

Lithothamnion sp. as biostimulant in plant cultivation¹

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ABSTRACT

One of the various seaweed species with biostimulating properties is *Lithothamnion* sp., a calcareous seaweed recognized for its nutritional attributes. This review article aimed to gather information on *Lithothamnion* sp. and its applications in agricultural cultivation, focusing on identifying and analyzing its biostimulant effects. The practical use of this biostimulant in farming has been confirmed by studies highlighting its efficiency, which varies according to the source material (deposit and particle fraction), application methodology (dosage, methods and frequency) and specific crop (genotype and development stages). *Lithothamnion* sp. is notable for promoting vegetative growth and has established itself as an invaluable biostimulant in producing seedlings of various species. Its application, either via soil or by foliar methods, has led to improvements in the yield and quality of vegetables, fruits, oilseed crops, grains and forage plants. Although the underlying mechanisms need further investigation, the results suggest that *Lithothamnion* sp. contributes to amplifying photosynthesis, water-use efficiency and phytoalexin production.

KEYWORDS: Coralline algae, rhodolite, biofertilizers.

Agriculture is significantly challenged by climate change, which has intensified environmental pressures and increased the demand for agricultural inputs to maintain yield and reduce economic losses (Malhi et al. 2021). In the quest for more sustainable agricultural practices, research has shifted towards developing new inputs capable of effectively addressing these challenges (Raza et al. 2019). In Brazil, the National Bioinput Program was established in May 2020 through Decree n° 10,375 (Brasil 2020b).

Bioinputs are defined as products, processes or technologies derived from biological sources that

RESUMO

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Diversas algas possuem efeitos bioestimulantes, como o *Lithothamnion* sp., uma alga marinha calcária conhecida por seu valor nutricional. Objetivou-se, neste artigo de revisão, compilar informações sobre o *Lithothamnion* sp. e suas aplicações em cultivos agrícolas, visando identificar e analisar seus efeitos bioestimulantes. A aplicação prática desse bioestimulante na agricultura tem sido verificada por meio de estudos que demonstram sua eficácia dependendo da matéria prima utilizada (jazida e fração da partícula), tecnologia de aplicação (dosagem, formas e intervalos) e cultura (genótipo e estádios de desenvolvimento). O *Lithothamnion* sp. se destaca por induzir o desenvolvimento vegetativo e ser uma ferramenta valiosa na produção de mudas de diversas espécies. Sua aplicação, seja via solo ou foliar, tem revelado melhorias na produção e qualidade de hortaliças, frutas, culturas oleaginosas, grãos e forrageiras. Ainda que os mecanismos subjacentes exijam investigações mais aprofundadas, os resultados sugerem que o *Lithothamnion* sp. contribui para o aumento na fotossíntese, eficiência do uso da água e produção de fitoalexinas.

PALAVRAS-CHAVE: Algas coralinas, rodolito, biofertilizantes.

produce positive impacts on the chemical, physical and biological processes of target organisms (Brasil 2020b). This definition aligns with the concept of biostimulants, even though this specific term was not directly mentioned in the decree. Biostimulants, in turn, include substances or microorganisms applied to plants to enhance nutritional efficiency, improve tolerance to abiotic stresses and/or the inherent qualities of crops, regardless of their nutritional content (Du Jardin 2015). This perspective aligns with European Union regulations, which classify biostimulants on par with fertilizers under Regulation

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(EU) 2019/1009, dated June 5, 2019 (União Europeia 2019).

In Brazil, products with biostimulant compounds are classified as biofertilizers. This category encompasses products derived from amino acids, humic substances, algae extracts, plant extracts or combinations thereof, as long as their bioactive properties are verified (Brasil 2020a). The regulation for this classification falls under the Ministry of Agriculture, Livestock and Supply, as per Normative Instruction nº 61, dated July 8, 2020. This regulation requires the scientific validation of bioactivity using bioassays (Brasil 2020a). However, in Brazil, most biostimulant products are registered under the category of organic fertilizers, which do not require proof of bioactivity.

