The effect of abiotic parameters and nitrogen enrichment on growth rates and biochemical composition of the Eastern Mediterranean alga *Codium taylorii*

By: Itai Kolsky

THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE

REQUIREMENTS FOR THE MASTER'S DEGREE

University of Haifa

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The Leon H. Charney School of Marine Sciences

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Supervised by: Prof. Tali Mass

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I would like to thank my thesis supervisors, Professor Tali Mass and Professor Muki Shpigel, for your patience, guidance, and support. I have benefited greatly from your wealth of knowledge and meticulous editing. I am incredibly grateful that you took me on as a student and continued to have faith in me over the years.

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Thank you.

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Itai Kolsky

Abstract

Seaweeds have been suggested as a sustainable and potentially excellent source of edible proteins and other bio-materials, such as pigments and polysaccharides. Due to its often rough sea and oligotrophic conditions, the Levant basin represents a challenge for establishing sea-based commercial cultivation of seaweeds. Nevertheless, algae species that thrive in such harsh conditions may possess unique properties that could well justify their culture. In this work, the local alga *Codium taylorii* (class Ulvophyceae) was examined in a multifactorial setup of light, salinity, and temperature levels, and its growth rates and biochemical performance were recorded. In terms of growth rates and protein content, *C. taylorii* best performed when grown at 100µE light intensity, reaching almost 2% daily growth and 12% protein. However, a colder environment (15ºC) and lower light intensity (50µE) were more efficient in preventing epiphyte settling and keeping algae typical rigidity. A high nutrient enrichment, with great emphasis on Nitrogen (above 280 µM), had a substantial morphological impact on the alga. *C. taylorii* tolerated a wide range of salinities (30 – 50‰) and temperatures (15-25°C) remarkably well. These features would allow combining its cultivation in controlled land-based facilities with commercial fish farms' effluents, making *C. taylorii* a valuable secondary aquaculture product.

This novel work regarding the Levant basin can lay the baseline for further research toward new sustainable products in the local algaculture.

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Introduction

New and sustainable protein sources and natural alternatives for other less traditional materials for the food industry, such as colorants and saccharides, are in demand worldwide[1]. This new demand comes as a consequence of agricultural lands is being converted to other uses, combined with the increasing need for food to feed the world's rapidly growing population[1], lack of fresh water in warm regions[2], and the growing ecological awareness for the need to protect nature[3]. Smart and intensified inland agriculture is one solution[2]. Another solution implemented in many regions is the use of seawater bodies for mariculture, especially where freshwater scarcity has become a great concern[2,3]. However, one such solution, fish-cages, became an ecological issue in some regions[4]. When fish cages become an ecological issue, bio-filtering organisms, such as seaweeds, were suggested as a cost-effective solution for obtaining food sources with lower environmental impact[5]. The ocean's potential is yet to be fully exploited, not just by agricultural growing areas but by new organisms that can become a valuable source for proteins and other raw materials[5–7], such as seaweed. Seaweed culture occupies around 25% of total worldwide aquaculture yield[8–10]. However, most of it remains with extensive productivity, and it heavily relies on seasonal culturing in natural water basins or harvesting part of the natural community, reaching about 31 million tons annually in 2015[9,10]and rising to almost 36 million tons in 2019[3,11]. In general, seaweed includes macroscopic multicellular marine algae, primitive plants with no mosses, ferns, roots, or other differentiation known in higher plants. The formed structure of seaweeds is called a thallus. The thallus can change morphology depending on environmental factors and form higher plant-like organs, but the tissue structure will remain similar[12]. Throughout the Levant basin, seaweed culture is not feasible due to its oligotrophic conditions[13] and due to the often rough sea conditions. Land-based culturing can be more intensive and possesses great advantages such as all-year production and high-quality products[1,14,15]. Seaweed intensively cultivated in industrial land-based facilities can be cost-effective if they are utilized as a bioremediation agent for growing fish and shellfish (such as in Integrated Multi-Trophic Aquaculture (IMTA) facilities[16]) or if high-value raw materials such as polyunsaturated fatty acids, proteins, saccharides, and pigments can be extracted[17,18]. From a general perspective, adopting the biorefinery approach,

where the process of growing algae is sustainable, and the biomass can produce a variety of marketable substances, can enhance the industry's profitability. Thus, it is being adopted by producers and growers[1,17,19]. The Levant basin algaculture is very limited and relies on very few species (mainly *Gracilaria sp.* and *Ulva sp.*). Introducing new local species to the alga industry can help establish new valuable protein sources and other raw materials. One such species can be *Codium* sp., which is very abundant in the eastern Mediterranean. Even though *Codium* sp. is not entirely new to the industry, no industrial facility is known to grow it on the Levant Basin. There are *Codium sp.* products on the East Asia markets as food and food additives with higher market value compared to *Porphyra* (also known as *Pyropia*) and *Undaria*[20,21], which have high demand. Therefore, it could be more easily supplied to Eastern markets and possibly introduced to Western markets.

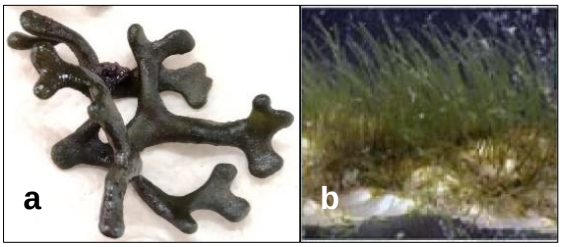

Figure 1 **a-b** - *Codium sp.* **a**. branched thalli **b**. medullary filaments [20]

Codium (phylum Chlorophyta) is one of the widest distributed marine alga genus worldwide, with approximately 150 species[22]. *Codium* species have at least two morphologically different thalli: spongy (Figure 1a) and filamentous (Figure 1b). The spongy polymorph individuals present a cylindrical thallus, regularly dichotomous with elastic consistency and a dark green color. This seaweed can grow up to 30 cm in length, and the branches, thin and cylindrical, can grow up to 8 to 10 millimeters in diameter. Terminal segments are often long, with apices rounded or slightly pointed. It commonly presents numerous hairs (or hair scars) below the apex[23]. In contrast, the thalli of the filamentous form fine-branched filaments (Figure 1b). This morphology formed from isolated utricles, medullary filaments, zygotes, and parthenogenetic female gametes of the spongy thalli[24]. *Codium* has a diploid life cycle with gametic meiosis. In sexual reproduction, zygotes are obtained by merging the haploid gametes[25].

Several studies showed that *Codium sp.* consists of interesting compounds such as novel sterols, carotenoids, halogenated metabolites, and other bioactive compounds with high potential market values[26,27]. Another research conducted on *C. fragile* spp. showed that the ideal depth to cultivate this seaweed in the open sea

differs depending on its life stage[20]. All factors can be manipulated with ease on an in-land growing facility.

Using a locally distributed seaweed species has two significant advantages; firstly, we are not introducing new species to the local ecosystem. Secondly, the often rough condition that the local species grow in may affect its biochemical composition or growing mechanism[28], possibly enhancing its commercial value.

Research objectives

To examine the effect of physical and chemical environmental parameters (temperature, salinity, light intensity, and dissolved nitrogen levels) on the performances (growth rate, yield, and biochemical composition) of *Codium sp.* from the eastern shore of the Levant basin (Israeli coast), ultimately offering a base for developing local industrial cultivation.

Specific objectives:

1. Characterize local *Codium sp.* from the Israeli coast, using morphological and molecular tools.
2. Examine the effect of abiotic parameters (temperatures, salinity, light intensity, and nutrient levels) on growth rates and yield, biochemical composition, nutrients uptake, and polymorphism.
3. Examine the biochemical composition of *Codium sp.* during the succession period (spring) in terms of total protein and pigments composition.

Working hypothesis

By manipulating growing conditions (abiotic and nutrients), we will be able to control seaweed growth rate, yield, biochemical composition, and nutrient uptake.

In particular, we expect:

Higher growth rates when high nitrogen concentration and strong light intensity are supplied.

Higher content of carotenoids accumulation at high-temperature conditions, strong light intensity, and Nitrogen depletion.

Methods

The experiments were carried out during spring 2021 at the Morris Kahn research station of the Leon H. Charney School of marine sciences of the University of Haifa.

Algae collection

The first batch of seaweeds was harvested in August 2019 from the prefilter pools of the Soreq desalination plant and used for the preliminary observation (Figure 2).

Figure 2 - Codium sp. collection from the pre filter of the Soreq desalination plant, August 2019.

Seaweed samples for the experiment were collected at 5-12 m depth, from the abrasion platforms near Kibbutz Sdot-Yam, Israel, during October 2020. All samples were cleaned from debris and epiphytes by hand, using reverse osmosis water (RO). The clean algae samples were acclimated for at least four weeks (prior to the experiment) in one acclimation tank with artificial seawater (ASW) prepared using RO with sea salts (Red Sea Reef Salt) and ambient temperature and light at the time of harvest (20°C, 50µE).

Preliminary observations

A five-week preliminary experiment was conducted to define basic physical parameters suitable for growing different forms of *Codium* sp. indoor. *Codium* sp. samples were divided into four different morphology forms (sprouting like thallus, self-fragmented thallus, typically branched thallus, and spherical branched thallus) and set into three replicates each. All growing tanks received the same conditions, using ASW in a recirculated system, 50µE, and 25°c (Figure 3). The seaweeds weighed once a week, and cultivation tanks were cleaned to prevent contamination. A small amount of commercial fertilizer (Haifa group

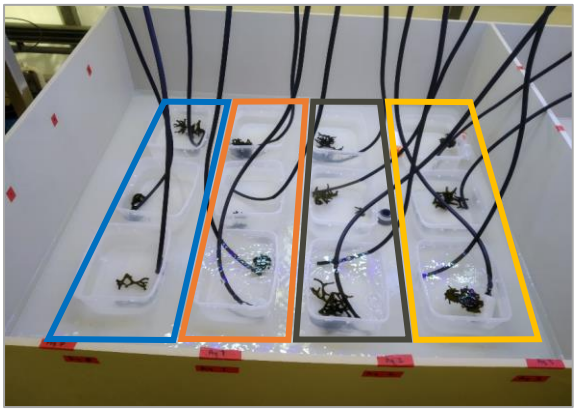

Figure 3 - Preliminary experiment setup, used to define the basic growing condition. sprouting like thallus, self-fragmented thallus, normally branched thallus, and spherical branched thallus (blue, orange, grey, and yellow, respectively)

Deshen Kol 20-20-20) was added weekly to achieve 15µM of Nitrogen (as Ammonium and Nitrate). After five weeks of the experiment, the temperature dropped accidentally from 25°C to 16°C for 48 hours and then increased to 18°C. The observation continued for two more weeks, under a temperature of 18°C. During this preliminary test, no other parameters were measured except wet weight and morphology changes.

Species Identification

For *Codium* species identification, 14 random samples were taken out for DNA analysis. Two genes were used for species identification, the rubisco large unit (RbcL) [29] and elongation factor tu (TufA)[30]. In order to extract DNA, 50mg wet weight from each sample was shredded using a scalpel, and DNA was extracted using the CTAB protocol of Bioline ISOLATE II Plant DNA Kit (cat no. BIO-52069). Genes were amplified using specific primers (Table 1). Products length and concentrations were validated using agarose gel and nanodrop (ThermoFisher NonoDrop one) and then sent for sequencing at Hy Laboratories Ltd (hylabs). Sequences were aligned using Mega- X software and BLAST on the NCBI website.

