



Cultivation and Domestication of *Kappaphycus alvarezii* Strains at Ubatuba Bay, São Paulo State, Southeastern Brazil

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Abstract

The red seaweed *Kappaphycus alvarezii* (Doty) L.M. Liao was introduced in Brazil by Professor Édison José de Paula from São Paulo University, in January 1995. The initial tetrasporophytic segments came from Usa Bay, Shikoku Island, Japan, and were originated from commercial crops carried out in the Philippines. The process of domestication has been ongoing since 1995-1996 through the cultivation of brown tetrasporophytic strain into the Experimental Marine Farm of Fisheries Institute, Ubatuba Bay, São Paulo State, Brazil. The initial brown tetrasporophyte originated red and green tetrasporophytic strains, and, after selection of tetraspore progeny, a pale brown gametophytic strain (initially named G11, and later Edison de Paula strain) was also originated. During the domestication process, the occurrence of spontaneous color strains of both diploid and haploid phases was recorded, and environmental data of temperature, salinity, and water transparency at the cultivation site were monitored. Currently, 12 color strains are being cultivated in Ubatuba Bay. The present chapter provides a comprehensive review on cultivation methods, grow rates, productivity, carrageenan yield and characterization of *K. alvarezii* strains cultivated at Ubatuba Bay. Characterization of these new strains should encompass factors such as refined and semirefined carrageenan content, biofertilizer yield, temperature resilience, epibiont presence, growth rate, yield, and identification of economically valuable biomolecules.

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To facilitate the preservation of these strains, a biobank with all color strains is implemented in both natural environment and laboratory conditions. These initiatives ensure secure strain storage and supply of *K. alvarezii* strains for local farmers.

Keywords

Biobank · Color strains · Edison de Paula Strain · Mariculture · Seaweed

9.1 Historic

The red alga *Kappaphycus alvarezii* (Doty) L.M. Liao (Rhodophyta, Solieriaceae) was introduced in Brazil in January, 1995, by Professor Édison José de Paula of São Paulo University. The initial thallus segments came from an experimental cultivation maintained in Usa Bay, Kochi, Shikoku Island, Japan, and originated from commercial crops carried out in the Philippines (Paula et al. 1998). The material was provided by Professor Masao Ohno with whom Prof. Paula worked with during his postdoctoral degree at the Usa Marine Biological Institute (Kochi University) in the second semester of 1994 (Paula 2001). Back in Brazil, Prof. Paula began to cultivate a branch 2.5 cm in length (about 2.5 g) in the Laboratory of Marine Algae, LAM (today called LAM Édison José de Paula), Institute of Biosciences, São Paulo University, São Paulo City. A unialgal non-axenic culture of the brown tetrasporophyte was established to eliminate associated species, the presence of which is considered one of the greatest risks in the introduction of exotic species (Paula 2001). This procedure was decisive in ensuring the introduction of the species into the sea without risking the simultaneous introduction of undesirable organisms.

The first seedlings of *K. alvarezii* destined for seawater experiments were produced after 10 months of vegetative propagation of the brown tetrasporophytic strain in the laboratory (Paula and Pereira 1998; Paula et al. 1998). A total of



Fig. 9.1 Original *Kappaphycus alvarezii* brown tetrasporophyte cultivated at the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. (Photo by Valéria Gelli)

20 branches (2.0–4.0 g) were produced and transferred to the subtropical waters of the southwestern Atlantic at the Experimental Marine Farm of the Fisheries Institute (EMFFI) (23° 26'90"S, 45°0'30"W), Paulista Technological Agribusiness Agency, Department of Agriculture and Supply, Ubatuba Bay, São Paulo State (Paula et al. 1999), in accordance with Brazilian legislation under the IBAMA Environmental License Process (IBAMA/MMA n° 02027.009179/1996-11) (Paula 2001; Paula et al. 2002). These *K. alvarezii* crops remain in cultivation to this day (Gelli 2019) (Fig. 9.1). Some studies reported on the introduction of a female gametophyte (Hayashi et al. 2007b), but it died after some years (Hayashi, L. pers. comm., 2022).