Biofertilizers from the class of seaweed extracts or processed seaweeds must declare a minimum content of 1 % of alginic acid for liquid products and 5 % for solid products (Brasil 2020a). Although various algae exhibit biostimulant effects, only certain large brown algae, predominantly found in temperate climates, contain alginic acid (Hurtado et al. 2020, Gómez-Matos et al. 2023). Since few marine macroalgae and no red algae (Rhodophyta) contain alginic acid (Gómez-Matos et al. 2023), products containing these algae cannot be registered as biofertilizers. Such gap highlights the shortcomings of the Brazilian legislation concerning biostimulants, leading to uncertainty throughout the production chain, especially among farmers.

Among red marine algae, the *Lithothamnion* genus, also known as *Lithothamnium*, is the only one classified under the category of mineral fertilizer, as per Normative Instruction nº 39, dated August 8, 2018, which considers only the mineral fraction of the algae (Brasil 2018). This classification overlooks the organic fraction that is rich in humic acids with auxinic activity (Amatucci et al. 2020).

This review article aims to gather information about *Lithothamnion* sp. and its implications on agricultural cultivation, focusing on identifying and analyzing its biostimulant effects.

Lithothamnion is classified under the Eukaryota domain/empire, Plantae kingdom, Biliphyta subkingdom, Rhodaria infrakingdom, Rhodophyta phylum, Eurhodophytina subphylum, Florideophyceae class, Corallinophycidae subclass, Corallinales order, Mesophyllineae suborder, Hapalidiaceae family and *Lithothamnion* genus (AlgaeBase 2023). This genus is commonly referred

to as coralline algae and is a member of the red algae group (Coutinho et al. 2022).

The *Lithothamnion* genus comprises 76 species, three varieties and 10 forms recognized in the taxonomic database. Additionally, it includes 51 names with an uncertain status, and three names that are yet to be verified (AlgaeBase 2023). Despite efforts, including the application of molecular techniques (Coutinho et al. 2022), an accurate and exhaustive taxonomic identification of *Lithothamnion* sp. remains complex and insufficiently explored (Sissini et al. 2022).

Lithothamnion is characterized by its ultraporous material, which has a high specific surface area. It contains minerals such as magnesian calcite, calcite and aragonite with xenomorphic and granular crystals (Silva et al. 2021). Additionally, it has isotropic calcium particles (Raut & Gadani 2021) and humic substances (Amatucci et al. 2020). Its complex composition encompasses calcium and magnesium carbonates, as well as over 20 trace elements, which vary in composition and quantity (Dias 2000).

From an economic perspective, the significance lies in the unconsolidated sedimentary materials derived from algae, which are evident as rhodoliths, nodules and bioclastic fragments (Paiva et al. 2023). Bioclastic granules, which represent a highly fragmented portion of sedimentary structures, are formed through processes such as water movement, bioturbation, grazing and dragging (Foster 2001). This granular composition can also include additional benthic materials such as shells, algae and animal remains (Helias & Burel 2023).

The extraction of sedimentary material from algae is conducted using the dredging method (Paiva et al. 2023), which is characterized by its low selectivity, enabling the collection of all unconsolidated material. During processing, the raw material undergoes washing to eliminate the excess of salts and impurities (Souza & Martins 2008), which is then followed by drying and grinding (Dias 2000). Products available in the market differ in their particle sizes, which depend on the grinding intensity. Those with smaller particle sizes are termed 'micronized', which entail mechanical breakdown due to friction between particles, resulting in sizes ranging from 1 to 10 µm (Amatucci et al. 2020).

Various terms describe the sedimentary materials of calcareous algae, including maerl

(Helias & Burel 2023, Ingrassia et al. 2023), bioclastic granules (Veneu et al. 2018 and 2019) and rhodoliths (Rendina et al. 2020, Sissini et al. 2022). The term “rhodoliths” is particularly relevant for characterizing these structures. However, the concept of bioclastic granules is more encompassing (Vale et al. 2022). Rhodoliths are free-living structures mainly comprised of 50 % non-geniculated coralline algae. They display a diverse range of shapes, sizes and species, and can exist in both living and fossil forms (Foster 2001). Rhodoliths often associate with other benthic organisms, bioclastic sediments and various species of coralline or epiphytic algae (Carvalho et al. 2020, Vale et al. 2022), delivering a plethora of environmental services, notably in preserving biodiversity (Ingrassia et al. 2023). Unregulated commercial exploitation of these areas may lead to significant environmental challenges, as observed in France (Bernard et al. 2019). This underscores the importance of innovative conservation strategies for these organisms and their habitats (Carvalho et al. 2020), not only to Brazil - which has a considerable potential for the commercial exploitation of rhodoliths - but also to other countries, due to its global significance (Tuya et al. 2023).