Table 1 – Primers and PCR conditions used to classify the collected seaweeds

Fragment	Primers		PCR cycling conditions	TAQ kit	Target size
RbcL	Fwd:	AACTGAAACTAAAGCAGGTGCAG	45" 94°C, 35X(15" 94°C, 20" 53°C, 45" 72°C), 2' 72°C	GoTaq G2 Green Mix + 1µL BSA	600 bp
	Rev:	GCATRATAATAGGTACGCCRAA			
TufA	Fwd:	GGNGCNGCNCAAATGGAYGG			750 bp[31]
	Rev:	CCTTCNCGAATMGCRAAWCGC			

Experimental system setup

The experiment was conducted in 52, 5L buckets submerged in a 1.2X2.8m water table for temperature control, equipped with an aeration system and eight configurable dedicated LED systems (AquaDecor 100W) (Figure 4 **B**). Using ASW (R.O. water and Red-sea reef salt) enriched with modified F/2 (Mod. F/2) growth medium to include both ammonium and nitrate as a source of Nitrogen ($(NH_4)_2SO_4$ 18.5mg/L, KNO_3 28.3mg/L, KH_2PO_4 2.5mg/L, Na_2-EDTA 4.16mg/L, $FeCl_36H_2O$ 3.15ml/L, $CuSO_45H_2O$ 0.01ml/L, $ZnSO_47H_2O$ 0.02mg/L, $CoCl_26H_2O$ 0.01mg/L, $MnCl_24H_2O$ 18mg/L, $Na2MoO_42H_2O$ 0.006mg/L). The expected nitrogen uptake, calculated from the preliminary observation's specific growth rate (SGR), was 70µM/L/week, and Mod. F/2 concentrated stock was added to enrich the media four times the expected uptake (280µM/L) from each source. The effects of the most

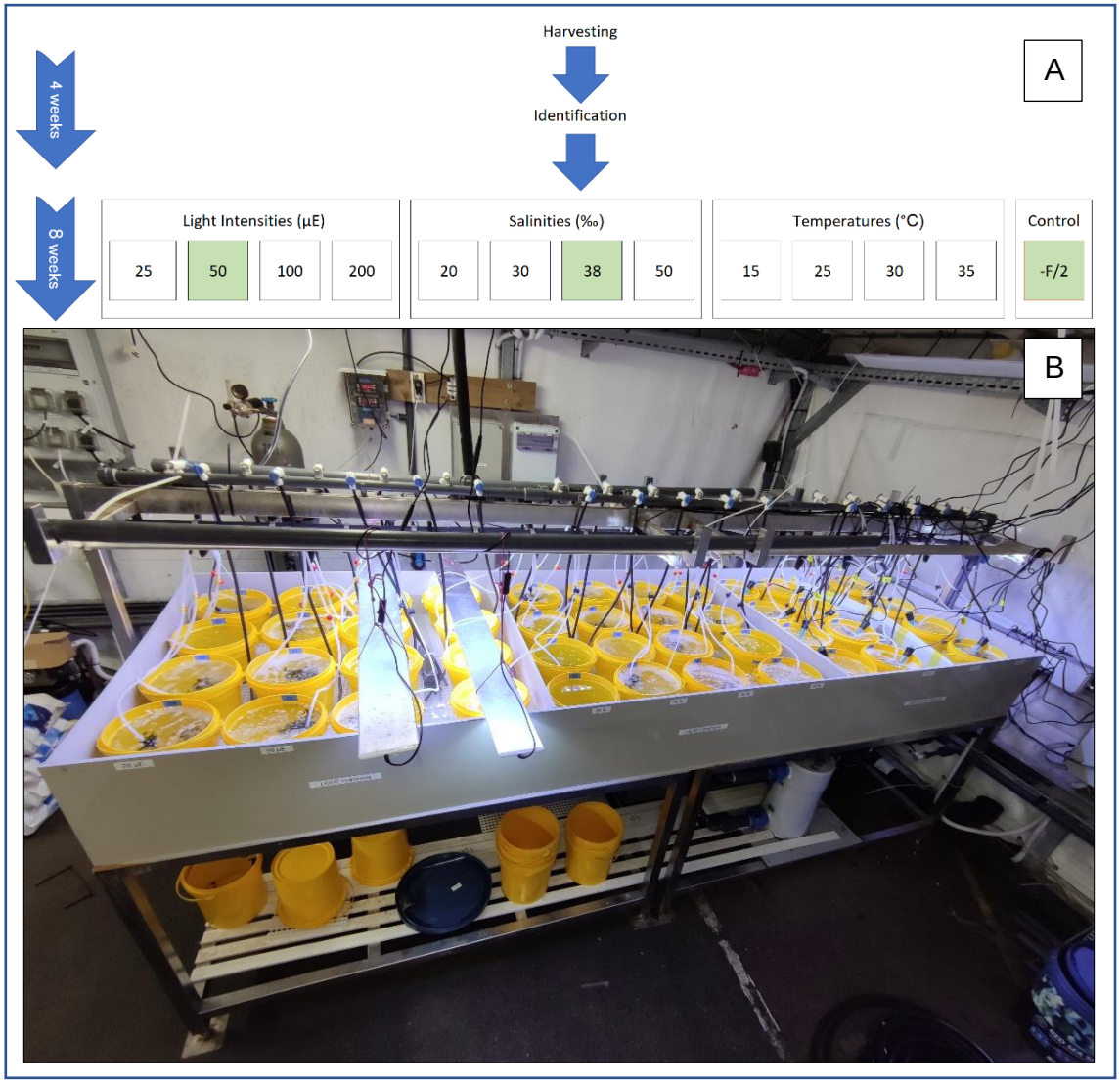

Figure 4 - Experiment system setup design, 15 treatments, 4 replicates each.
***A**- Flowchart of the conceptual design. **B**- photo of the actual system at Morris Kahn research station*

important environmental factors affecting algal growth: light intensity, salinity, and temperature were examined. Each factor was divided into four levels of treatments with four replicates (Table 2, Figure 4 **A**), control factors set according to the preliminary trial (38‰, 20°c, 50µE, Mod. F/2). Four replicates of control conditions with no nutrients (C-F/2) were added to the trial to better understand the effect of nitrogen enrichment on the seaweeds and stand as a negative control.

Table 2 – Growth conditions for all treatments, yellow marking indicate the changing levels of the examined factor, and red letters indicate the control treatments.

Treatment	Temperature (°C)	Salinity (‰)	Light intensity (µE)	Nutrients	Volume (L)	Initial stock (gr)	Replicates
Temperature	15	38	50	Mod. F/2	5	35	4
	25	38	50	Mod. F/2	5	35	4
	30	38	50	Mod. F/2	5	35	4
	35	38	50	Mod. F/2	5	35	4
Salinity	20	20	50	Mod. F/2	5	35	4
	20	30	50	Mod. F/2	5	35	4
	20	38	50	Mod. F/2	5	35	4
	20	50	50	Mod. F/2	5	35	4
Light Intensity	20	38	25	Mod. F/2	5	35	4
	20	38	50	Mod. F/2	5	35	4
	20	38	100	Mod. F/2	5	35	4
	20	38	200	Mod. F/2	5	35	4
Control C-F/2	20	38	50	-	5	35	4

The growing period was eight weeks, and water was replaced weekly to prevent possible starvation or contamination. Every week before water replacement, 50ml of 0.22µm filtered water from fresh growth media and all growing tanks were collected and kept at -20°C for further chemical analysis. Algae photosynthetic activity was measured at four different time points (T_0, T_{+2}, T_{+6}, T_{+8}). Biomass was weighed once a week for growth rate calculation. At the end of the experiment, all samples were lyophilized, weighed, and kept at -80°c for correlation of dry and wet weight, protein content, and pigment analysis.

System controls

Environmental factors (temperatures, pH, light intensity, and salinity) were logged using loggers (Onset HOBO pendant mx2202, fourtec MicroLite USB logger connected to Thermo Scientific Alpha 190 pH controller, Apogee SQ-500 PAR with microCache logger) and manually. At the end of the experiment, logged data were downloaded from the sensors for analysis. Throughout the experiments, pH was controlled (8.2±0.2) using CO_2 injected into the water using an aeration stone. The different temperatures were controlled using GHL ProfiLux 4 controller with dedicated cooler and aquarium heaters (NEWA Therm 150w) for the temperature treatments. Water evaporation was compensated daily using DDW to prevent salinity and nutrient concentration changes.

Samples analysis

Morphology and epiphytes

Four morphological factors were examined, rigidity, fragmentation, filaments formation ("hair"), and epiphyte succession. In order to quantify these qualitative factors, we created indexes from 1 to 5 as shown in Table 3 and Figure 5 and measured weekly. Linear correlations were calculated to compare the impact of the changes.

Table 3 – Indexes for morphological assessment

Index	1	5
Rigidity	No rigidity, flexible structure with no shape.	Typical structure, rigid bush-like formation.
Fragmentation	No fragmentation.	The thallus separates into many fragments.
"Hair" like filaments formation	Formation of a few "hair"-like filaments on the sample primary thallus.	An irregular thallus structure built mainly from separated filaments.
Epiphytes	No visible epiphytes on thallus.	The thallus is fully covered with epiphytes.

