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Response of Marine Primary Producers to Olivine Additions

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Abstract. Carbon dioxide removal (CDR) technologies are gaining increasing attention as a potential solution to reduce atmospheric CO_2 concentrations and combat climate change. Ocean alkalinity enhancement (OAE) seeks to enhance the ocean's CO_2 absorption capacity by introducing powdered minerals or dissolved alkaline substances into the surface ocean. Nevertheless, the impact of OAE on marine ecosystems remains largely uncharted. In this study, we explored the impact of olivine on a diversity of cosmopolitan primary producers, including Coccolithophores, Diatoms, Dinophyceae, *Micromonas sp.*, *Prochlorococcus* sp., and *Synechococcus* sp. Here, we show that most primary producers were not impacted negatively by the concentrations of olivine additions that were applied despite olivine additions increasing nickel concentrations in our cultures. Additions of olivine did not lead to growth inhibition but resulted in a slight increase in growth in most cultures, with picoplankton benefiting the most. However, phytoplankton responses were species-specific and subject to the media used in a combination with the olivine addition. Additionally, it is essential to mention the pitfalls and concerns associated with our experimental setup, particularly regarding the impact of the medias and considerations of carbonate chemistry. Our findings raise confidence in applying olivine for carbonation to generate CO_2 removal without harming primary producers; however, future studies should include tests on an ecosystem level to investigate potential effects on different trophic levels and natural settings.

1 Introduction

Ocean alkalinity enhancement (OAE), a carbon dioxide removal (CDR) technology, is gaining substantial attention as a strategy to reduce atmospheric CO_2 concentrations and thus mitigate climate change (Fawzy et al., 2020; IPCC, 2023; Pörtner et al., 2023). OAE aims to increase the alkalinity of surface ocean waters and sediments of entire regions, enhancing the ocean's capacity

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to absorb more CO_2 from the atmosphere (Campbell et al., 2022; Bach et al., 2019), while at the same time mitigating ocean acidification (Kheshgi, 1995; Doney et al., 2009; Egleston et al., 2010; Heinze et al., 2015; Renforth and Henderson, 2017; Bach et al., 2019).

Mineral-based OAE utilizes the addition of ground alkaline minerals to ocean waters to increase seawater alkalinity (Renforth and Henderson, 2017). OAE approaches are based on the natural process of rock weathering as a key process of stabilizing Earth's temperature (Urey, 1957; Walker et al., 1981; Berner et al., 1983; Volk, 1987) and are closely linked to another CDR technology known as enhanced weathering (EW) (Köhler et al., 2010; Goll et al., 2021; Calabrese et al., 2022). EW processes enhance weathering rates by, for instance, spreading ground silicate rocks or basaltic rocks on terrestrial land, leveraging natural weathering mechanisms (Renforth et al., 2015; Meysman and Montserrat, 2017; Beerling et al., 2020). The permeance of CO_2 removal achieved through OAE is estimated to last more than ten thousand years, depending on the duration of HCO_3^- in solution and the absence of reverse reactions due to a changed equilibrium (Archer, 2005; Rau, 2011; Middelburg et al., 2020) Notably, reverse reactions of the carbonate-silicate weathering are reduced in ocean regions with low salinity, faster mixing, and dilution (Archer, 2005; Kirchner et al., 2020; Bertagni and Porporato, 2022).

Rising concentrations of CO_2 in the atmosphere have profound impacts on the marine environment (IPCC, 2023; Smith et al., 2023), leading to, for example, heat waves and ocean acidification (Doney et al., 2009; Oliver et al., 2018). These changes cause significant alterations to marine biodiversity, including changes in primary producer community composition and activity (Doney et al., 2012). A change in the primary producer community in response to alkaline substances, such as olivine and quicklime, has been suggested (Bach et al., 2019), with certain taxonomic groups being favored over others. The addition of olivine was, for instance, suggested to promote a diatom- and cyanobacteria-rich environment, producing a "Greener" ocean, as these groups have high requirements for one or more elements present in olivine, including silicon (Si), iron (Fe), and nickel (Ni). On the other hand, the addition of quicklime was suggested to lead to a coccolithophore-rich environment, generating a "Whiter" ocean (Bach et al., 2019).