Coralline algae have been documented in both living and fossil forms across various regions, including the Mediterranean, Gulf of California, Atlantic coast of Norway, Ireland, northeastern Canada, eastern Caribbean, Brazil, southern Japan and western Australia (Foster 2001). Computational studies suggest an even broader distribution for these organisms (Fragkopoulou et al. 2021). Notably, the Brazilian coast is home to the world’s largest rhodolith reserve (Amado-Filho 2012). The diversity of calcareous algae species comprising the rhodoliths is significant along Brazil’s coast, as evidenced by studies in the states of Espírito Santo (Amado-Filho et al. 2010, Sissini et al. 2022), Bahia (Costa et al. 2014), São Paulo (Pereira Filho et al. 2019), Sergipe and Alagoas (Vale et al. 2022), the Amazon river delta (Moura et al. 2016, Vale et al. 2018) and Ceará (Carneiro et al. 2021).

The rich diversity of calcareous algae species in the Espírito Santo state, which includes endemic species, stems from its position in a transitional zone between the tropical and warm temperate regions of Brazil (Carvalho et al. 2020), offering a diverse environment that fosters an increased biodiversity.

Commercial deposits of calcareous algae in Brazil are found along the coastlines of the states

of Maranhão, Bahia and Espírito Santo. These are operated by the licensed companies Oceana Minerals, PrimaSea and Supramar, respectively. No comparative analysis of the nutritional composition of these commercial products was found in existing literature. Therefore, in this review, we conducted a detailed analysis of both macro and micronutrients, as well as humic substances found in these different sources (Table 1). This information will facilitate a deeper understanding of the biostimulant effects associated with these products. This topic will be further discussed in subsequent sections of this review article. Based on our analyses, noticeable variations in composition from different extraction regions were observed, especially concerning the levels of iron, manganese, zinc, sodium and humic acid.

Moreover, the presence of amino acids in the commercial product LT Supra® was assessed. A sample was sent to the Instituto Campineiro de Análise de Solo e Adubo Ltda. This institute used the pre-column derivatization with phenyl isothiocyanate method, combined with liquid chromatography for amino acid analysis in foods (Hagen et al. 1989). The Walters Pico-Tag System (White et al. 1986) was used for the aminogram analysis. The results indicated a concentration of 1,400 mg kg⁻¹ of free amino acids

Table 1. Result for the chemical analysis of nutrients and humic substances of the commercial product from the companies Oceana Minerals (Algen®), Prima Sea (Primaz®) and Supramar (LT Supra®).

Parameter*	Unit	Analyzed products		
		Algen®	Primaz®	LT Supra®
Nitrogen		0.05	0.05	0.06
Phosphor		0.06	0.06	0.09
Potassium	%	0.03	0.04	0.06
Calcium		34.28	30.29	31.19
Magnesium		3.21	3.62	2.06
Sulfur		0.22	0.28	0.29
Boron		31.17	30.24	48.06
Copper		0.16	0.94	0.97
Iron	mg kg ⁻¹	717.01	4,527.96	14,765.56
Manganese		11.91	49.67	481.89
Zinc		1.19	2.88	10.50
Sodium		3,744.25	4,899.77	8,084.48
Fulvic acid	%	6.77	5.40	9.31
Humic acid		3.16	4.91	0.93

* The products analyses were carried out in the ABCLab commercial laboratory of the Fundação ABC from samples of the commercial products, using the following methodology: EPA 6010/3051 (Brasil 2017), AOAC - Official Method 993.13 - Nitrogen (Total) in Fertilizers (Tate 1994).

(0.15 %). Notably, glycine and tryptophan were predominant, each present at 400 mg kg⁻¹ of the product. Furthermore, the analysis identified aspartic acid (200 mg kg⁻¹), alanine (200 mg kg⁻¹), proline (100 mg kg⁻¹), valine (100 mg kg⁻¹) and glutamic acid (0.01 mg kg⁻¹).