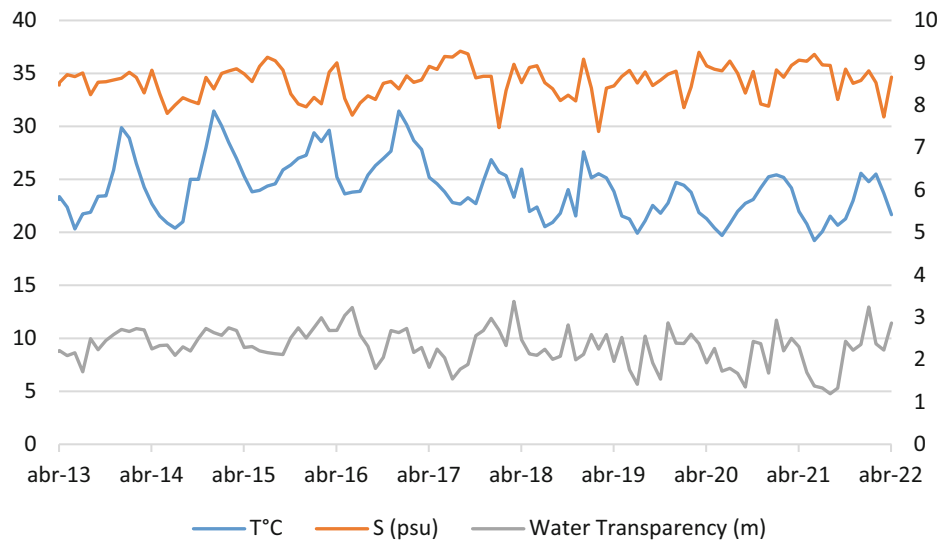
The initiative to introduce an exotic seaweed in Brazil and invest in techniques for its cultivation arose from the need to mitigate overexploitation of the natural banks of the native kappa carragenophyte *Hypnea pseudomusciformis* Nauer, Cassano and M.C. Oliveira (cited previously as *Hypnea musciformis*) and meet the demand for national importation of this product (Paula and Pereira 1998; Paula et al. 1999).

Kappaphycus alvarezii seedlings introduced in Ubatuba Bay were monitored and showed satisfactory growth (Paula et al. 1998, 2002). From some of these seedlings, originally brown in color, green and red branches appeared spontaneously and in low frequency (Fig. 9.2). These colored branches were then propagated and kept in cultivation at



Fig. 9.2 Brown, green, and red tetrasporophytic strains and pale brown EP gametophytic strain of *K. alvarezii* (from left to right) cultivated at the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. (Photos by Valéria Gelli)

Fig. 9.3 Average variations of temperature, salinity, and water transparency from April 2013 to May 2022 in the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. Source: Fisheries Institute (2022)



sea, showing the stability of these characteristics (Paula 2001). These color strains were evaluated for their growth rates, carrageenan content (Hayashi et al. 2007a), and pigments (Paula 2001) compared to the original strain, and they also remain in the seawaters of EMFFI.

In the summer and autumn months, December 1995 to May 1996, tetraspores were produced in cultivation experiments at sea, quite unexpectedly, though, since the strain had been considered infertile (Paula et al. 1999). Experiments on the germination of these tetraspores showed low viability (Paula et al. 1999). They were produced from brown plants and both green and red color variants (Fig. 9.2) (Bulboa et al. 2008). Rare plantlets were finally obtained in the laboratory after great experimental effort. These plantlets were very delicate and sensitive (Paula 2001). After fertility observation, there was monitoring in the tidal region near the crops and in deeper places, through scuba diving. However, no fixed plants were found in any substrates. These results indicated that the establishment of *K. alvarezii* via spore production in the natural environment of the southeastern coast of Brazil is remote (Bulboa et al. 2008). These important conclusions provide scientific bases to the legislation for the sea cultivation of *K. alvarezii* along Brazilian coast.