Primary producers comprise the first trophic level of the marine food web and play a crucial role as the foundation of global ecosystems (Michaels and Silver, 1988; Longhurst et al., 1995; Buesseler, 1998). Marine phytoplankton is responsible for approximately half of Earth's carbon

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fixation (Longhurst et al., 1995; Field et al., 1998) and a driver of the biological carbon pump exporting CO_2 to waters below the mixed layer, thus removing CO_2 on longer timescales (Volk and Hoffert, 1985). The transfer of organic matter by the biological carbon pump creates a vertical dissolved inorganic carbon (DIC) gradient, enhancing the ocean's ability to absorb CO_2 from the atmosphere through air-sea exchange to replace the DIC that was converted into organic matter (Kwon et al., 2009). Approximately 20% of carbon fixed during primary production in the euphotic zone is transported into the deep ocean through various mechanisms (Dunne et al., 2007; Devries and Weber, 2017; Nowicki et al., 2022).

The primary producers we have chosen to work with are cosmopolitan and quantitatively important in the marine environment (Waterbury et al., 1979; Chisholm et al., 1988; Quere et al., 2005; Worden et al., 2015). Prochlorococcus and Synechococcus are cyanobacteria, a diverse phylum of photoautotrophic prokaryotes (Chisholm et al., 1988; Chisholm et al., 1992). The eukaryotic primary producers in this study included coccolithophores (*Emiliania huxleyi)*, diatoms (*Thalassiosira weissflogii*, *Skeltonema marinoi)*, dinophyceae (*Scrippsiella trochoidea)* and a green alga (*Micromonas commoda).* Coccolithophores are calcifying organisms with exoskeletons made of calcium carbonate called coccoliths, making them particularly vulnerable to ocean acidification (Iglesias-Rodriguez et al., 2008; Meyer and Riebesell, 2015). They are ecologically and biogeochemically essential and play a vital role in the marine biological carbon pump (Poulton et al., 2013; De Vries et al., 2021). Diatoms are responsible for about one-fifth of Earth's photosynthesis (Treguer et al., 1995). They are present in all marine environments and are responsible for generating as much organic carbon as all the global rainforests combined, which illustrates their importance for the global carbon cycle (Treguer et al., 1995; Field et al., 1998). In the ocean, diatoms account for approximately 40 % of the total primary production (Treguer et al., 1995; Tréguer and Pondaven, 2000; Sarthou et al., 2005). Diatoms require silicate to construct their siliceous cell walls. Therefore, elevated silicate concentrations in the ocean could promote the proliferation of diatoms in comparison to non-siliceous types of phytoplankton, an aspect potentially relevant in the context of OAE-mineral additions (Tréguer and Pondaven, 2000). Dinophyceae, coccolithophores, and diatoms initiated a major shift in the global carbon cycle, which began an era of increasing atmospheric O_2 concentrations and declining atmospheric CO_2 concentrations (Katz et al., 2005). Dinophyceaes are motile phytoplankton, allowing them to migrate in the water column (Margalef, 1978). It is a diverse group of organisms in which some

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representatives can produce toxins, bioluminesce, or even be parasitic or symbiotic (Hackett et al., 2004). Lastly, *Micromonas sp*. is a unicellular picoplanktonic (≤ 2 µm cell diameter) marine green alga which thrives in most ocean environments, from coastal to open waters (Not et al., 2004; Worden et al., 2009; Cuvelier et al., 2017). Several studies over the past decades have observed that there has been an increased abundance of picophytoplankton species, including *Micromonas* spp. In comparison, larger phytoplankton have decreased in abundance. These changes are strongly linked to climate change, which could lead to reduced biological production at higher trophic levels and points towards the importance of *Micromonas* spp. in a future ocean (Li et al., 2009; Mckie-Krisberg and Sanders, 2014; Van Baren et al., 2016).

In this study, we explored the impact of olivine additions (grain sizes <63 µm), on ten strains of primary producers, including coccolithophores, diatoms, dinophyceae, the green alga *Micromonas commoda*, as well as on cyanobacteria (Prochlorococcus, and Synechococcus). We monitored the response of primary producers using their overall growth and snapshots of carbon fixation rates in response to olivine additions. Those biological parameters were complemented with quantification of trace metal and ion concentrations, including Si, Ni, magnesium (Mg), manganese (Mn), calcium (Ca), and Fe, to explore potential ecotoxicological or growth-stimulating effects on primary producer cultures. In addition, we assessed how olivine additions affected the carbonate system during the experimental periods. Taken together, we hope that our study can inform and contribute to the design of experiments in relevant ecosystems, model explorations of OAE applicability, and decision-making on the applicability of olivine additions for OAE as a tool to mitigate climate change.