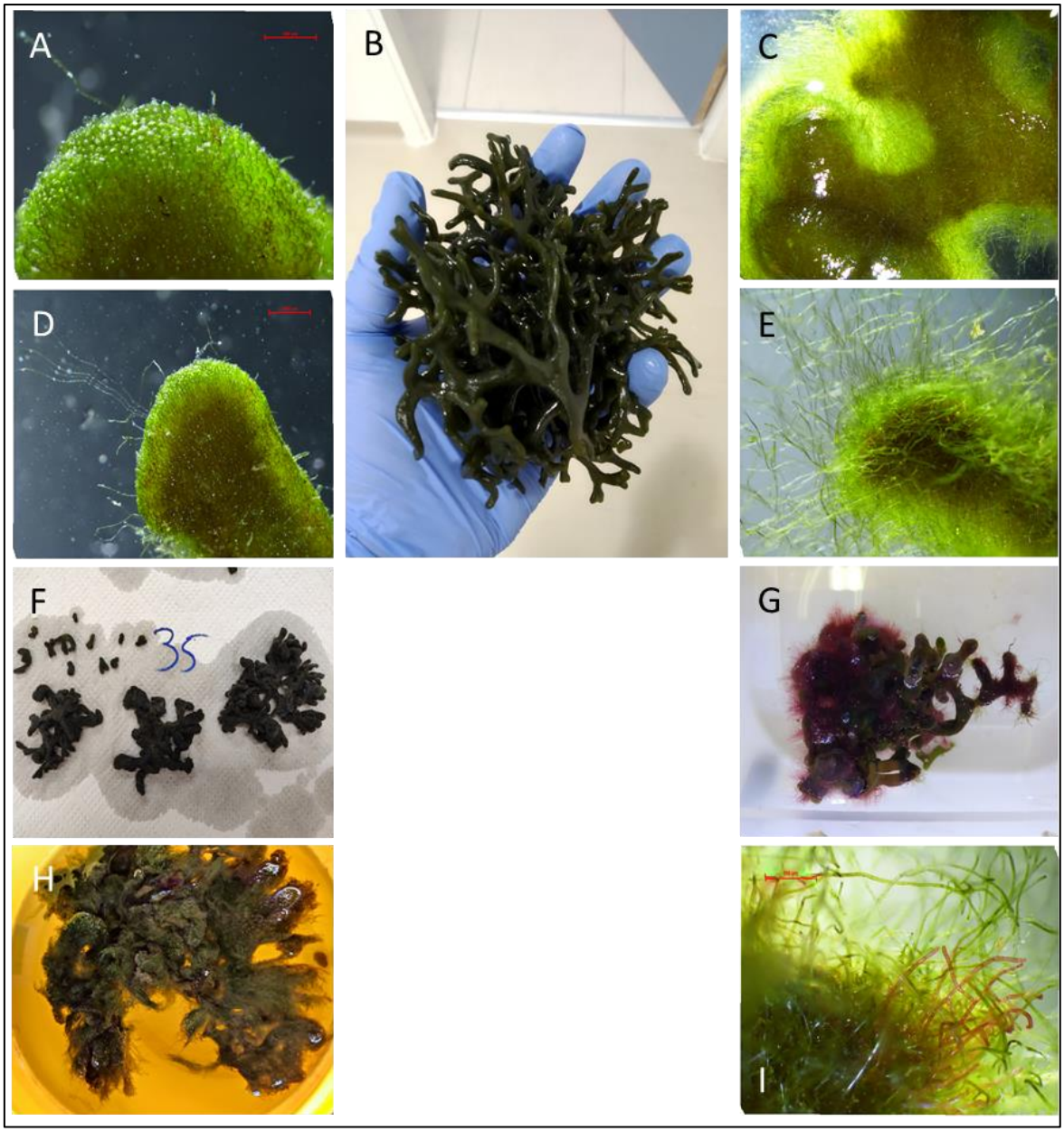

*Figure 5 – Codium taylorii different morphologies resemble the indexes used to describe the morphology changes: **A**, **D** – normal morphology with no "hair" formation or epiphytes. **B**- a typical structure of the seaweed. **C**, **E**- "hair" formation of filaments morphology. **F**- high fragmentation, **H**- unrigid formation, with no regular shape. **G**, **I** – epiphytes grow on the seaweed.*

Weight (dry and wet) measurement

Wet biomass of the seaweeds was weighed weekly, using a semi-analytic balance (Sartorius Entris 4202) with an accuracy of 0.01 gr (Figure 6). At the end of the experiment, all seaweeds were lyophilized, and dry biomass weight was measured using analytical Sartorius balance (Entris 2241-1S), with an accuracy of 0.001 gr. The specific growth rate (SGR) was calculated as growth per day[32] using Equation 1.

Figure 6 - Field measurements of wet weight and morphology

Equation 1 - SGR equation, specific growth rates.

$$SGR_{g\ g^{-1}\ day^{-1}} = \frac{\ln\left(wet\ weight_{week\ x+y}\right) - \ln(wet\ weight_{week\ x})}{\Delta days_{(week\ x+y)\ -(week\ x)}}$$

Lyophilization

All biomass samples were kept at -80°c before drying. Drying was done using lyophilizer (IIlshin FD5508 with PFEIFFER Penta 10 vacuum pump), at -52°c and 5mTorr for 48 hours. The dry biomass was kept at -80°c for further analysis.

Water Chemistry measurements

NH_4, NO_3, NO_2, and PO_4 were quantified using Macherey-Nagel Visicolor Eco kits Ammonium 3, Nitrate, Nitrite, and Phosphate, respectively. All reactions were read using a dedicated compact photometer (PF-12Plus). Weekly seaweed nutrients uptake was calculated as the delta of nutrients measures from the former fresh media and the current of all growing tanks (e.g., $\Delta NO_3\text{-}N_{Tank1\ week\ x} = NO_3\text{-}N_{Media\ week\ x\text{-}1} - NO_3\text{-}N_{Tank1\ week\ x}$), assuming neglectable uptake by other organisms.

Figure 7 - PF-12PLUS photometer used for water chemistry analysis

Protein quantification

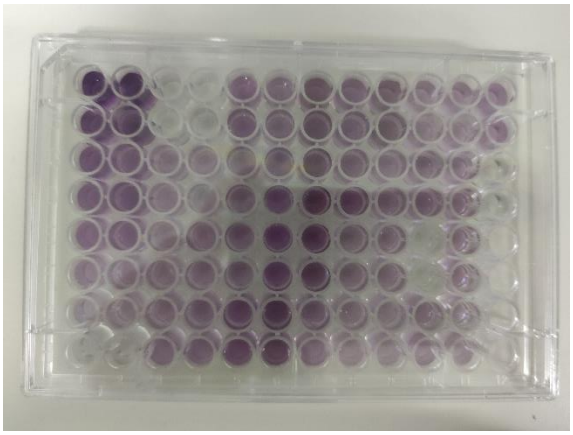

Figure 8 - 96 wells plate with samples colored with BCA for protein quantification

For the protein quantification, ten mg of dry biomass from each sample was hydrated for 1 hour in 3000µL H_2O and homogenized using CAT x120 homogenizer, then sonicated three times for 10 minutes in cold water and centrifuged for 5 min in 500g. The supernatant was then collected and used for total protein quantification using Bicinchoninic Acid Assay (BCA) method (Cyanagen QPro Bicinchoninic Acid Assay (BCA) kit). Bovine serum albumin (BSA) was used as a standard for calibration, following the kit manufacture protocol. The results were expressed as a percentage of dry weight (% DW).

Pigments analysis

Pigments were separated using UPLC (ACQUITY UPLC system with PDAeλ detector, Waters). C8 column, 1.7 µm particle size, 2.1 mm internal diameter, 50 mm column length (ACQUITY UPLC BEH, 186002877), 0.5 ml/min flow rate with column and injection heating to 50ºC and 30ºC respectively, following Daniel Sher's lab protocols based on LOV method[33]. For pigment separation, 10 mg of shredded lyophilized biomass was dissolved in 3000 µL of analytical grade MeOH for one hour in the dark, then sonicated three times for 15 minutes and kept overnight in the dark at 4°C. 1000 µL from each sample solution was filtered using 0.22µm PTFE filters to a dedicated vial with PTFE septum for UPLC autosampler and stored at -80°C until the analysis. UPLC data was analyzed using Empower 2 software.

Photosynthetic activity

Figure 9 - Actinic light over a sample of Codium during photosynthetic activity measurement using Walz Imaging PAM

During the experiment, photosynthetic activity was measured at four different time points (T_0, T_{+3}, T_{+6}, T_{+8}). Seaweeds from all treatments were dark acclimated for 30 minutes before measurement. Samples were measured using imaging PAM (Walz imaging PAM IMAG-MAXI with IMAG-K4 camera). The illumination method was set to reach 1250µE in 16 legs, each of 0.2 sec with 20 sec of relaxation time between, and an actinic saturating light (2700 µE). All seaweeds were sampled at three different points on the thalli. After photosynthetic activity measurements, all samples were returned to the experiment tanks for recovery. Data collected was used to calculate photosystem II (PSII) maximal quantum yield (F_v/F_m), initial slope of the light response curve (α), relative maximal electron transport rate ($rETR_{max}$), and minimum saturating irradiance (E_k).

Results

Preliminary observation

Preliminary observation showed no significant differences in growth rates between the polymorphs (Wilcoxon pairwise test, χ^2=5.299, df=3, P=0.151), with an average of 0.012g*g^{-1}*day^{-1}. The rapid temperature decrease caused the sprout-like formation of the thallus, exhibiting full cover of young branches in most of the algae samples.

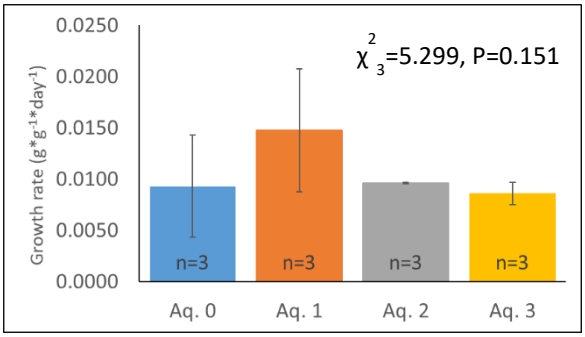

Figure 10 - Specific Growth Rates of the preliminary observation after six weeks. Sprouting like individuals- blue, typical form individuals- orange, fragmented individuals- grey, spherical fragments - yellow

Species characterization

For both RbcL and TufA gene sequences, out of 14 different algae samples, 13 samples showed >99% similarity to *Codium taylorii,* and only one was matched in 100% to *Codium parvulum* genes sequences. The last-mentioned showed a thinner and more transparent thallus morphology with long open dichotomous terminals that differ significantly from *C. taylorii* typical formation (Supplementary Figure 19, Supplementary Figure 20).

During the experiment, around the fourth week, an unknown general fault negatively affected all treatments, mainly regarding biomass accumulation. Therefore, all analyses were divided into two time points where it was possible (weeks 0-4 and weeks 5-8, respectively). This act was done to try and understand the strength of this event on the different treatments and minimize the effect on the data interpretation.

Growth rates and yields

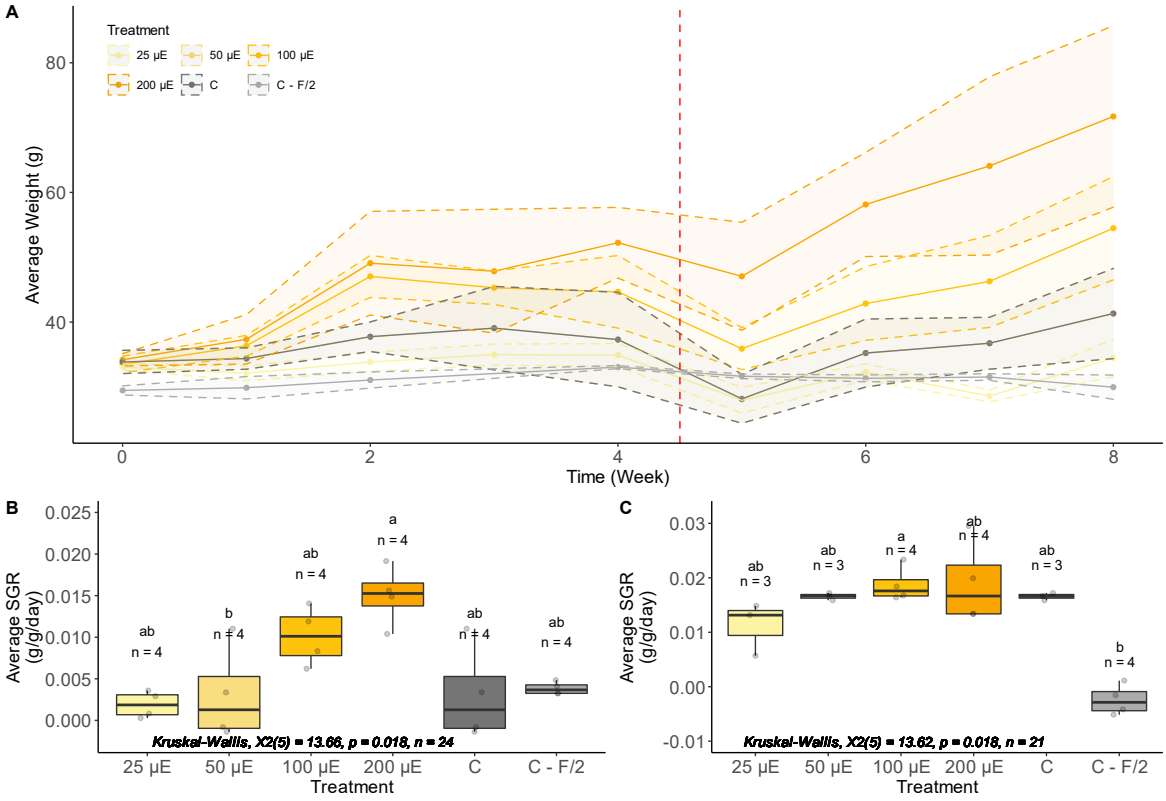

*Figure 11 - Light treatments growth curves and growth rates. **A**- wet weight gain during the experiment (the red dashed line defines the analyzed growing periods, and faded ribbons define SD), **B**- SGR during weeks 0-4 period (p=0.018), **C**- SGR during weeks 5-8 period (p=0.018). Different letters were attributed to significantly different treatments based on Dunn's test, n = number of samples.*

In the light treatments, the algae daily growth (Table 4) during weeks 0-4 were 0.2%, 0.3%, 1%, and 1.3%, respectively to the ascending light intensity levels and 0.3-0.4% on the control treatments, where a significant difference was observed only between the highest light treatment and C-F/2 treatment (Kruskal Wallis, χ^2= 18.1, df= 5. p= 0.0285) (Figure 11 **B**). During the 5-8 weeks period, daily growth rates were 1.1%, 1.6%, 1.8%, and 1.9%, respectively, at the ascending light intensity levels, while a decrease of -0.2% was observed in the C-F/2 treatment (Figure 11 **C**). Only the 100 µE treatment was significantly different from the negative control (C-F/2) (Kruskal Wallis, χ^2= 15.5, df= 5. p= 0.0085).

