merizalde and Mancera-Pineda, 2016).

In Cienfuegos Bay, Cuba and adjacent coasts, ~20 potentially harmful species were identified between 2007 and 2009. *Gymnodinium catenatum*, *Pyrodinium bahamense* and *Dinophysis ovum* were recorded for the first time (Moreira-González et al., 2013). A bloom of *Heterocapsa circularisquama* occurred in 2009 (Moreira-González, 2010); and another of *Vulcanodinium rugosum* in 2015, left 60 cases of skin lesions (Moreira-González et al., 2016a). The HABs in Cienfuegos Bay were associated with the restricted water circulation in small enclosed areas and discharges of urban/industrial effluents (Moreira-González et al., 2014). A bloom of *Cochlodinium polykrikoides* in 2014 in Guanaroca Lagoon was related with extreme weather conditions associated with ENSO (Moreira-González et al., 2016b). Outbreaks by *Phaeocystis* sp. occurred in Cayo Largo del Sur in 2012 (Loza et al., 2013), and by *Chattonella* sp. in La Redonda Lagoon in 2013 (Moreira-González and Comas-González,

2014). *Gambierdiscus*, *Ostreopsis* and *Prorocentrum* have been quantified in the southern Cuban coast, and a high diversity of *Gambierdiscus* species was revealed through qPCR assays (Díaz-Asencio et al., 2016).

In El Salvador, *P. bahamense* blooms and high levels of saxitoxins in shellfish occurred in 2011 and 2012 (Espinoza et al., 2013a). Also, in 2012, a bloom of *Alexandrium peruvianum/ostenfeldii* at Los Cóbano was associated with sea turtle deaths (Espinoza et al., 2013b); and a bloom by *C. polykrikoides* caused abundant scum and mass fish mortalities in the coast of La Libertad (Espinoza et al., 2013b). A massive sea turtle death in 2013, was associated with *G. catenatum*, and *P. bahamense* (Amaya et al., 2014). Regarding epiphytic dinoflagellates, the most abundant taxon is *Prorocentrum lima* (80 cells/g), and *Gambierdiscus* sp. and *Ostreopsis* sp. are present in low densities (4 and 9 cell/g respectively; Quintanilla and Amaya, 2017).

On the Pacific coast of Nicaragua, in 2005, a *P. bahamense* bloom caused 50 human intoxications and one death

(Chow et al., 2010). In Punta de El Este and La Paloma, Uruguay, between 2010 and 2018, fourteen toxic events by *Dinophysis* cf. *ovum*, two by *G. catenatum* and two by both species, produced PST concentrations in bivalves above the regulatory level (0.8 mg STXeq/kg meat) and positive mouse bioassay results for Diarrhetic Shellfish Poisoning (DSP). In the Caribbean coast of Guatemala, monitoring reports since 2015, include *Prorocentrum* spp. (14–5,492 cell/g), *Ostreopsis* sp. (0–165 cell/g), *Gambierdiscus* spp. (0–16 cell/g, **Supplementary Figure S2**) and *Coolia* sp. (0–7 cell/g) (unpublished data). Whereas, in the southern Caribbean coast of Costa Rica, the reports since 2016, included *Coolia monotis*, *C. tropicalis*, *Ostreopsis* spp., *Prorocentrum* spp. and *Gambierdiscus* spp. (unpublished data).

Despite *Gambierdiscus* species being responsible for CFP, and having been reported in Colombia, Cuba, El Salvador, Guatemala and Costa Rica, none of these studies informed intoxication cases owing to this taxon.

Nuclear Techniques for the Study of HABs

Toxin analysis

In El Salvador, between 2010 and 2017, more than 300 RBA analyses of PST have been performed in marine biota (e.g., puffer fish, snails, crabs, mussels, clams, oysters, shells, phytoplankton and sea turtles). PST concentrations at risk for consumption have been regularly found in *Crassostrea iridiscens* oysters, with concentrations reaching levels as high as 28.09 mg STXeq/kg in 2011 (Amaya et al., 2012, 2014). In 2013, ~200 sea turtles died, and RBA analysis indicated the presence of saxitoxins in diverse tissues of some turtle (Amaya et al., 2014). Owing to the permanent HAB monitoring program, and the toxins quantification by RBA, the intoxication cases associated with PSP have diminished in El Salvador, since both actions allow providing scientific information and early warnings to decision makers, which facilitates rapid responses to enforce shellfish harvest and trade bans, and protect the population from the consumption of contaminated seafood.

Ciguatoxins were detected for the first time in fish tissues from Cuba (Díaz-Asencio et al., 2016) using a recently optimized RBA method. Guidance on RBA use and quality control checks for toxin screening of fish samples is provided in Díaz-Asencio et al. (2018). This optimized protocol supports a full validation of the assay, a necessary step to develop and implement a regulatory monitoring program for ciguatoxins in seafood products.

Retrospective studies of the occurrence of potential harmful species

Historical reconstruction of HAB events have been conducted in Mexico through the analysis of dinocyst abundances in ²¹⁰Pb dated sediment cores. A sediment core collected in April 2016, in Punta Caracol (Mexican Caribbean coast; **Figure 1**), recorded very scarce dinocysts (total abundance: 178 ± 34 cyst/g) with all species belonging to *Spiniferites* taxa (*Spiniferites* spp., *S. ramosus*, *S. hyperacanthus*, *S. belerius*, *S. mirabilis*, **Supplementary Figure S2**, unpublished data).