After vegetative propagation, some *K. alvarezii* plantlets derived from tetraspores were transferred to the sea and showed lower growth rates than parental tetrasporophytes, though mostly unviable in winter (Paula et al. 1999). However, one plantlet, G11, was more promising. Although it presented lower growth rates, this new pale brown gametophytic strain had higher carrageenan content (Paula 2001; Hayashi et al. 2007a). Later, in honor of Prof. Édison José de Paula, this strain was called EP (Hayashi et al. 2008a). However, in the older papers mentioned in this review, it is still termed G11. The EP strain is still cultivated at EMFFI. The haploidy (n) of the EP gametophyte was confirmed by confocal fluorescence microscopy (Zitta et al. 2012).

9.2 Cultivation of *Kappaphycus alvarezii* Strains in the Natural Environment (Ubatuba Bay) and Laboratory

9.2.1 Cultivation

Since 1995, *K. alvarezii* has been cultivated in a floating raft system (Fig. 9.2) in Ubatuba Bay and periodically replanted using the tie-tie system (Paula et al. 1999; Hayashi et al. 2007a; Solorzano-Chavez et al. 2019). In this system, ten fragments of each strain weighing about 100 g were tied in ropes, 2 m in length, and spaced 20 cm apart (Fig. 9.2). Harvesting and planting were then carried out every 30 days according to monthly marine farm management records.

9.2.2 Monitoring of Environmental Parameters

Temperature (°C), salinity (psu), and seawater transparency (m) were recorded in situ near the *K. alvarezii* culture raft every day from April 2013 to May 2022 in Ubatuba Bay. Figure 9.3 shows the variation of abiotic parameters.

9.2.3 New Spontaneous Strains

Throughout these years of cultivation, other variants of *K. alvarezii* have arisen spontaneously from the four strains already described, and these strains also continue to be propagated and maintained in both seawater and laboratory. They have been photographed and catalogued. As it appeared, each new strain was cultivated in the natural environment on 2-m ropes and with 10 branches. Furthermore, apical segments (Fig. 9.4) of these strains (about 10 g) were cultivated at the Seaweed Laboratory of the Fisheries

Fig. 9.4 Thallus aspect at the beginning of culture (left) and more developed thallus (right) of the new green color variant of *Kappaphycus alvarezii* originated from a red tetrasporophytic branch cultivated at the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. (Photo by Valéria Gelli)



Institute (SLFI) with two replicates for each color strain. According to SLFI protocol, they were kept in a temperature-controlled room at 25 °C with a photoperiod of 12 h and maintained in seawater (35 psu) changed weekly (Fig. 9.5).

Since the introduction of the EP gametophytic strain to the sea in 1998 (Paula et al. 1999), its phenotypic plasticity has been observed, and two new color variants have arisen from it (Fig. 9.6). Moreover, six new variants arose from the tetrasporophytic strains (Fig. 9.7).

9.3 Brief Review of Publications on *Kappaphycus alvarezii* Cultivated in Ubatuba Bay

9.3.1 Physiological Responses

Experimental conditions and the main results on growth rates and viability of tetrasporophyte and gametophyte in both field and laboratory conditions are summarized in Table 9.1. In sea cultivation, seasonal variation of growth rates was clearly related to seawater temperature (Paula et al. 2002).

Bulboa et al. (2008) evaluated tetraspore viability and concluded that tetraspores had low survival rates, mostly dying after 20 days, and that the recruitment of tetraspores did not occur in the field, indicating that the establishment of *K. alvarezii* via spore production in the natural environment of the southeastern coast of Brazil is remote. These important conclusions provide scientific bases to support legislation aimed at cultivating *K. alvarezii* along Brazilian coastal seawaters.