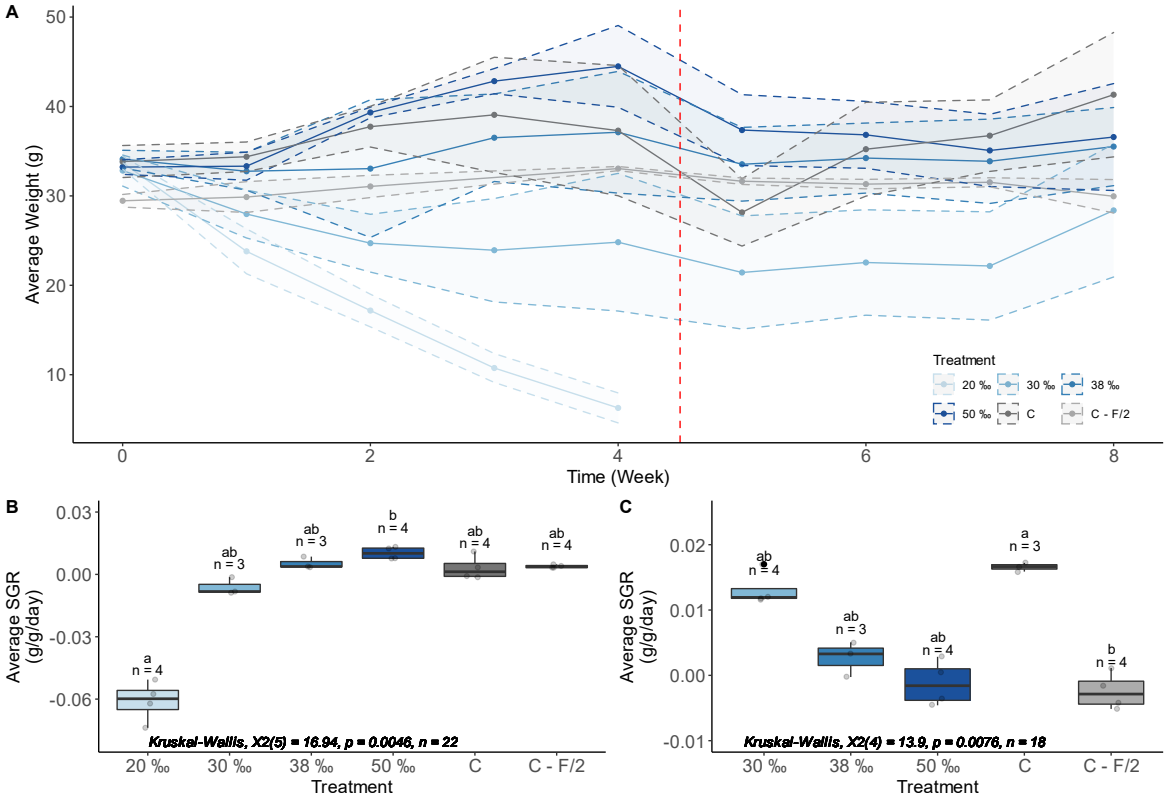

*Figure 12 - Salinity treatments growth curves and growth rates. **A**- wet weight gain during the experiment (the red dashed line defines the analyzed growing periods, and faded ribbons define SD), **B**- SGR during weeks 0-4 period (p=0.004), **C**- SGR during weeks 5-8 period (p=0.007). Different letters were attributed to significantly different treatments based on Dunn's test, n = number of samples.*

In the salinity treatments, daily growths (Table 4) during the period of 0-4 weeks were -6%, -0.6%, 0.5%, and 1%, respectively, in the ascending salinity levels, followed by the control treatments showed 0.3-0.4% daily growth. A significant difference was observed between the two highest salinity levels and the lowest (20‰) (Kruskal Wallis, χ^2= 18.1, df= 5. p= 0.0285). The algae on the 20‰ treatment did not acclimate to this salinity level and crashed on the fourth week (Figure 12 **B, A**). During the period of 5-8 weeks, the salinity treatments showed 1.3%, 0.27%, and -0.1% daily growth (30‰, 38‰, 50‰ respectively), compared to the control treatments that showed 1.6% daily growth for the control and -0.2% daily growth for the C-F/2 (Figure 12 **C**, **A**). The control showed a significant difference relative to C-F/2 and the 50‰ treatment, while C-F/2 showed a significant difference to 30‰ treatment as well (Kruskal Wallis, χ^2= 15.9, df= 4. p= 0.0316).

In the temperature treatments, daily growths (Table 4) were 0.2%, -1.25%, -4.6% at 15°C, 25°C, and 30°C, respectively, while at 35°C treatment, the algae did not survive the second week. A significant difference was recorded between the control

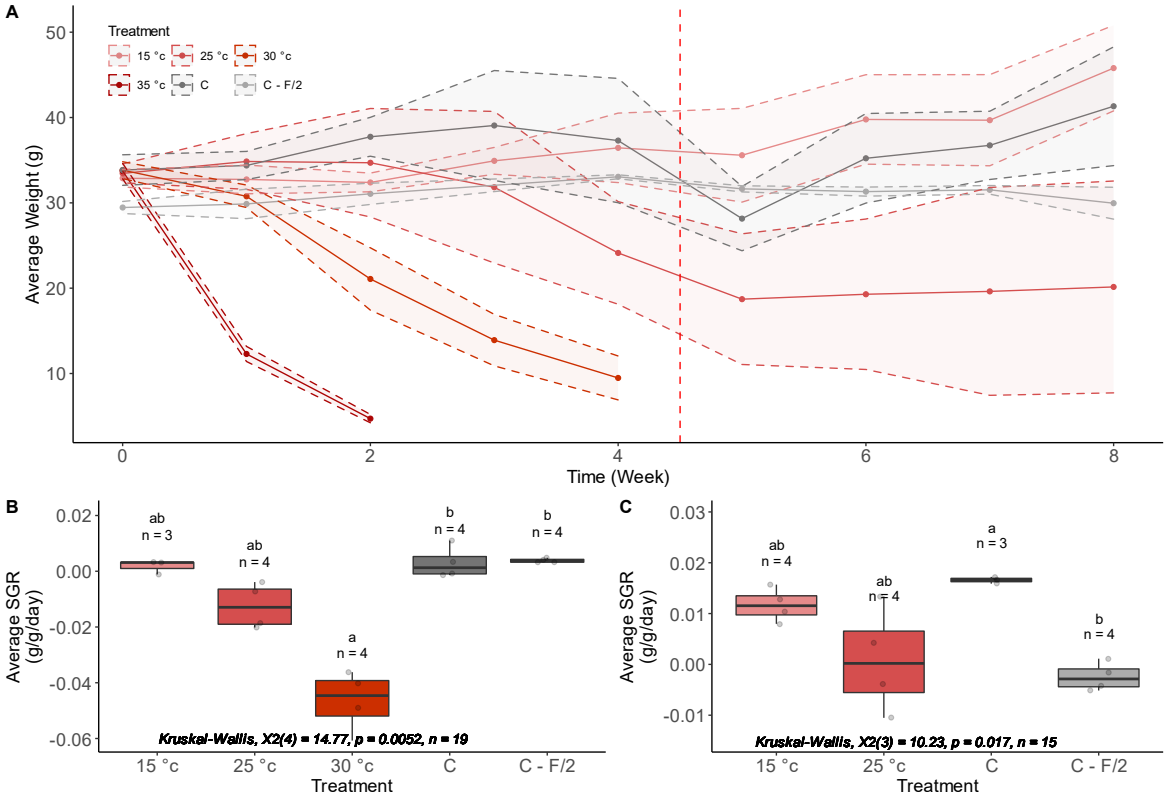

*Figure 13 - Temperature treatments growth curves and growth rates. **A**- wet weight gain during the experiment (the red dashed line defines the analyzed growing periods, and faded ribbons define SD), **B**- SGR during weeks 0-4 period (p=0.005), **C**- SGR during weeks 5-8 period (p=0.017). Different letters were attributed to significantly different treatments based on Dunn's test, n = number of samples.*

treatments and the 30°C treatment (Figure 13 **B, A**) (Kruskal Wallis, χ^2= 14.8, df= 4. p= 0.0051). During the 5-8 weeks period, the algae at 15°C reached 1.2% and at 25°C reached almost 0.1% daily growth. The control treatment was found to be significantly different from the 25°C and the C-F/2 treatments (Figure 13 **C, A**) (Kruskal Wallis, χ^2= 11.5, df= 3. p= 0.0091). The algae at 30°C treatment did not survive after the fourth week.

*Table 4 – Average specific growth rates (SGR) for all treatments for the two periods of the experiment. The average numbers ± SD expressed in gr*gr^{-1}*day^{-1}, numbers in brackets express n.*

Treat. Type	Treatment	T_{0-4}	T_{5-8}
Light	25 µE	0.0019±0.0016 (4)	0.0112±0.0049 (3)
	50 µE	0.0031±0.0057 (4)	0.0166±0.0007 (3)
	100 µE	0.0101±0.0035 (4)	0.0187±0.0032 (4)
	200 µE	0.015±0.0036 (4)	0.019±0.0076 (4)

Treat. Type	Treatment	T_{0-4}	T_{5-8}
Salinity	20 ‰	-0.061±0.0097 (4)	
	30 ‰	-0.0061±0.0042 (3)	0.0131±0.0026 (4)
	38 ‰	0.0053±0.0029 (3)	0.0027±0.0027 (3)
	50 ‰	0.0103±0.003 (4)	-0.0012±0.0035 (4)
Temperature	15 °C	0.0018±0.0025 (3)	0.0117±0.0033 (4)
	25 °C	-0.0125±0.0081 (4)	0.0008±0.0103 (4)
	30 °C	-0.0465±0.0109 (4)	
Control	C-F/2	0.0039±0.0008 (4)	-0.0024±0.0028 (4)
	C	0.0031±0.0057 (4)	0.0166±0.0007 (3)

Morphology

Light treatments were divided into two treatment groups that performed similarly concerning rigidity and filaments formation (100µE with 200µE and 25µE with 50µE) with a much higher trend to rigidity and lower trend to filaments formation at the low light intensities (Table 5, Figure 14**a**, **b**). The epiphytes and fragmentation factors showed both similarities at the two lowest intensities, while trends got stronger when intensities increased, a difference between the 200µE to 100µE with a higher trend at the higher intensity (Table 5, Figure 14**c**, **d**).