Lithothamnion sp. has been studied for its effects on the seedling production of various agricultural crops. However, its impact varies depending on the dosage used. For physic nut seedlings (*Jatropha curcas* L.), incorporating a dose of 6.68 kg m⁻³ into the substrate led to increased height, stem diameter, leaf area and total biomass (Evangelista et al. 2016). In the case of sweet passion fruit seedlings (*Passiflora alata* C.), adding 2 kg m⁻³ to the substrate led to increased shoot growth (Souza et al. 2007). For Japanese quince seedlings [*Chaenomoles senensis* (Koehne)], the optimal dose was found to be 5 kg m⁻³ (Sabino et al. 2013). For Cleóprata mandarin seedlings (*Citrus reshni* Hort ex Tan.), the optimum dose was determined to be 5 kg m⁻³ (Cruz et al. 2008). *Lithothamnion* sp. also exhibited genotypic effects on papaya seedlings (*Carica papaya* L.). The Formosa group showed a favorable response at 2 kg m⁻³ (Teixeira et al. 2009), while the Solo group benefited from a dose of 3 kg m⁻³ (Hafle et al. 2009).

The interaction with the substrate plays a crucial role in the *Lithothamnion* sp. effects. The application of 5.25 kg m⁻³ of *Lithothamnion* sp., when combined with cattle manure, resulted in increased root growth and height of Arabica coffee seedlings (*Coffea arabica* L.). A similar effect was observed with 1.75 kg m⁻³ of *Lithothamnion* sp. when combined with filter cake (Rodriguez et al. 2017). Furthermore, the initial growth of Swingle citrumelo seedlings (*Citrus paradisi* Mac × *Poncirus trifoliata* L. Raf.) was enhanced with the addition of *Lithothamnion* sp. at the concentration of 5 (v:v), depending on the substrate composition (Araújo et al. 2007).

Another application in seedling production is for the acidity correction, attributed to its calcium and magnesium carbonate contents. The incorporation of *Lithothamnion* sp. at a dosage 1.4 times higher than the conventional recommendation through the base saturation method led to an increased total dry mass in passion fruit (*Passiflora alata* C.) (Souza et al. 2009).

A promising path for further research is the use of *Lithothamnion* sp. in seedlings propagated from cuttings. While the auxinic bioactivity of

Lithothamnion sp. has been confirmed (Amatussi et al. 2020), few studies have specifically focused on its use in seedlings propagated from cuttings. This gap presents numerous opportunities to delve into its potential within this propagation method.

The application of *Lithothamnion* sp. to agricultural crops has shown significant and diverse effects consistent with the definition of biostimulants. The dose-dependent response is evident, similar to other algal species (Kapoore et al. 2021). The application method varies: it can be incorporated into the soil or applied as foliar sprays. For the latter, *Lithothamnion* sp. is used in its micronized form.

Adding *Lithothamnion* sp. to soil enhances plant growth and boosts sweet pepper (*Capsicum annum* L.) yield (Evangelista et al. 2016). When combined with poultry and cattle manure, there was a noticeable increase in red dragon fruit [*Hylocereus undatus* (Haw.) Britton & Rose] yield (Moreira et al. 2011 and 2012b, Costa et al. 2015). Improvements were also observed in height, leaf dry weight, stem dry weight and stem diameter for castor bean (*Ricinus communis* L.), sunflower (*Helianthus annuus* L.) and radish (*Raphanus sativus* L.) (Evangelista et al. 2015).

The production quality can also be improved when *Lithothamnion* sp. is incorporated into the soil, especially when combined with the practice of chemical thinning using 600 mg L⁻¹ of Ethepon in the cultivation of pokan tangerine (*Citrus reticulata* Blanco), resulting in an increase in both fruit size and soluble solids content (Moreira et al. 2012a).

However, it is important to note that the dosage and interactions between system components can influence the results. Negative effects were observed on carrot (*Daucus carota* L.) yield when *Lithothamnion* sp. was used as a source of calcium and magnesium in combination with potassium (Rodrigues Neto et al. 2021). This result can be attributed to the high dosage used (200 kg ha⁻¹). Previous studies and researches on other biostimulant algae suggest that growth stimuli are more effective when multiple doses of very low values are applied (Shukla et al. 2019). Additionally, biostimulants should not be used as a substitute for mineral nutrients. Instead, they should complement fertilizers, aiming to enhance the efficiency of these fertilizers and reduce nutrient application rates (União Europeia 2019).