9.3.2 Carrageenan Yields and Characterization

Carrageenan yields and properties of brown, red, and green tetrasporophytes, EP gametophyte, and red female gametophyte are summarized in Table 9.2. Although the EP gametophyte showed lower growth rates when compared to tetrasporophytic strains (see Table 9.1), carrageenan yields were higher in EP gametophyte than those of other strains (Table 9.2). Comparing cultivation period, depth, and planting density, the only parameter that influenced carrageenan quality was the cultivation period (Hayashi et al. 2007b).

9.3.3 Tissue Culture and Micropropagation

Tissue culture and micropropagation techniques were used in five *K. alvarezii* strains: brown (BR), green (GR), and red (RD) tetrasporophytes; pale brown female gametophyte (BFG); and a strain originating from tetraspore germination (“Edison de Paula,” EP). Several experimental conditions were tested, such as culture media (half-strength of von Stosch (VS 50), Guillard and Ryther (F/2 50), and synthetic ASP 12-NTA medium) and the addition of glycerol or phytohormones (indole-3-acetic acid (IAA), 2,4-dichlorophenoxyacetic acid (2,4-D), and benzylaminopurine (BA), either alone or in combination) and colchicine (0.01%). The EP strain showed higher potential for micropropagation than the other strains. Regeneration was stimulated by the addition of glycerol, IAA: BA (5: 1 mg L⁻¹), and colchicine, which proved useful for improving the micropropagation of *K. alvarezii* (Hayashi et al. 2008a).

Fig. 9.5 Different color strains of *Kappaphycus alvarezii* cultivated at the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. EP = EP gametophytic strain; Brown, Green, Red = brown, green, and red tetrasporophytic strains, respectively. (Photo by Valéria Gelli)



Fig. 9.6 EP gametophytic strain (a) which originated two spontaneous color strains (b and c) of *Kappaphycus alvarezii* cultivated at the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. (Photo by Valeria Gelli)



9.3.4 Ecological Traits

Kappaphycus alvarezii showed potential as a biofilter when cultivated with fish effluents (*Trachinotus carolinus*), removing nitrate (18.2%), nitrite (50.8%), ammonium (70.5%), and phosphate (26.8%) (Hayashi et al. 2008b). However, growth rates of algae cultivated with fish effluents were lower than

those observed in sea cultivation as further claimed by the authors. Still, carrageenan yields did not differ between cultivation systems. Therefore, *K. alvarezii* cultivation with fish effluents not only reduced eutrophication but also produced carrageenan.

The environmental monitoring of *K. alvarezii* cultivation in Ubatuba Bay was performed from November 2016 to



Fig. 9.7 Tetrasporophytic strains (a, b, and c) and its spontaneous color strains (d, e, f, g, h, and i) of *Kappaphycus alvarezii* cultivated at the Experimental Marine Farm of the Fisheries Institute, Ubatuba Bay, São Paulo State. (Photo by Valéria Gelli)

Table 9.1 Physiological responses of *Kappaphycus alvarezii* strains cultivated in the field (Ubatuba Bay, São Paulo State) or in laboratory (Institute of Biosciences, São Paulo University)