Temperature and salinity treatments showed a significant difference between all treatments on all measured factors, with a higher trend at treatments that collapsed prior to the eighth week (20‰, 30°C, 35°C treatments). At 15° C and at the negative control (C-F/2), almost no impact was observed on the measured morphology factors throughout the experiment, except the fragmentation factor (Table 5, Figure 14**a**, **b**, **c**, **d**).

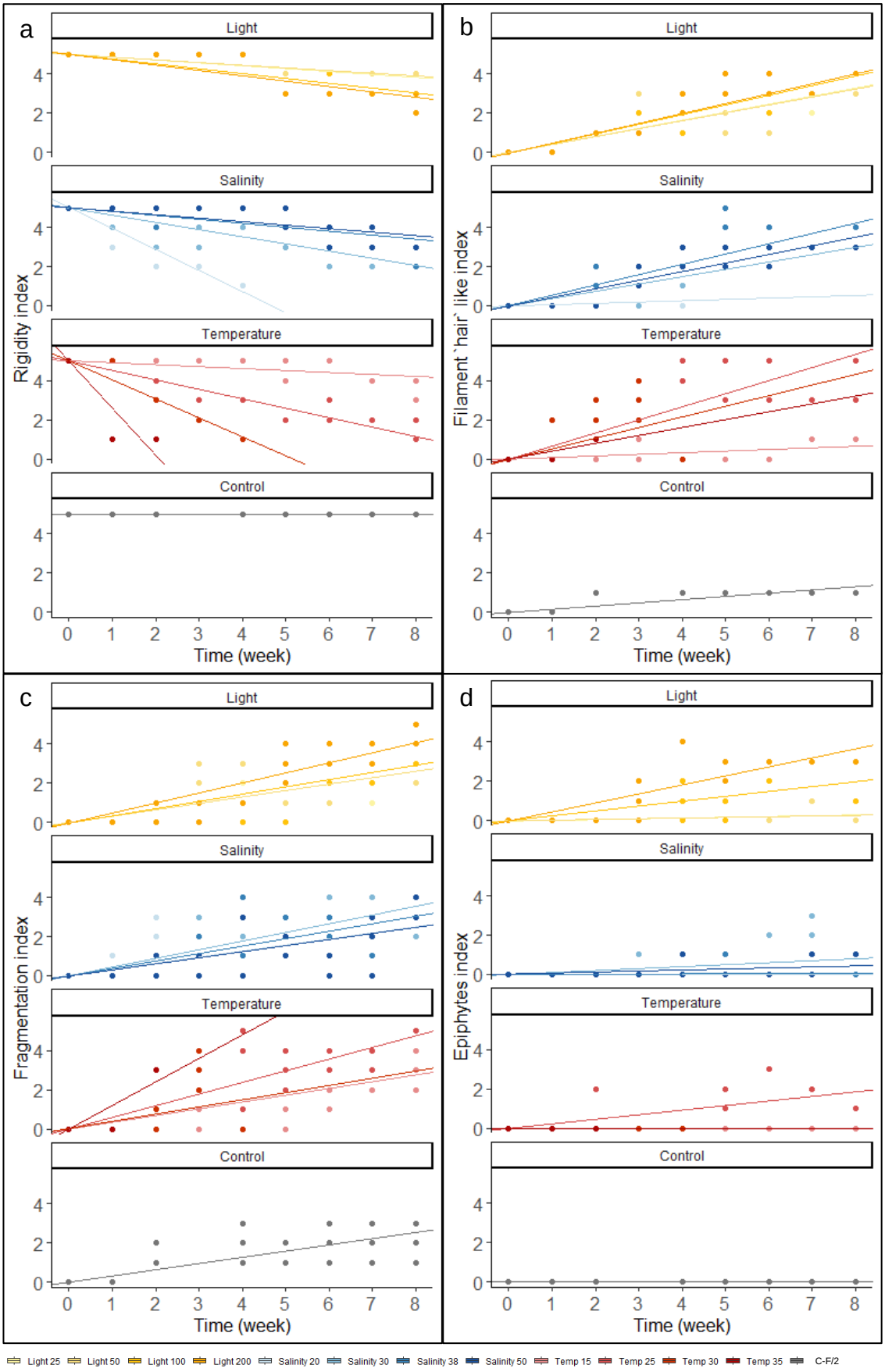

Figure 14 - Correlation between morphological factors examined against all treatments. ***a****- Rigidity factor,* ***b****- Filaments formation factor,* ***c****- Fragmentation factor,* ***d****- Epiphytes factor.*

Table 5 - Morphology indexes – a qualitative correlation between four morphology factors and the different treatments. When no coefficient is available, it means either the factor was not affected by the treatment, or either the treatment was crushed at an early stage of the experiment. All coefficients expressed with ± SD.

Measure	Treat. Type	Treatment	Coefficient	Adj. r^2	statistics
Rigidity index	Light	25µE	-0.147±0.012	0.80	F--11.91, p-0, n-36
	Light	50µE	-0.137±0.011	0.80	F--12.18, p-0, n-36
	Light	100µE	-0.248±0.02	0.81	F--12.62, p-0, n-36
	Light	200µE	-0.275±0.021	0.83	F--13.12, p-0, n-36
	Salinity	20‰	-1.075±0.048	0.96	F--22.24, p-0, n-16
	Salinity	30‰	-0.366±0.022	0.88	F--16.3, p-0, n-36
	Salinity	38‰	-0.201±0.019	0.76	F--10.72, p-0, n-36
	Salinity	50‰	-0.179±0.018	0.74	F--10.15, p-0, n-36
	Temperature	15° C	-0.099±0.016	0.52	F--6.36, p-0, n-36
	Temperature	25° C	-0.483±0.019	0.94	F--24.88, p-0, n-36
	Temperature	30° C	-0.967±0.041	0.96	F--23.47, p-0, n-16
	Temperature	35° C	-2.4±0.241	0.89	F--9.95, p-0, n-4
	Control	C-F/2	0±0	NA	n-32
Filaments 'hair' like index	Light	25µE	0.409±0.015	0.95	F-27.58, p-0, n-36
	Light	50µE	0.404±0.022	0.91	F-18.57, p-0, n-36
	Light	100µE	0.494±0.017	0.96	F-29.57, p-0, n-36
	Light	200µE	0.509±0.018	0.96	F-28.75, p-0, n-36
	Salinity	20‰	0.067±0.039	0.09	F-1.71, p-0.1036, n-16
	Salinity	30‰	0.371±0.018	0.92	F-20.37, p-0, n-36
	Salinity	38‰	0.526±0.029	0.90	F-17.96, p-0, n-36
	Salinity	50‰	0.44±0.021	0.93	F-21.41, p-0, n-36
	Temperature	15° C	0.081±0.013	0.52	F-6.33, p-0, n-36
	Temperature	25° C	0.667±0.047	0.85	F-14.16, p-0, n-36
	Temperature	30° C	0.542±0.143	0.40	F-3.78, p-0.0013, n-16
	Temperature	35° C	0.4±0.06	0.78	F-6.63, p-0, n-4
	Control	C-F/2	0.164±0.011	0.87	F-14.75, p-0, n-32
Fragmentation index	Light	25µE	0.328±0.031	0.76	F-10.69, p-0, n-36
	Light	50µE	0.328±0.03	0.77	F-10.89, p-0, n-36
	Light	100µE	0.369±0.025	0.86	F-14.71, p-0, n-36
	Light	200µE	0.513±0.026	0.91	F-19.48, p-0, n-36
	Salinity	20‰	0.308±0.1	0.30	F-3.1, p-0.0059, n-16
	Salinity	30‰	0.442±0.032	0.84	F-13.81, p-0, n-36
	Salinity	38‰	0.379±0.03	0.82	F-12.8, p-0, n-36
	Salinity	50‰	0.31±0.034	0.69	F-9.05, p-0, n-36
	Temperature	15° C	0.344±0.029	0.80	F-12.07, p-0, n-36
	Temperature	25° C	0.593±0.032	0.90	F-18.32, p-0, n-36
	Temperature	30° C	0.367±0.118	0.30	F-3.1, p-0.0059, n-16
	Temperature	35° C	1.2±0.181	0.78	F-6.63, p-0, n-4
	Control	C-F/2	0.318±0.025	0.83	F-12.7, p-0, n-32

Measure	Treat. Type	Treatment	Coefficient	Adj. r^2	statistics
Epiphytes index	Light	25µE	0.039±0.011	0.23	F-3.42, p-0.0016, n-36
	Light	50µE	0.034±0.01	0.22	F-3.33, p-0.0021, n-36
	Light	100µE	0.246±0.027	0.70	F-9.2, p-0, n-36
	Light	200µE	0.461±0.032	0.85	F-14.52, p-0, n-36
	Salinity	20‰	0±0	NA	n-16
	Salinity	30‰	0.1±0.026	0.29	F-3.92, p-0.0004, n-36
	Salinity	38‰	0.006±0.006	0.00	F-1.05, p-0.3001, n-36
	Salinity	50‰	0.053±0.011	0.36	F-4.61, p-0.0001, n-36
	Temperature	15° C	0±0	NA	n-36
	Temperature	25° C	0.232±0.029	0.64	F-8.12, p-0, n-36
	Temperature	30° C	0±0	NA	n-16
	Temperature	35° C	0±0	NA	n-4
	Control	C-F/2	0±0	NA	n-32

Water chemistry analysis and nutrients uptake

All Nitrogen measurements, except that of Ammonium (NH_4-N), were disqualified because of the significant interference of Nitrite (>0.16mg/L). Therefore, they were not used.