In the Gulf of Tehuantepec (Pacific coast) and San Jose Lagoon (Gulf of California), *Polysphaeridium zoharyi* (cyst

of *P. bahamense*) was the most representative species and was found in the sediments since more than 100 years ago. Both studies indicated that climate variability might be an important driver in the dynamics of this species. The highest *P. zoharyi* fluxes in the Gulf of Tehuantepec were associated with low sea surface temperatures (La Niña episodes) and increasing rainfall (El Niño events) (Sanchez-Cabeza et al., 2012); and in San Jose Lagoon, with increments in rainfall and in minimum atmospheric temperatures (Cuellar-Martinez et al., 2018).

HAB Records in an International Database

The network recognized the lack of HAB records in LAC and found synergy with HAEDAT to integrate information about HABs that have occurred in the region. The network laboratories have provided 184 new data within the past 6 years. The database HAEDAT was consulted in June 2018 and until then, 223 HAB events occurred in countries from the network (between 1 and 53 records per country; **Table 1**), the oldest being from 1956.

The common taxa in the region are *Alexandrium*, *Pyrodinium*, *Cochlodinium* and *Gymnodinium*. The most reported HAB forming species are *Alexandrium catenella* (Chile), *G. catenatum* (Mexico and Uruguay) and *P. bahamense* (Mexico, El Salvador, Costa Rica and Guatemala). In recent years the occurrence of *C. polykrikoides* blooms has become evident in Cuba and El Salvador.

Harmful Algae Event Database is a good platform to highlight the problem of HAB occurrence in LAC countries and facilitate the incorporation of results emanating from the monitoring programs. However, based on the current records available, the appraisal of the present situation regarding HABs in LAC would be largely underestimated, since not all HAB events are registered yet. For instance, despite CFP being a threat to human health in the Caribbean (~2400 intoxication cases, **Supplementary Table S1**), the records in HAEDAT are still scarce (19 events). It is important to maintain constant efforts to update the information to maximize the benefits of the database for coastal management and to improve the global perspective about HABs.

CHALLENGES AND PERSPECTIVES ON HAB RESEARCH IN LAC THROUGH THE NETWORK

The main obstacles encountered by the network to establish and sustain continued HAB monitoring programs, are (a) lack of knowledge and awareness of the magnitude of the HAB problem in the region; (b) dependency on importation for many reagents and materials needed for the monitoring programs; (c) high turn-over of trained personnel; and (d) lack of financial resources.

Currently, under the project RLA/7/022 “Strengthening Regional Monitoring and Response for Sustainable Marine and Coastal Environments” the immediate objectives include: dissemination of the information on the installed capacity

of nuclear technologies and their applications for monitoring of stressors in the marine-coastal environments in LAC; the integration of strategic partners in the network activities to establish long lasting flows of validated information; and the consolidation of processes to transfer network-generated information to stakeholders, for evaluation of socio-economic impacts derived from environmental damage in the region, which would allow mitigation and remediation decisions.

Recent network efforts aim at communicating the activities and major accomplishments to decision makers, international institutions and general public. The dissemination of these achievements is expected to help improving the understanding and recognition of the threat that HABs represent to public health and sustainable coastal management. Improving the network visibility can also facilitate access to international funding and promote cooperation with other HAB-related networks. Locally, this could lead to stronger governmental engagement to maintain and expand HAB and biotoxin monitoring programs, with the ultimate goal of safeguarding seafood safety and the generation of scientific knowledge to better prevent, control and mitigate HAB occurrence and impacts.

The establishment of early warning systems in the rest of the network countries, the adoption of molecular techniques to improve the identification of HAB forming species, and the study of HABs in freshwater environments, certainly should be included in further efforts. Also, the network intends to improve the study of other marine stressors such as sea level rise and contaminants; and capacity is currently being built for ocean acidification monitoring and microplastic quantification.

CONCLUSION

Despite recurrent toxic blooms and drastic consequences for human health and the economy of LAC countries, response strategies have been quite unequal in the region. The creation of a network enabled countries to work together, with the support of IAEA technical cooperation projects, to build on their existing skills and capacity. Regional efforts have led to successful establishment of mature, standardized and up-to-date methodologies for identification and quantification of potentially harmful species, for toxin analysis and for temporal reconstruction of HAB occurrences. Scientific information and HAB occurrence data have been generated, and pilot surveillance of HABs biotoxins allowed the establishment of an early warning system in El Salvador.

Most countries in the region are challenged by the lack of resources to maintain continuous monitoring programs or

expand monitoring efforts beyond HABs. It is therefore essential to strengthen south-south collaborations within the network laboratories and among other LAC countries, to promote alliances with local and international institutions, and take steps forward the sensitization and awareness of local authorities and general public about the threat of HABs in the region. The effective transfer of the scientific results obtained through the network are a key step to support the coastal management and to facilitate decision making toward the prevention and mitigation of HAB impacts in LAC.

AUTHOR CONTRIBUTIONS

MYDB and ACRF contributed to the conception and design of the study. TCM, RQ, and LRVG organized the database. TCM performed the statistical analysis. TCM and ACRF wrote the first draft of the manuscript. MYDB, CAH, OAM, RQ, HLCO, NA, LDA, SMM, MV, and NFCW wrote sections of the manuscript. All authors contributed to manuscript revision, read and approved the submitted version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fmars.2018.00409/full#supplementary-material>

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