Cultivation location	Strain(s)	Cultivation conditions	Physiological responses	References
Laboratory	BR tetrasporophyte	Fertile branches (10 cm length, 50 g FW) were selected from the sea during the reproductive period (December 1995 to May 1996). After cleaning, these branches were cultured at 25 ± 2 °C, under $60\text{--}100$ $\mu\text{mol photon m}^{-2} \text{s}^{-1}$, 14:10 h L:D cycle	Tetraspores were released and germinated but the majority died after 2 to 4 days after release. Only 20 plants derived from tetraspores were grown successfully for over a year in the laboratory. These haploid plants differed in morphology, color, size, and growth rates since the early developmental stages.	Paula et al. (1999)
Laboratory	BR tetrasporophyte	Tested conditions: temperature, light, salinity, and culture media (Provasoli = PES, Guillard and Ryther = F/2, and von Stosch = VS)	Growth rates of about $3\% \text{ day}^{-1}$ ($2.8\text{--}3.6\% \text{ day}^{-1}$) were obtained using PES, pulse-fed 24 h per week, or one-quarter strength 'F/2', and half-strength VS added continuously. Optimal conditions for growth were: 25 ± 2 °C, $170\text{--}210$ $\mu\text{mol photons m}^{-2} \text{s}^{-1}$, 14:10 h L:D cycle, and 32–35 psu	Paula et al. (2001)
Field	BR tetrasporophyte	Branches (3 g) produced in laboratory were transplanted to the sea and cultivated using floating system during 1 year (October 1995–1996)	Growth rates of transplanted branches were higher during the first 2 months ($6.5\text{--}10.7\% \text{ day}^{-1}$) and decreased in subsequent months ($4.5\text{--}8.2\% \text{ day}^{-1}$). Seasonal variation of GR was related to seawater temperature	Paula et al. (2002)
Field	BR tetrasporophyte	Branches were cultivated using floating system during 48 months (October 1995–December 1996). Factors analyzed: temperature, salinity, rainfall, and number of sunny hours	Growth rates of $3.6\text{--}8.9\% \text{ day}^{-1}$ correlated to seawater temperature: mean monthly values from 19.9 to 29.0 °C and extreme values from 17.0 to 33 °C low salinity and rainfall affected negatively the GR.	Paula and Pereira (2003)
Laboratory	BR tetrasporophyte	Experimental design: Temperatures 15, 18, 21, 24, 27, and 30 °C X PFDs 50, 100, and 150 $\mu\text{mol photon m}^{-2} \text{s}^{-1}$	<i>K. alvarezii</i> grew in temperatures from 18 to 30 °C but died at 15 °C, and the highest growth rate was $5.7\% \text{ day}^{-1}$ at 30 °C and 100 $\mu\text{mol photon m}^{-2} \text{s}^{-1}$	Bulboa and Paula (2005)
Field	BR, RE and GR tetrasporophytes, and EP	Cultivation from February to December 2001	Growth rates of brown, green, and red tetrasporophytes were similar ($2.5\text{--}6.6\% \text{ day}^{-1}$) but higher than EP gametophytes ($1.3\text{--}4.8\% \text{ day}^{-1}$). For all strains, including EP strain, GR were higher from February to May and decreased from July to December	Hayashi et al. (2007a)

(continued)

Table 9.1 (continued)

Cultivation location	Strain(s)	Cultivation conditions	Physiological responses	References
Field	BR female gametophyte	Tested conditions: cultivation period (28, 44, and 59 days), depth (surface, 0.5 m and 1.0 m), and planting density (24, 12, and 8.4 plants m ⁻²) during the winter	Growth rates ranged 5.2–7.2% day ⁻¹ , and the optimal conditions for growth were as follows: cultivation for 28 days, at 0–0.5 m depth and planting density of 12 and 8.4 plants m ⁻²	Hayashi et al. (2007b)
Field and laboratory	BR, RE and GR tetrasporophytes, and EP	Laboratory experiments: temperatures (20, 25, and 30 °C) X PFDs (50, 100, 150 μmol photon m ⁻² s ⁻¹) photoperiods (LD cycles): 14:10 h, 12:12 h, and 10:14 h. Field experiments: substratum for tetraspore settlement was 25 cm ² acrylic plates (<i>n</i> = 84). Forty-two cords were hung vertically from the culture raft, each with two settlement plates at 50 and 150 cm depth	Tetraspore survival (%) of brown (13%), red (3%), and green (5%) strains declined with time, showing high mortality after 20 days in all temperatures, PFDs, and photoperiods. Recruitment of tetraspores did not occur in the field; what indicated that the establishment of <i>K. alvarezii</i> via spore production in the natural environment is rather remote	Bulboa et al. (2008)
Field	BR tetrasporophyte	Cultivation in raft anchored in the bay, density of 6.7 plants m ⁻² for 30 days (May 2014)	Growth rate of 6.3% day ⁻¹ and productivity of 50.9 g m ⁻² day ⁻¹ (corresponding to 18.3 kg m ⁻² year ⁻¹)	Roldán et al. (2017)
Field	BR, RE and GR tetrasporophytes, and EP	Cultivation in raft anchored in the bay, from May to June 2013	Growth rates ranged from 3.8 to 6.2% day ⁻¹ , and productivity ranged from 15.9 to 46.0 g m ⁻² day ⁻¹	Masarin et al. (2016); Oliveira et al. (2019)
Field	BR, RE and GR tetrasporophytes, and EP	Cultivation in raft anchored in the bay, density of 6.7 plants m ⁻² , from August 2013 to July 2014	Growth rates differed among strains: 5.0–8.5% day ⁻¹ (RE), 5.3–9.0% day ⁻¹ (BR), 4.8–9.0% day ⁻¹ (GR), and 3.3–6.6% day ⁻¹ (G11)	Solorzano-Chavez et al. (2019)