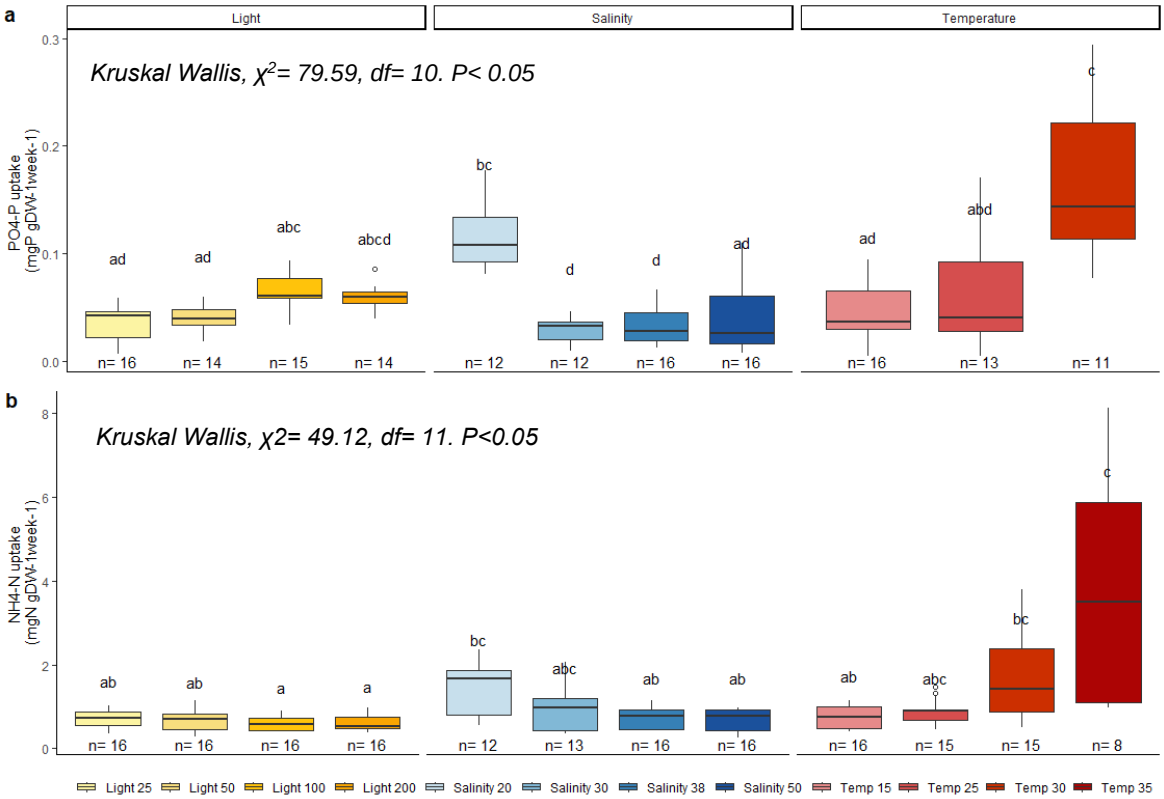

*Figure 15 – Nutrient's weekly uptake normalized to DW, **a**- Phosphate (PO4-P) uptake (p<0.05). **b**- Ammonium (NH4-N) uptake (p<0.05). Different letters were attributed to significantly different treatments based on Dunn's test, n = number of samples.*

To allow correct comparisons of uptakes, data normalized to treatments biomass. Phosphate and Ammonium (Figure 15 **a**, **b**) did not show significant differences between the uptakes, except at treatments that collapsed prior to the eighth week (20‰, 30°C, 35°C treatments) (PO_4-P, Kruskal Wallis, χ^2= 79.59, df= 10. P< 0.05) (NH_4-N, Kruskal Wallis, χ^2= 49.12, df= 11. P<0.05).

Photosynthetic activity

With all measured photosynthetic factors (ETR_{max}, E_k, F_v/F_m, Alpha), some significant differences were found among treatments observed, but without consistency during the time. Significant differences were observed for the treatments that collapse before the end of the eighth week (20 ‰, 35 °c, 30 °c) or because of a faulty measure (100 µE at the sixth week, and 50µE at the eighth week) (Supplementary Table 8).

Total proteins

The algae at the high light treatments (100 µE, 200 µE) and the 20 ‰ salinity treatment showed a significantly higher protein content (9.8, 11.9% and 8.6% respectively) while in the rest of the treatment's protein content does not appears to be significantly different (5.1-6.8%) (Table 6) (Kruskal Wallis, χ^2= 13.66, df= 3. p= 0.0034) (Figure 16 **a**), (Kruskal Wallis, χ^2= 12.97, df= 3. p= 0.0047) (Figure 16 **b**), (Kruskal Wallis, χ^2= 1.64, df= 3. p= 0.65) (Figure 16 **c**). T_0 samples results were excluded from analysis because of an error during extraction.

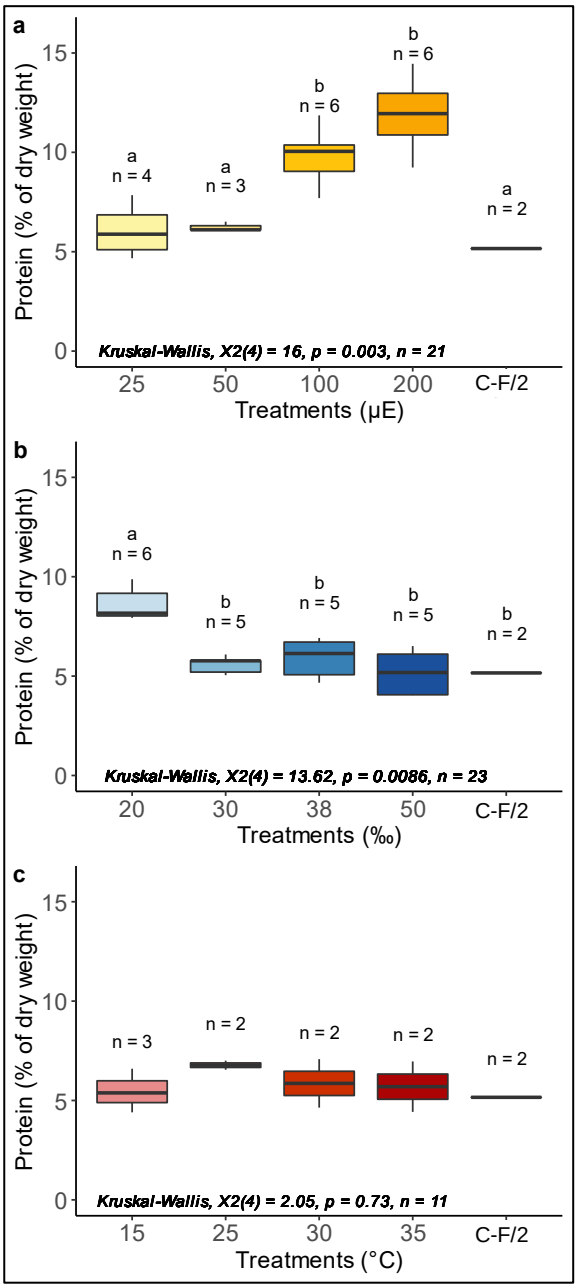

*Figure 16 - Protein as % content of dry biomass weight, **a**- Light treatments (p=0.003), **b**-Salinity treatments (p=0.008), **c**- Temperature treatments (p=0.73). Different letters were attributed to significantly different treatments based on Dunn's test, n = number of samples.*

Table 6 - Average protein content (%) of all treatments

Treat. Type	Treatment	Protein (%) Mean ± SD	n
Light	25 µE	6.07±1.41%	4
	50 µE	6.24±0.24%	3
	100 µE	9.81±1.43%	6
	200 µE	11.90±1.85%	6
Salinity	20 ‰	8.60±0.85%	6
	30 ‰	5.58±0.43%	5
	38 ‰	5.90±1.00%	5
	50 ‰	5.18±1.14%	5
Temperature	15° C	5.46±1.10%	3
	25° C	6.78±0.32%	2
	30° C	5.86±1.72%	2
	35° C	5.70±1.79%	2
Control	C-F/2	5.16±0.002%	2

Pigment content

All chromatograms (Figure 17) aligned retention time (RT) manually, comparing the UV/Vis absorption spectra of each peak to allow comparison, and the RT axis was

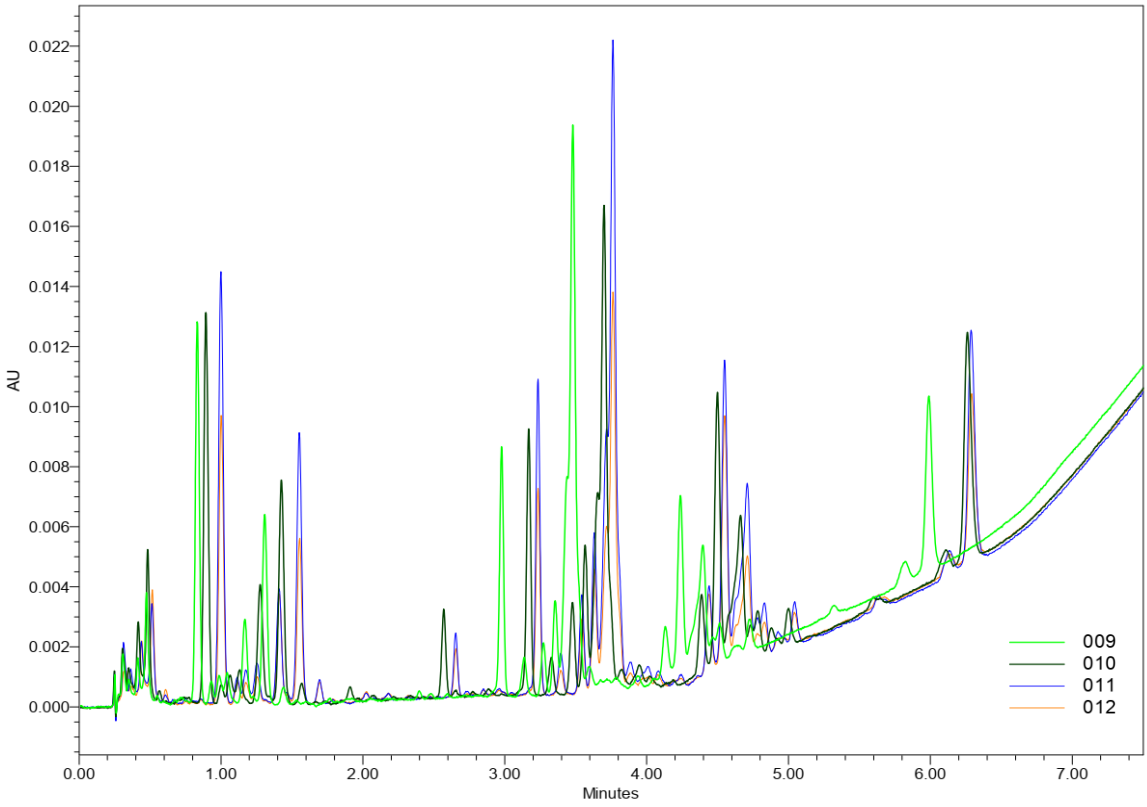

Figure 17 - A typical unaligned chromatograms. 100µE treatment chromatograms needed a manual alignment to allow comparison.

then replaced with a generalized peak numbers axis. Since no standards run during this work, we could not define the peak's substance and concentration. For this reason, the peak area (AUC) was divided by the dry weight of the samples to normalize the results. Heatmap processed (Figure 18) to visualize the substances' changes following the different treatments. 2-Way ANOVA was then used to define the significant difference between the treatments at each peak. Out of 47 Aligned peaks, only 19 were statistically affected by treatment levels (Table 7). The temperature had the most substantial effect on the algae, followed by salinity and light intensity levels. Light intensity levels positively affected peaks 5, 36, 37, 44, 47, and negatively on peak 29 (2Way ANOVA, Df-107, F-28.677, $p<0.05$). The 25µE treatment showed a great variability that masked the trends of peaks 5, 29, 44. The ascending salinities showed a significant positive effect on peaks 6, 9, 16, 227, 29, 36, 39, 42, 44, and 47, and negatively on peaks 5 and 15 (2Way ANOVA, Df-111, F-18.536, $p<0.05$). The increasing temperature gave a significant negative effect at peaks 1, 5, 9, 13, 16, 24, 25, 29, 36, 37, 39, 42, 44, 46, 47 and a positive effect at peaks 3 and 15 (2Way ANOVA, Df-110, F-3.192, $p<0.05$). The 35°C treatment showed a great variability that masked the trends at peaks 15 and 37, while it was the only effector at peaks 1, 5, 25, 36, and 46.