The soil acidity correction in agricultural crops is another significant application of *Lithothamnion*

sp., owing to its content of calcium and magnesium carbonate. When applied at a base saturation below 70 %, it promoted increased bean (*Phaseolus vulgaris* L.) yield (Melo & Furtini Neto 2003). Similar results were observed in maize (*Zea mays* L.) at 70 % base saturation, in which case there was a rise in shoot dry mass (Chaves et al. 2022).

There are few studies on the effects of *Lithothamnion* sp. in enhancing nutritional efficiency. This aspect requires further investigation, especially since one anticipated benefit of biostimulants is improved nutrient use efficiency (Ricci et al. 2019). Only one study was found that examined maize's ability to improve nitrogen absorption using urea as source. However, there were no significant effects observed when using doses of 27, 44 and 53 kg ha⁻¹ of *Lithothamnion* sp. applied to the soil (Bernardes et al. 2016).

With foliar application, *Lithothamnion* sp. enhances plant development, yield and product quality, influencing various physiological and biochemical characteristics. Weekly foliar applications of *Lithothamnion* sp. nanoparticles in melon (*Cucumis melo* L.) cultivation resulted in increased shoot and root biomass (Negreiros et al. 2019). Similarly, weekly foliar sprays with micronized *Lithothamnion* sp. boosted the growth of both the shoot and roots of tomato (*Solanum lycopersicum* L.) plants. This treatment also raised the levels of total free amino acids and sugars in the leaves and roots, increased the protein content in the leaves and augmented the sugar levels in the fruits (Amatucci et al. 2020).

The induction of root development, yield characteristics and biochemical parameters (Amatucci et al. 2020) have been demonstrated in the cultivation of onion (*Allium cepa* L.) using micronized *Lithothamnion* sp. with foliar application, both independently (Mógor et al. 2021) and in combination with the *Arthrospira platensis* cyanobacterium (Amatucci et al. 2023).

The foliar application of a commercial product containing *Lithothamnion* sp. led to an increase in the foliar levels of the boron and copper micronutrients in grape (*Vitis vinifera* L.) (Carvalho et al. 2019). This result suggests a biostimulant effect of *Lithothamnion* sp. on improved nutrient absorption. However, further studies are needed to identify the underlying mechanisms and interactions with other minerals.

The association of *Lithothamnion* sp. foliar application with water-soluble nutrients and free

amino acids enhanced the commercial quality of Kent mangoes (*Mangifera indica* L.), leading to a reduced seasonality and, consequently, increased yield (Lobo et al. 2019). Foliar applications of a commercial product containing 70 % of *Lithothamnion* sp. resulted in a 20 % increase in the Palmer mango yield (Simões et al. 2022). Both studies involving mangoes indicate that the effects are contingent upon the dosage and climatic conditions. Many factors can influence the performance of a biostimulant in the field (Ricci et al. 2019). These results further confirm the biostimulant effect of *Lithothamnion* sp.

The mechanisms by which *Lithothamnion* sp. exerts biostimulant effects on plants remain poorly understood. An auxinic effect was observed following the application of 1.76 g L⁻¹ of micronized *Lithothamnion* sp. to the roots of *Vigna radiata* L. This effect is attributed to the presence of 31.36 µg L⁻¹ of humic acids (Amatucci et al. 2020). These results elucidate the pronounced rhizogenesis-inducing effect observed in several species of agronomic significance, as previously discussed in this article.

Foliar applications of a commercial product containing *Lithothamnion* sp. led to increased photosynthetic rates, transpiration, stomatal conductance, water-use efficiency, carboxylation efficiency and chlorophyll content in grape (*Vitis vinifera* L.) (Carvalho et al. 2019). These results elucidate the stimulatory effects observed on the growth and yield of several species discussed in this article.