BR, RE, and GR = brown, red, and green strains, respectively; EP—gametophyte previously cited as “G11,” which was originated from tetraspore germination

January 2018, and vegetative dispersion and reproductive development were evaluated in four strains, three tetrasporophytes (brown, green, and red) and one gametophyte (EP) (Araújo et al. 2020b). No evidence of vegetative dispersion and/or establishment in Ubatuba Bay was observed. Also, reproductive structures were not found during the monitoring period, indicating the low invasive potential of *K. alvarezii* in this region (Araújo et al. 2020b).

9.3.5 Chemical Composition, Utilization, and Biotechnological Potential

The high and sustainable biomass production from *K. alvarezii* cultivation in Ubatuba Bay has provided raw material for chemical analysis and for biotechnological applications, such as biofuel production (Masarin et al. 2016; Roldán et al. 2017; Oliveira et al. 2019; Solorzano-Chavez et al. 2019), production of monomeric sugars for bioethanol or fine chemical production (Paz-Cedeno et al. 2019), hydrogen production (Rodrigues et al. 2019; Fonseca et al. 2020), antioxidants (Araújo et al. 2020a, 2022), and biofertilizers (Gelli et al. 2020).

An alternative found to leverage São Paulo’s algiculture would be the new processing of fresh macroalgae by the

producers themselves (Gelli and Barbieri 2015). Recent studies were carried out on the extraction of agricultural biostimulant using equal parts of the four cultivated strains (original brown, green, and red tetrasporophytes and the EP gametophyte (Gelli et al. 2020)). The results can be seen in Table 9.3.

9.4 Perspectives and Final Remarks

The commercial cultivation of *Kappaphycus alvarezii* was prohibited on the north coast of São Paulo. However, in June of 2022, commercial cultivation was finally authorized by environmental legislation in São Paulo State (Brazil 2020; São Paulo 2022).

In São Paulo State, algiculture activities can be expanded in a gradual and planned manner in the coming years (Gelli 2019). In this emerging phase, it will be necessary to develop new processing methods for the commercialization of fresh macroalgae directly by the mariculturist to stimulate the production chain at the artisanal level, followed by the development of the industrial production chain.

K. alvarezii color strains that originated spontaneously from the four initial strains need to be studied before they are made available to the production sector. More

Table 9.2 Carrageenan yield and properties of *Kappaphycus alvarezii* strains cultivated in Ubatuba Bay, São Paulo State, southeastern Brazil