2 Experimental methods

2.1 Mineral analysis

Olivine used for our addition experiments were obtained from Norway. To increase the surface area of the raw minerals, olivine minerals was initially crushed in a Laboratory Jaw Crusher (Type BB51, Retsch, Haan, Germany) and then further crushed using a Planetary Ball Mill PM 100 (Retsch, Haan, Germany) for 5-10 minutes at 500 Relative Centrifugal Force (RCF). The crushed materials were sieved using a 63 µm pore size sieve to select particles with diameters at or below

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<63 µm (Horowitz, 1985; Kersten and Smedes, 2002). The olivine was then ground with ball mills, requiring less energy than other ultra-fine grinding methods (Summers et al., 2005).

The specific surface area of the particles was determined using a Brunauer–Emmett–Teller (BET) approach. Samples were analyzed in duplicates (Tab. 1); a Micromeritics Gemini 2390 (Micromeritics, Norcross, USA) was used to determine further the particle sizes (Particle Analytical ApS, Hørsholm, Denmark). The results for olivine showed the following average surface areas (BET analysis) 1.81 m^2 g^{-1}(Tab. 1).

Major and trace elements of the applied olivine were analyzed with inductively coupled plasma optical emission spectroscopy (ICP-OES, Agilent 7700 ICP-MS, Agilent Technologies, Santa Clara, USA). Olivine (forsterite) is an ultramafic rock primarily composed of Mg_2SiO_4 (Bragg and Brown, 1926). The major element composition analysis displayed in Tab. 1 confirms the composition of olivine. Olivine is primarily composed of Mg (29.65 %) and Si (18.97 %).

Table 1. Specific surface area (BET, m^2 g^{-1}) and major element composition of olivine (%).

		Major elements in %						
Minerals	**BET, m^2 g^{-1}**	**Na**	**Mg**	**Al**	**Si**	**K**	**Ca**	**Fe**
Olivine	1.81	0.006	29.654	0.079	18.966	0.005	0.085	5.191

Olivine's trace metal content, a concentration of Mn (728.286 mg kg^{-1}), and a concentration of zinc (Zn) (29.1 mg kg^{-1}) was detected in olivine. Both trace metals are crucial for primary producers' growth (Blaby-Haas and Merchant, 2017) (Tab. 2). Ni, arsenic (As), and chromium (Cr) can negatively affect the growth of primary producers (Cervantes et al., 2001; Tripathi and Poluri, 2021; Guo et al., 2022). Olivine contained a concentration of As (0.066 mg kg^{-1}), and of Cr (453.3 mg kg^{-1}), and Ni (2709.2 mg kg^{-1}), all of which could impair ecosystem health. It is essential to consider that Mn, Zn, As, Cr, and Ni toxicity can vary depending on the specific aquatic setting. Guidelines on acceptable trace metal concentrations might, therefore, vary.

Table 2. Trace element concentrations of olivine in mg kg^{-1} applied in the experiments.

	Trace elements in mg kg^{-1}							
Minerals	**Cr**	**Mn**	**Co**	**Ni**	**Cu**	**Zn**	**As**	**Pb**
Olivine	453.294	728.286	106.306	2709.208	3.308	29.133	0.066	0.646

2.2 Strains and cultivation conditions

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The primary producers used in this study included cyanobacteria, dinophyceae, chlorophyta, diatoms, and coccolithophores. We explored how they responded to abrupt exposure to olivine additions, thus imitating a water column mineral addition. The following strains were all related to the open oceans, specifically the Atlantic and Pacific oceans. The primary producers were pure cultures purchased from Roscoff Culture Collection (CNRS-Sorbonne Université, Station Biologique, Place G. Tessier 29680 Roscoff, France) or Bigelow, National Center for Marine Algae and Microbiota (Bigelow, East Boothbay, ME, USA).

Table 3. Overview of the phytoplankton strains, cultivation media, geographic origin, experimental period [d], and if the phytoplankton cultures were grown on a shaker during the experimental period.