The biostimulant effects of *Lithothamnion* sp. on inducing tolerance to abiotic stress have been poorly studied. However, it has been shown to enhance yield, sucrose content and fiber in sugarcane (*Saccharum* sp. L.), when used in conjunction with vinasse, both in rainfed and irrigated cultivation (Rodriguez et al. 2018). These results demonstrate improved water-use efficiency in sugarcane, similarly to observations made regarding photosynthetic parameters in grape (*Vitis vinifera* L.) (Carvalho et al. 2019).

While the role of inducing plant defenses against biotic stresses has been excluded from the definition of biostimulants, specifically to differentiate them from pesticides (Du Jardin 2015), they function to alleviate the negative impacts of both biotic and abiotic stresses on plants through various mechanisms (molecular alterations, physiological, biochemical and anatomical modulations) (Del

Buono 2021). *Lithothamnion* sp., when combined with rosemary (*Rosmarinus officinalis* L.), has been shown to enhance the production of phytoalexins in bean (*Phaseolus vulgaris* L.) and soybean [*Glycine max* (L.) Merrill] (Faria et al. 2022), demonstrating its potential as a defense mechanism inducer.

The application of *Lithothamnion* sp. has been shown to stimulate the accumulation of root biomass in agricultural crops such as peppers, Arabica coffee and onion. Furthermore, it stimulates the accumulation of shoot biomass in papaya, castor bean, sunflower, forage turnip, pepper, melon, tomato, bean and maize. Additionally, it enhances

yield and quality in pepper, red dragon fruit, pokan tangerine, sugarcane, tomato, onion, mango and bean. It also impacts physiological rates in grape and acts as a resistance inducer in soybean and bean. The biostimulating effects of *Lithothamnion* sp. on agricultural plants are dose-dependent (Table 2).

Lithothamnion sp. is a calcareous alga that belongs to the red algae group. Its mineral composition and humic substance content vary based on the extraction location. Consequently, commercial products derived from it have differences among them, which can result in varied responses when stimulating plants. However, no studies have been

Table 2. Results from *Lithothamnion* sp. according to dose and species.

Crop	Treatments	Optimal dose	Increments (%)	References
Jatropha		6.68 kg m ⁻³	PH (34 %) and TDM (51 %)	Evangelista et al. (2016)
Sweet passion fruit	Doses	2 kg m ⁻³	PH (94 %)	Souza et al. (2007)
		1.4 times the base saturation	TDM (107 %)	Souza et al. (2009)
Papaya		2 kg m ⁻³	PH (6 %)	Teixeira et al. (2009)
		3 kg m ⁻³	PH (4 %)	Hafle et al. (2009)
Arabica coffee	Doses and associations	1.75-5.25 kg m ⁻³	RL (13 %)	Rodriguez et al. (2017)
Citrumelo		5 % (vv)	TDM (22 %)	Araújo et al. (2007)
Quince	Doses	5 kg m ⁻³	TDM (106 %)	Sabino et al. (2013)
Tangerine		5 kg m ⁻³	SDM (65 %)	Cruz et al. (2008)
Pepper		0.54 kg m ⁻³	RDM (100 %) and YLD (6 %)	Evangelista et al. (2016)
		2.2 kg m ⁻³	YLD (119 %)	Moreira et al. (2011)
Dragon fruit	Doses and associations	1.9 kg m ⁻³	YLD (+ 104 %)	Moreira et al. (2012b)
		2.5 kg m ⁻³	YLD (+ 716 %)	Costa et al. (2015)
Tangerine		0.8 kg plant ⁻¹	YLD (35 %)	Moreira et al. (2012a)
Sugar cane		200 kg ha ⁻¹	YLD (45 %)	Rodriguez et al. (2018)
Castor bean		428 kg ha ⁻¹	SDM (125 %)	
Sunflower	Doses	432 kg ha ⁻¹	SDM (653 %)	Evangelista et al. (2015)
Fodder turnip		527 kg ha ⁻¹	SDM (292 %)	
Melon		1-5 kg ha ⁻¹	TDM (32 %)	Negreiros et al. (2019)
Tomato		1.4-1.93 g L ⁻¹	TDM (15 %), AA (23 %), PTN (96 %) and GLC (22 %)	Amatussi et al. (2020)
Onion	Doses and associations	1.5 g L ⁻¹	RDM (124 %), YLD (20 %), AA (45 %) and GLC (20 %)	Amatussi et al. (2023)
	Doses		RDM (30 %) and YLD (16 %)	Mógor et al. (2021)
Mango	Doses and associations	2.5 mL L ⁻¹	YLD (56 %)	Lobo et al. (2019)
	Commercial product (70 % of seaweed)	12 L plant ⁻¹	YLD (20 %)	Simões et al. (2022)
Grape	Commercial product	0.6 %	P _N (16 %), g _s (21 %), WUE (10 %), EiC (13 %), Chl (4.3 %), B (18 %) and Cu (33 %)	Carvalho et al. (2019)
Bean and soybean		0.5 g L ⁻¹	PHY bean (123 %) and soybean (401 %)	Faria et al. (2022)
Bean	Doses and associations	0.61-1.09 t ha ⁻¹	YLD (+ 350 %)	Melo & Furtini Neto 2003
Corn		70 % of base saturation	SDM (75 %)	Chaves et al. (2022)