Strain(s)	Extraction method	Carrageenan yields	Carrageenan characterization	References
BR, RE and GR tetrasporophytes and EP	Semi-refined and refined carrageenans were obtained from 20–50 g DW after alkaline transformation	Semi-refined carrageenan yields of EP gametophyte were higher (31–43%) than tetrasporophytic strains; all strains showed lower yields in May and higher from August to December Refined carrageenan yields were also higher in EP strain (15%–28%) than tetrasporophytic strains	Higher 3,6-anhydrogalactose contents were observed in the EP (30.62%) and green (24.51%) strains, while the brown strain showed the lowest values (7.45%). Sulfate contents varied from 23.08% to 33.48%, and gel strength varied from 688 to 926 g cm ⁻² , but these variations did not differ significantly among the strains	Hayashi et al. (2007a)
BR female gametophyte	Native carrageenan was extracted in distilled water, and alkali-modified carrageenan was produced by alkali transformation with isopropanol and KOH solution	Yields of native and alkali modified carrageenan varied, respectively, from 21 to 35% and from 20 to 32%, and the higher values were observed in samples cultivated for 28 days	The highest values of iota fraction, molecular weight, and gel strength of native and alkali-modified carrageenan were observed in samples cultivated for 59 days	Hayashi et al. (2007b)
BR and RE tetrasporophytes	Semi-refined carrageenan was extracted in distilled water using a biomass pretreated in cold alkaline solution	Yields varied between the strain: 63.5% (BR strain) and 60% (RE strain)	Carrageenans from BR and RE strains presented, respectively, 42.6 and 46.6% (galactan), 24.2 and 28.5% (ash), 0.3 and 0.3% (protein), 1.1 and 1.4% (insoluble aromatics), and 13.3 and 14.0% (sulfate groups).	Masarin et al. (2016)
BR tetrasporophyte	Semi-refined carrageenan was extracted in distilled water using a biomass pretreated in cold alkaline solution	Yield of 64.5 ± 1.2%	Galactan and sulfate contents were 44.3% and 13.8%, respectively	Roldán et al. (2017)
BR, RE and GR tetrasporophytes	Refined carrageenan was extracted in distilled water using a biomass pretreated in cold alkaline solution	The yields varied among strains: 53.2–69.1% (brown strain), 59.5–63.1% (red strain), and 56.1–63.2% (green strain). Higher yields in samples cultivated during the summer-autumn season	The main components found in the biomass: 26.9–33.1% (galactans), 12.7–16.6% (glucans), 14.4–17.1% (ashes), 1.8–5.6% (proteins), 2.5–4% (insoluble aromatics), 10.7–13.8% (sulfate groups), 1.5–8.6% (lipids), and 77.7–89.7% (sum of components) (w/w)	Solorzano-Chavez et al. (2019)

BR, RE and GR = brown, red and green strains, respectively; EP—gametophyte previously cited as “G11”, which was originated from tetraspore germination

Table 9.3 Chemical characteristics of extract composed by equal parts of the brown, green, red, and EP strains of *Kappaphycus alvarezii* cultivated in Ubatuba Bay (Gelli et al. 2020)

	Values
pH	5.97
Density	0.95 g mL ⁻¹
Organic matter	6.89 g L ⁻¹
Soluble mineral residue	36.53 g L ⁻¹
Total Carbon	3.83 g L ⁻¹
Total mineral residue	36.57 g L ⁻¹
Mineral insoluble residue	0.036 g L ⁻¹
Potassium (K ₂ O)	20.17 g L ⁻¹
Total Carbon	3.83 g L ⁻¹
Total Nitrogen	0.42 g L ⁻¹
Sulfur (S)	0.35 g L ⁻¹
Calcium (Ca)	0.25 g L ⁻¹
Magnesium (Mg)	0.2367 g L ⁻¹
Phosphorus (P ₂ O ₅)	0.090 g L ⁻¹
Iron (Fe)	0.0037 g L ⁻¹
Zinc (Zn)	0.0010 g L ⁻¹
Manganese (Mn)	0.0003 g L ⁻¹
Copper (Cu)	0.0 g L ⁻¹

investigations will be needed to characterize these new strains, such as the amount of refined and semi-refined carrageenan, biofertilizer yield, temperature resistance, epibionts, growth rate and yield, as well as characterization of biomolecules of economic interest.

The implanted biobank (SLFI) and the strain bank in a natural environment (EMFFI) will guarantee the deposit of these variants and the supply of different strains adapted to the needs and conditions of each farmer.

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