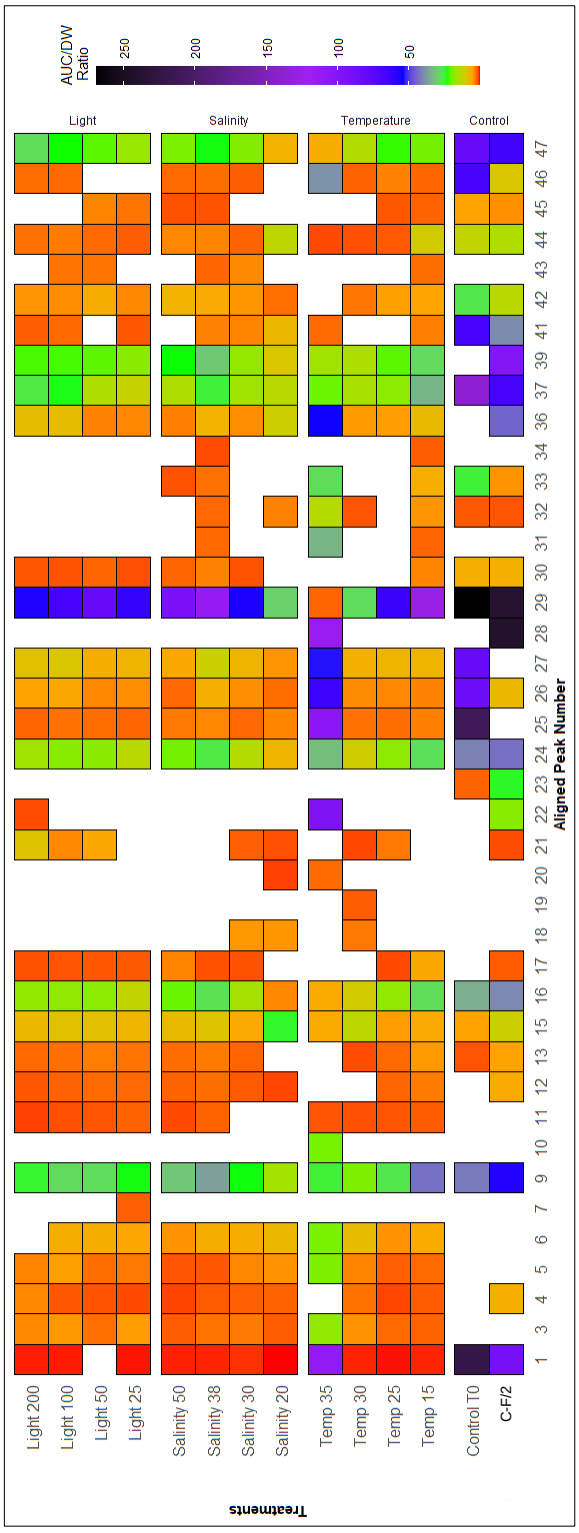

Figure 18 - Heatmap of the aligned peaks, the normalized area under the curve (AUC) to DW (mg) of the pigments UPLC chromatograms outputs

*Table 7 - UPLC chromatograms aligned peaks affected by the different treatments. '+' indicates an increasing effect compared to increasing levels of treatment. '-' indicates an increasing effect compared to the decreasing levels of treatment. *- trend masked by the most variable effector. **- only the most variable treatment with significant different*

	1	3	5	6	9	13	15	16	24	25	27	29	36	37	39	42	44	46	47	Stats
Light			+*									-*	+	+			+*		+	2Way ANOVA, Df-107, F-28.677, p<0.05
Salinity			-	+**	+		-	+	+		+	+	+		+	+	+		+	2Way ANOVA, Df-111, F-18.536, p<0.05
Temperature	-**	+	-**		-	-	+*	-	-	-**		-	-**	-*	-	-	-	-**	-	2Way ANOVA, Df-110, F-3.192, p<0.05

Discussion

Seaweed mariculture is rising worldwide, constantly[3,9,10,34]. Following the demand for food substances such as proteins and other bio-materials such as pigments and polysacharides[1,17,27], there is an increased interest in mariculture of seaweed such as the *Codium* sp. [34] [20,35].

Codium spp. are widely distributed worldwide in cold and tempered seas, including the Mediterranean Levant Basin[22], where the most common is *Codium taylorii*. Genetic identification of the dominant species on the coast of Israel revealed that the local *Codium* population includes the dominant species *Codium taylorii* and a second invading species, *Codium parvulum*, which is known to be present over a decade[36].

Codium taylorii showed a remarkable adaptation to a variety of salinities starting from 38‰ and up to 50‰, but it is best performing at 30‰-38‰, allowing continuity after stress events[37,38]. Our results support these observations, wherein in all of our treatments that survived, the SGRs' from weeks 0-4 to weeks 5-8 significantly increased, showing the adaptability of this species[38]. This understanding can manipulate the growth and avoid epiphytes' succession.

The most outstanding effect was that the different temperatures had on the algae growth. Where the two highest temperature treatments did not survive the whole period of the experiment, it seems that *Codium* prefers cold water, with a limit below 30°C, preferably 20°C, as described by Hanisak[37], that reached more than 4% daily growth at temperatures between 18-24°C. This finding raises ecological concerns about the success of the *Codium* population in the Levant basin, as this area responds rapidly to climate change[39,40].

Daily growth rates reached almost 2%, at 100µE light intensity and above levels. This finding is in agreement with Hanisak[37] for a similar setup. Furthermore, the slight relative difference compared to 50 µE SGR results shows that these algae can grow well under low light intensities[41], suggesting higher intensities can be avoided. Interestingly, observing *Codium* in its natural habitat reveals higher densities in low light environments such as under overhangs. Nevertheless, some obseravtions[42,43] on *Codium tomentosum* at 120µE and *Codium fragile* at 56µE and

560 µM of NO_3 showed more than double our highest SGR, suggesting that the fullest potential is yet to be reached.

The higher protein content was almost 12% of DW at the high light intensity and 6% on average for all other treatments. Similar to former observations[43,44], showed variation between 5-19% of protein content in *C. fragile* and *C. tomentosum*.

Environmental factors usually trigger morphological changes as a defense mechanism against an environment that has become hostile. Indeed, we observed a significant effect on diverse morphology factors in this study. One such factor is rigidity, showing some thresholds crossed above 50µE light intensity and below 38‰ salinity levels. No effect on rigidity was observed at the C-F/2 treatment. However, the major factor that affected most the algae rigidity was temperature, where the algae lost entirely its typical shape and culture collapsed, above 30°C.

Filaments formation is assumed to be related to rigidity plasticity[45,46], as a vegetative reproduction mechanism. Our observation showed high filamenting behavior on almost all treatments, except 15°C and C-F/2, not related to rigidity difference, suggesting that filamenting behavior is highly related to nutrients availability, contrary to former observation[43]; where algae in high nutrients cultures grew without any morphological changes. The fact that algae can be manipulated to produce filaments without losing their rigidity, hence, stay vital, can be used as a vegetative reproduction[46–48] multiplier in mariculture.

Interestingly, fragmentation was related to the rising temperatures by Bégin and Scheibling[49], wherein this study fragmentation was observed as typical behavior of *Codium* and was not affected by all the measured parameters in contrary to their findings. This finding supports a vegetative reproduction mechanism as shown in the past[48–50].

Photosynthetic activity was expected to change among the treatments[51]. However, we did not observe any significant differences between treatments, indicating a possible methodology error or faster adaptation than expected.

We next compared the effect of the different treatments on the pigment's composition. We expected that pigment composition would increase as the light, salinity, or temperature levels increases[52,53]. However, we observed the opposite

trend. In addition, high variability was found in the 25µE, 50‰, and 35°C treatments, indicating that the algae at these treatments might be stressed rather than adopted. The pigment composition significantly differs between the treatments (Table 7), suggesting that we can manipulate pigment composition to achieve desired components. Nevertheless, more comprehensive research should be done to define the nature of the separated substances and their concentrations on the different treatments.

Combining the SGR results with the protein content results, we found a significant advantage to grow the algae with 100µE intensity, showing lower epiphytes impact regarding higher light intensities while keeping higher protein content. However, nutrient amounts should be investigated, as they significantly affect rigidity. Growing *Codium* on land-based facilities should be combined with commercial fish farms and industrial effluents, considering the alga plasticity at the local range of salinities and seasonal temperatures.

This novel local research gives a baseline for further feasibility studies for growing *Codium taylorii* in the local algaculture industry. The following steps should examine the effect of nutrient levels on the examined factors and upscale the water volumes and yields.

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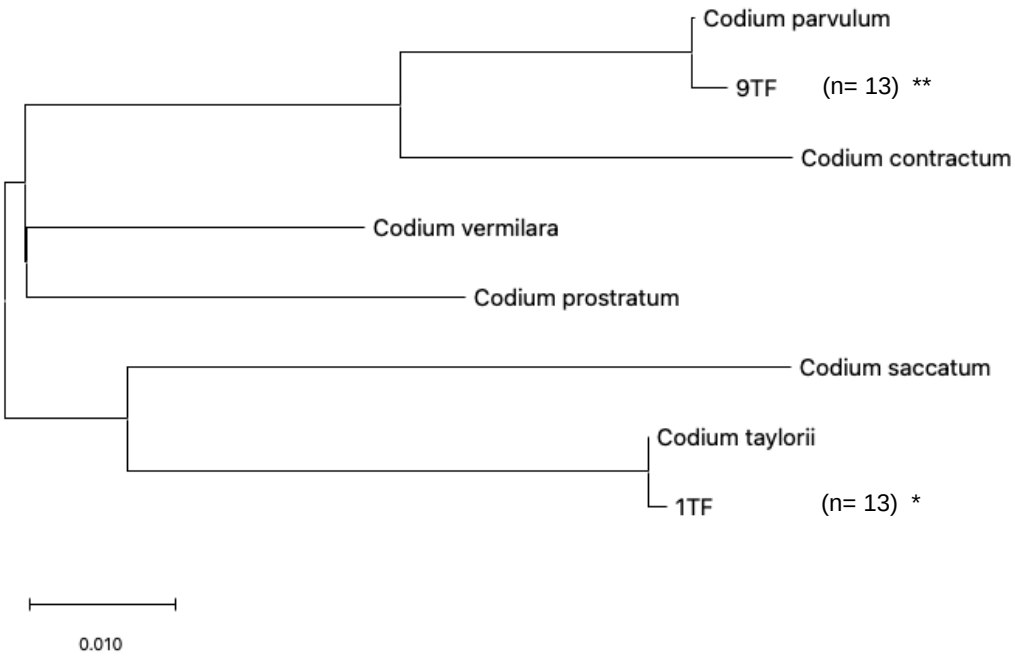
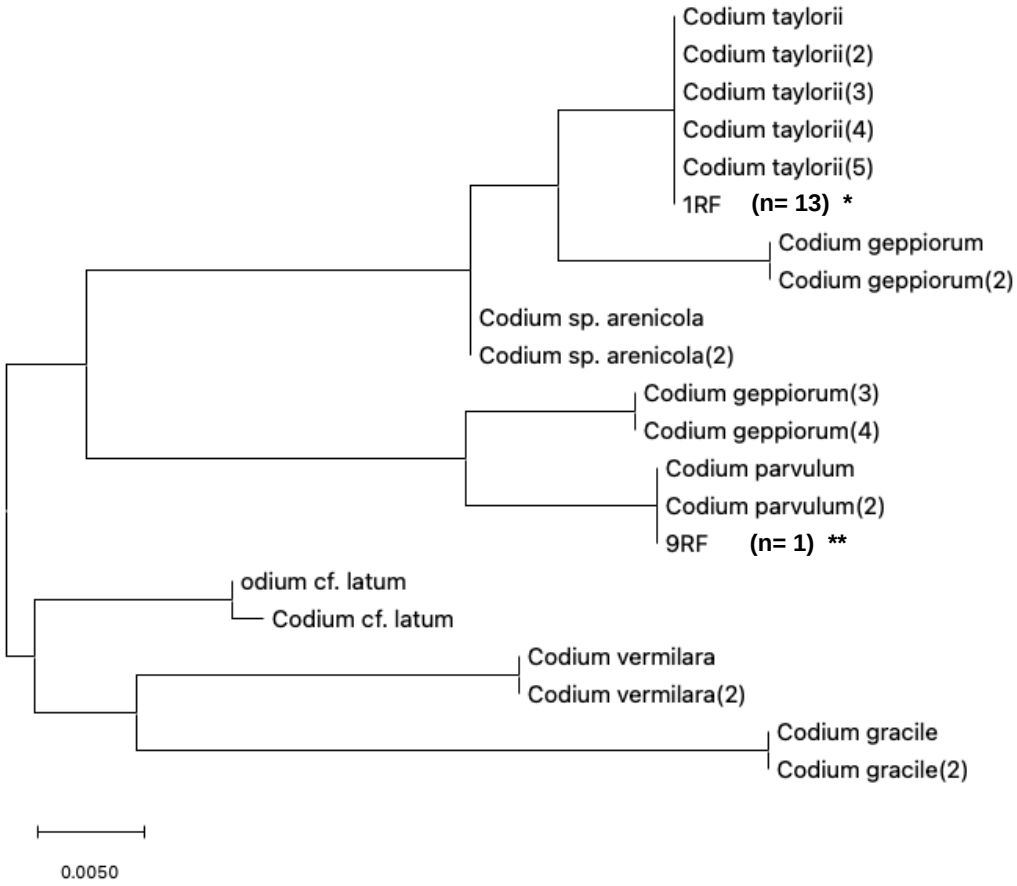
Supplementary material