PH: plant height; RL: root length; TDM: total dry mass; SDM: shoot dry mass; RDM: root dry mass; YLD: yield; AA: amino acids; PTN: proteins; GLC: glucose; P_N: photosynthetic rate; g_s: stomatal conductance; WUE: water-use efficiency; EiC: intrinsic carboxylation efficiency; Chl: chlorophyll content; B: boron; Cu: copper; PHY: phytoalexins.

conducted comparing the effects of these different commercial products.

The *Lithothamnion* sp. potential for agricultural use has been demonstrated in seedling production and in the cultivation of horticultural, fruit, oilseed, grain and forage species. However, its application needs to be extended to various plant species.

The *Lithothamnion* sp. biostimulant role as an inducer of plant development was observed in species such as papaya and Arabica coffee, enhancing the growth of both the plant parts and roots. This effect was also observed in jatropha, citromelo, quince, sweet pepper, melon and tomato, in which it increased the total biomass. Additionally, *Lithothamnion* sp. has been shown to improve the production quality, boosting both the yield and quality of pepper, mango, bean, onion, dragon fruit, tangerine and sugarcane. It also enhances the accumulation of amino acids, proteins and carbohydrates in vegetable products, notably in tomato and onion.

Additionally, it increases the tolerance to water stress in sugarcane cultivation and enhances the water-use efficiency in hose irrigation. However, its effects need to be verified for other species and need to be further investigated for inducing tolerance to other abiotic stresses.

Studies confirming the enhanced efficiency of nutrient use still need to be conducted, even though a higher accumulation of boron and copper in vine leaves has already been demonstrated (Carvalho et al. 2019). No research has been identified concerning the availability of nutrients confined in the soil or rhizosphere. Therefore, the biostimulant effect of *Lithothamnion* sp. pertaining the reduction of nutrient use remains to be verified.

While the ability to induce tolerance to biotic stress is not included in the definition of biostimulants, it has been demonstrated that *Lithothamnion* sp. promotes the accumulation of phytoalexins in bean and soybean (Faria et al. 2022). However, no study has yet confirmed its effect on reducing pests and diseases in agricultural crops.

Research has shown that the effectiveness of these biostimulants depends on dosage, methods and intervals of application, particle size, type of crop, genotype, phenological stages and interactions with other biostimulant molecules (Amatuzzi et al. 2020 and 2023). Given this wide array of potential effects, types of crops and cultivation conditions, it is imperative to extend field research. This will help

to develop technical recommendations suitable for various agricultural systems and plant species of agronomic significance.

The mechanisms through which *Lithothamnion* sp. promotes plant development have not been extensively researched. However, there is evidence to support an increase in photosynthesis and an auxinic effect related to the stimulation of root growth. It is thus proposed that future studies should elucidate the physiological mechanisms underlying the response to *Lithothamnion* sp., as well as validate its effects on reducing environmental stress.

Future studies should also identify the bioactive effects of products containing *Lithothamnion* sp., excluding the mineral fraction, to demonstrate the effects of the seaweed's organic fraction alone.

Additionally, it is recommended to broaden the phycology research in the field for the identification of *Lithothamnion* species and to investigate the environmental sustainability aspects of their commercial exploitation.

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