Species	**Strain Name**	**Cultivation Media**	**Ocean Region**	**Runtime of experiment [d]**	**Shaking**
Synechococcus sp.	A15-62 Clonal	PCR S11 Red Sea Medium (Roscoff Culture Collection, 2019) adopted from (Rippka et al., 2000)	North Atlantic, Tropical Atlantic	25	Shaking
Synechococcus sp.	EUM Syn10	PCR S11 Red Sea Medium (Roscoff Culture Collection, 2019) adopted from (Rippka et al., 2000)	North Atlantic, Tropical Atlantic	10	No Shaking
Synechococcus sp.	WH8102	PCR S11 Red Sea Medium (Roscoff Culture Collection, 2019) adopted from (Rippka et al., 2000)	North Atlantic, Caribbean Sea	9	Shaking
Prochlorococcus sp.	NATL2-M98	PCR S11 Red Sea Medium (Roscoff Culture Collection, 2019) adopted from (Rippka et al., 2000)	North Atlantic	34 (41) *	Shaking
Prochlorococcus sp.	EQPAC1-C	PCR S11 Red Sea Medium (Roscoff Culture Collection, 2019) adopted from (Rippka et al., 2000)	Pacific Ocean, Equatorial Pacific	9	Shaking
Scrippsiella trochoidea	CCMP1331	L1 medium (Biglow, 2020, DOC-051.000) adapted from (Guillard and Hargraves, 1993), silicate excluded	Unknown	44	No Shaking
Micromonas commoda	CCMP489	K medium (Biglow, 2020, DOC-050.000) adapted from (Keller, 1985; Keller et al., 1987)	North Atlantic, Sargasso Sea	9	No Shaking
*Thalassiosira weissflogii***	CCMP1051	L1 medium (Biglow, 2020, DOC-051.000) adapted from (Guillard and Hargraves, 1993), silicate included	North Pacific	42	Shaking
*Skeletonema marinoi***	CCMP2092	L1 medium (Biglow, 2020, DOC-051.000) adapted from (Guillard and Hargraves, 1993), silicate included	North Atlantic, Adriatic Sea	42	Shaking
*Emiliania huxleyi****	AC448	Adapted K medium, (Biglow, 2020, DOC-050.000) adapted from (Keller, 1985; Keller et al., 1987)*****	North Atlantic	32	Shaking

*Dolomite addition of NATL2-M98 was incubated for an extended 41 days.

**Diatom

***Coccolithophore

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**** Adaptions of K medium involving division of the additions of components by two.

The seawater for L1, K, and their adaptations have been taken from the Danish coastal North Sea (55°17'49.4" N 8°39'05.2" E) at the beginning of high tide on June 23, 2020, and July 6, 2021. Seawater salinity was between 30.9 – 31.0. The seawater was prefiltered with a bottle top 0.2 µm PES membrane filter (Avantor VWR® Radnor, Pa, USA). The primary producers were cultivated in biological triplicates. Salinity varied between the media. PCR S11 had a salinity of 36.0, while the others were mixed with North Sea water and had a salinity between 31.4 and 31.5.

2.3 Culture experiments

To ensure consistent and comparable results in cultivating the 10 primary producers affected by the olivine additions, we kept the primary producers in identical conditions in a cultivation room with a day/night cycle of 8 h of light and 16 h of darkness. The laboratory tables were consistently exposed to an average light intensity of 42.47 lux (lumen/m^2) measured with LI-250A Light Meter (LICOR®, Lincoln, USA) in the light period and maintained a controlled room temperature of 20.0 C°- 20.3° C throughout the experiments. All primary producers were cultivated in 1 L or 2 L conical flasks, with headspaces in exchange with the atmosphere. 400 mL of the algal-specific medium was mixed with 100 mL of the desired, pre-grown strain prior to the experimental start. Samples were continually collected at the same time, between 9:30 a.m. and 11:00 a.m. The primary producers, namely CCMP489, EUM Syn10, and CCMP1331, were not subjected to shaking during the cultivation experiments. CCMP489 and EUM Syn10 were cultivated in a growth chamber Model KBW 400 (Binder, Tuttlingen, Germany), where shakers were not used but were still cultivated under conditions similar to those mentioned. The remaining primary producers were subjected to shaking at 80 relative centrifugal force (RPG) using a Laboshaker (C. Gerhardt, Königswinter, Germany).

Carbonate chemistry was investigated to assess the influence of olivine additions on primary producers' growth. Ground additions of 0.5 g L^{-1} were added for olivine in the preliminary phase of the experiment, giving a solid-liquid ratio of 0.0005 kg L^{-1}. This amount has been chosen based on our earlier experiments (Rønning, in prep, 2024), in which we could obtain and maintain

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significant increases in TA and pH over a 35-day runtime. Olivine was not detected to fully dissolve in our experiments. Control batches for each strain were cultivated under the same conditions as those with mineral additions but without mineral supplements.