Evolutionary analysis



*Supplementary Figure 19 - Evolutionary analysis by Maximum Likelihood method for DNA TufA sequence of 14 samples compared to 6 sequences from NCBI database. Evolutionary analyses were conducted in MEGA X[54,55]. * - indicates Codium Taylorii (number of samples), **- indicates Codium parvulum (number of samples).*



Supplementary Figure 20 - Evolutionary analysis by Maximum Likelihood method for DNA RbcL sequence of 14 samples compared to 19 sequences from NCBI database. Evolutionary analyses were conducted in MEGA X[54,55]*. * - indicates Codium Taylorii (number of samples), **- indicates Codium parvulum (number of samples).*

PAM results

Supplementary Table 8 - Photosynthetic activity results obtained from the Imaging-PAM for all treatments during four time points. values expressed as mean ± SD (n)

Measure	Treat. Type	Treatment	T_0	T_{0+2}	T_{0+6}	T_{0+8}
ETR_{max}	Light	25 µE	10.54±2.44 (12)	22.06±1.22 (10)	23.71±3.69 (12)	13.41±1.92 (12)
		50 µE	10.54±2.44 (12)	21.25±2.55 (12)	24.67±2.66 (12)	15.42±1.16 (10)
		100 µE	10.54±2.44 (12)	22.95±2.14 (12)	22.77±3.37 (12)	15.92±1.67 (11)
		200 µE	10.54±2.44 (12)	21.06±2.52 (12)	25.56±3.16 (12)	18.66±1.04 (10)
	Salinity	20 ‰	10.54±2.44 (12)	7.17±2.89 (10)		
		30 ‰	10.54±2.44 (12)	20.92±2.51 (12)	24.03±1.91 (12)	18.04±2.64 (12)
		38 ‰	10.54±2.44 (12)	24.33±2.81 (11)	27.99±2.11 (12)	14.66±1.57 (11)
		50 ‰	10.54±2.44 (12)	20.09±2.97 (12)	24.22±1.2 (11)	18.32±3.5 (12)
	Temperature	15° C	10.54±2.44 (12)	13.44±5.97 (12)	27.38±4.19 (12)	23.39±6.11 (12)
		25° C	10.54±2.44 (12)	26.51±1.28 (10)	22.53±1.72 (12)	17.58±2.02 (11)
		30° C	10.54±2.44 (12)	17.59±3.62 (12)		
		35° C	10.54±2.44 (12)			
E_k	Light	25 µE	44.46±8.98 (12)	78.12±8.14 (12)	89.21±5.37 (9)	51.38±6.26 (12)
		50 µE	44.46±8.98 (12)	79.51±8.68 (12)	91.29±8.68 (12)	60.44±7.35 (12)
		100 µE	44.46±8.98 (12)	83.66±7.94 (12)	95.63±11.33 (12)	61±5.57 (12)
		200 µE	44.46±8.98 (12)	79.96±7.93 (12)	86.94±10.45 (12)	69.99±5.2 (12)
	Salinity	20 ‰	44.46±8.98 (12)	35.48±10.9 (10)		
		30 ‰	44.46±8.98 (12)	77.6±6.98 (12)	88.28±6.23 (12)	68.99±7.92 (12)
		38 ‰	44.46±8.98 (12)	28.69±3.38 (11)	103.12±7.33 (12)	18.56±2.11 (12)
		50 ‰	44.46±8.98 (12)	76.05±11.17 (12)	76.66±4.79 (11)	71.13±12.26 (12)
	Temperature	15° C	44.46±8.98 (12)	60.66±14.28 (11)	102.06±14.03 (12)	59.79±15.76 (12)
		25° C	44.46±8.98 (12)	90.17±10.37 (12)	26.16±2.79 (12)	23.17±2.3 (10)
		30° C	44.46±8.98 (12)	61.74±10.26 (12)		
		35° C	44.46±8.98 (12)			

Measure	Treat. Type	Treatment	T_0	T_{0+2}	T_{0+6}	T_{0+8}
F_v/F_m	Light	25 µE	0.7±0.04 (12)	0.76±0.01 (12)	0.77±0.02 (12)	0.76±0.01 (12)
		50 µE	0.7±0.04 (12)	0.77±0.01 (12)	0.76±0.01 (12)	0.83±0 (10)
		100 µE	0.7±0.04 (12)	0.75±0.01 (12)	0.7±0.01 (11)	0.76±0.01 (11)
		200 µE	0.7±0.04 (12)	0.73±0.02 (12)	0.76±0 (9)	0.76±0.01 (12)
	Salinity	20 ‰	0.7±0.04 (12)	0.61±0.03 (10)		
		30 ‰	0.7±0.04 (12)	0.76±0.01 (12)	0.76±0 (10)	0.77±0.01 (12)
		38 ‰	0.7±0.04 (12)	0.78±0.01 (12)	0.76±0.01 (12)	0.78±0.01 (12)
		50 ‰	0.7±0.04 (12)	0.77±0.01 (12)	0.73±0.01 (12)	0.72±0.01 (12)
	Temperature	15° C	0.7±0.04 (12)	0.68±0.06 (12)	0.74±0.01 (12)	0.74±0.02 (12)
		25° C	0.7±0.04 (12)	0.77±0 (10)	0.75±0.01 (12)	0.77±0.01 (10)
		30° C	0.7±0.04 (12)	0.77±0 (9)		
		35° C	0.7±0.04 (12)			
alpha	Light	25 µE	0.24±0.01 (12)	0.27±0 (11)	0.27±0.01 (11)	0.26±0.01 (11)
		50 µE	0.24±0.01 (12)	0.27±0.01 (11)	0.27±0 (12)	0.27±0.01 (12)
		100 µE	0.24±0.01 (12)	0.27±0 (11)	0.24±0 (11)	0.26±0.01 (11)
		200 µE	0.24±0.01 (12)	0.26±0.01 (12)	0.29±0.03 (12)	0.26±0.01 (12)
	Salinity	20 ‰	0.24±0.01 (12)	0.2±0.03 (11)		
		30 ‰	0.24±0.01 (12)	0.26±0.01 (10)	0.27±0.01 (12)	0.26±0.01 (11)
		38 ‰	0.24±0.01 (12)	0.85±0.03 (12)	0.27±0 (11)	0.77±0.05 (12)
		50 ‰	0.24±0.01 (12)	0.26±0 (11)	0.32±0.01 (12)	0.26±0.01 (12)
	Temperature	15° C	0.24±0.01 (12)	0.24±0.03 (11)	0.27±0.01 (12)	0.39±0.01 (12)
		25° C	0.24±0.01 (12)	0.29±0.02 (12)	0.86±0.04 (12)	0.78±0.07 (12)
		30° C	0.24±0.01 (12)	0.28±0.02 (12)		
		35° C	0.24±0.01 (12)			

השפעת תנאים א-ביוטים והעשרה בחנקן על ההרכב הביוכימי וקצבי הגידול של האצה קודיום באגן המזרחי של הים התיכון

מגיש: איתי קולסקי

תקציר

בעשורים האחרונים, קיימת מגמה למציאת מקורות חלבון אלטרנטיביים לאלו הזמינים כיום. הצורך במקור חדש נובע מהגידול באוכלוסיית העולם והקטנת השטחים החקלאיים הקיימים. פתרונות כמו חקלאות יבשתית מתועשת וחקלאות ימית קיימים. אצות אשר משמשות כפילטר ביולוגי מוצאות כפתרון לייצרת מקור מזון בר קיימא עם פגיעה מנימאלית בטבע. אזור האגן המזרחי של הים התיכון מהווה איזור מחייה קשה ל"מפעלי חלבון מהירי גידול" כמו אצות בגלל תנאי הים הקשים והעוני בחומרי הזנה (נוטריינטים). יתרה מזאת, אצות אשר מצליחות לפרוח בתנאים קשים אלו, מציגות פוטנציאל גדול, לחקלאות אצות יבשתית, בהתחשב בשמירה על ערכי הטבע ובערכים ביוכימיים שאצות אלו יכולות להכיל. בנוסף, אצות יכולות לגדול עת תוצרים שניוניים של מערכות חקלאות מים ולהוות נדבח במערכות אינגרטיביות לגידול דגים וחסרי חוליות (IMTA). גידול שכזה, ימחזר תוצרים רעילים כמו אמוניה, אשר נלווים למערכות חקלאות ימית רגילה. קודיום, אשר גדלה באופן טבעי באגן המזרחי של הים התיכון, כבר קיימת כמוצר בשווקים, בעיקר במזרח, עם שווי גבוה מהאצה פורפירה. אימוץ הגישה של "בתי זיקוק ביולוגיים", אצה מקומית זאת יכולה להניב מוצרים בעלי ערך גבוה דוגמאת חלבונים, גמנטים וסוכרים מורכבים. במחקר זה נבחנה האצה קודים טילוריי והראתה פוטנציאל גידול של כמעט 2% ביום, עם כ-12% תכולת חלבון. בנוסף ניתן לראות עמידות גבוהה לשינויים סביבתיים. מחקר זה יכול להוות בסיס להמשך פיתוח והצגת מוצר חדש לתעשיית החקלאות הימית באיזורינו.

השפעת תנאים א-ביוטים והעשרה בחנקן על ההרכב הביוכימי וקצבי הגידול של האצה קודיום באגן המזרחי של הים התיכון

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אוניברסיטת חיפה

הפקולטה למדעי הים

בית הספר למדעי הים ע"ש ליאון צ'רני

החוג לביולוגיה ימית

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