Parameters that were investigated during the cultivation study of the 10 primary producers of olivine additions were relative fluorescence units (RFU) or optical density (OD 680) (S2-supplemental material) and pigment concentration. RFU and pigment analysis was conducted on a Triology® Fluorometer (Model #7200-00) using a chlorophyll-a (chl-a) in vivo blue module (Model #7200-043, both Turner Designs, San Jose, CA, USA), set to use a method for chl-a. Chl-a samples were collected alongside every sample session to monitor growth over time by filtering the samples onto GF/F filters (GE Healthcare Life Sciences, Whatman, USA). Filtered volumes for chl-a were adjusted to the density of the primary producer cultures. Filters were stored in darkened 15 mL LightSafe centrifuge tubes (Merck, Rahway, NJ, USA) at -20 °C until further analysis. A calibration was generated for pigment analysis using a standard stock solution based on chl-a from spinach (Sigma Aldrich, Burlington, USA). The samples were submerged in 8 mL of 90% acetone overnight at a temperature of 5°C; the next day, 1 mL of the overnight liquid was added into a 1.5 mL glass vial (Mikrolab Aarhus A/S, Aarhus, Denmark) and then analyzed on the Triology® Fluorometer. RFU measurements were taken at each sampling point to monitor the real-time growth of primary producers. 1 mL samples were placed into glass vials with a pipette and analyzed in the Triology® Fluorometer. However, due to the unavailability of the Triology® fluorometer, two primary producers, namely the CCMP1331 and A15-62 clonal strains, had their growth analyzed by measuring the optical density at OD 680 with a Multiscan GO spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA).

2.4 Carbon fixation in the cultures

We determined carbon fixation rates in the cultures to assess the potential impacts of olivine additions on primary production. Samples were incubated with 20 µg mL^{-1} 98 % sodium bicarbonate ^{-13}C before starting the experiments (Lot #MBB9999 and #MBBD2322; Sigma-Aldrich, Burlington, USA). An initial set of biological triplicate samples was collected at the beginning of the experiments before the additions of olivine as T0. After the total incubation time for each primary producer (Tab. 3), samples were filtered onto pre-combusted GF/F filters (GE Healthcare Life Sciences, Whatman, USA). Filters were stored at -20 °C until further analysis.

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Filters were then acidified and dried before analysis using an Elemental Analyzer Flash 2000 (Thermo Fisher Scientific, Waltham, MA, USA) coupled to an isotope ratio mass spectrometer Finnigan Delta V Advantage (Thermo Fisher Scientific, Waltham, MA, USA), as described earlier (Reeder et al., 2022).

2.5 Carbonate system parameters (DIC / pH / TA)

Parameters measured within the carbonate system, including DIC, pH, and total TA, were collected alongside every sample session to assess the effects of olivine additions on the primary producers' effect on the carbonate system. The samples were taken up with a syringe that was equipped with an 8-10 cm Iso-Versinic tube 4 mm (Saint Gobain, Courbevoie, France) and filtered with 0.22 µm syringe filters (Avantor VWR® Radnor, Pa, USA). The first 1-3 mL that went through the syringe filter were discarded due to the production residues of CO_2. The filtered samples were stored in amber glass vials with closed caps bonded with a PTFE-faced silicone liner to ensure a secure sealing (Thermo Fisher Scientific, Waltham, MA, USA) at 5°C until further analysis. TA and pH samples were measured using a Titrando 809 (Metrohm, Herisau, Switzerland) and an attached pH microelectrode, 6.0234.100 (Metrohm, Herisau, Switzerland), which was calibrated against four standard buffers daily before use (DuraCal pH buffer solutions, 2.00, 4.00, 7.00 Reagecon, Clare, Ireland, and a 10.00 from Hamiltoncompany, Reno, USA). TA was measured based on Dickson SOP3b (Dickson et al., 2007) using an open cell titration with the Titrando 809. To obtain alkalinity measurements, 5 mL of each sample were analyzed on the Titrando 809 with mixing and 0.1 mol L^{-1} (0.1 N) HCl (Merck, Rahway, NJ, USA) while 0.6 mol L^{-1} NaCl was added under stirring. The analysis used the Tiamo 2.5 software (Metrohm, Herisau, Switzerland). To verify the accuracy of the TA laboratory protocol a certified seawater reference was applied (Batch 187, Scripps Institution of Oceanography, University of California, San Diego, USA). DIC samples were analyzed using an AS-C5 DIC analyzer (ApolloSciTech, Newark, Delaware, USA) with a laser-based CO_2 detector. Before analyzing the samples, 2 mM $NaHCO_3$ was used as a standard, following the manufacturer's protocol, with three standard volumes at 0.9 mL, 1.2 mL, and 1.5 mL in addition to a certified seawater reference for CO_2 measurements (Batch 187, Scripps Institution of Oceanography, University of California, San Diego, USA). For analysis, a sample volume of 0.9 mL was extracted from each sample; the best of 3 out of a maximum of 5 measurements within the precision range of 0.02 on the AS-C6L DIC analyzer determined a single DIC point.

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2.6 Trace metal concentrations

For the elemental analysis of the water, samples were analyzed for concentrations of dissolved trace elements, including aluminum (Al), As, copper (Cu), Fe, Mg, Mn, and Ni, using an Agilent 7900 Quadrupole Inductively Coupled Plasma (ICP) Mass Spectrometers instrument (Agilent Technologies, Ca, USA). Calibration standards (20, 40, 60, 80, and 100 mg L^{-1}) were prepared in 5% nitric acid from 1000 mg L^{-1} ICP standards. Samples were prepared in 5% nitric acid and stored at room temperature until further analysis (Olesik, 2013). Samples were collected both at the start and at the end of the experiments, following the procedure described above. However, elemental data for strain CCMP489 and EUM Syn103 are not available.

2.7 Nutrient concentrations

During the experimental runs, we monitored the development of nitrate (NO_3^-), nitrite (NO_2^-), and phosphate (PO_4^{3-}). Samples were collected alongside every session for analysis. The nutrient samples were taken up with a syringe and filtered with (Avantor VWR® Radnor, Pa, USA) 0.22 µm syringe filters and stored in 1.5 mL Eppendorf microtubes (Eppendorf, Hamburg, Germany) at -20°C until further analysis. The following standards were used: a stock solution of KNO_3 for NO_3^-, $NaNO_2$ for NO_2^-, and KH_2PO_4 for PO_4^{3-}. Standard dilution series were at 0, 5, 10, 20, and 30 µmol. These solutions served as reliable references for the analysis of the respective nutrients. The samples were analyzed with a Multiscan GO spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA) on OD at 680 nanometers, according to (Grasshoff et al., 1999) protocol. Samples are to be found in raw data on request.

3 Results

This study aimed to examine the impact of small amounts of ground olivine additions on isolated primary producers. To achieve this, we conducted controlled experiments on primary producers to evaluate the ability of OAE in both coastal and open ocean environments. We aimed to discern the impact of olivine additions on the growth of these primary producers.

3.1 Responses of carbonate chemistry to olivine additions

3.1.1 Total alkalinity over the course of the experiments

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TA concentrations displayed variability in cultures of different species, as exemplified by the three *Synechococcus* strains (Fig. 1. a-c), while differences between primary producers were generally detectable. A similar pattern was observed in *Synechococcus sp*. strain A15-62 Clonal (Fig. 1. a) and *Prochlorococcus sp.* strain NATL2-M98 (Fig. 1. d). The influence of olivine additions on TA and the control groups varied between strains. Olivine additions had a bigger effect on TA concentrations during the experimental period for Synechococcus sp. strain WH8102 cultures compared to the control (Fig. 1. c). Among the *Synechococcus sp*. strains examined the most pronounced variations in carbonate chemistry were observed in the case of strain A15-62 Clonal, where olivine additions elevated concentrations to 3159 µmol kg^{-1} on day 6 (Fig. 1. a). It is worth highlighting that the olivine treatments, including the control, experienced a decline in TA concentrations throughout the experiment (Fig. 1. a). The EUM Syn10 strain showed similar TA concentration fluctuations between the control and olivine-treated groups. The olivine addition exhibited a low decline, decreasing from 3158 to 3121 µmol kg^{-1} at the end of the experimental period (Fig. 1. b). The control group showed a decrease in TA concentrations from the beginning to the end of the experimental period, with fluctuations occurring intermittently between days 2 and 9 (Fig. 1. b). In the *Synechococcus* strain WH8102, olivine additions and the control strain had very similar patterns showing declines in TA concentrations during the experimental phase (Fig. 1. c). The trend in TA concentrations was considerably similar among the olivine addition and the control for the *Prochlorococcus sp.* strains, both experiencing a decline from the beginning to the end of the experiment (Fig. 1. d-e). However, olivine additions showed some fluctuations, with TA concentrations increasing in the first few days of the experiments, peaking at day 4 with 3236 µmol kg^{-1} before declining again (Fig. 1. d). Similar observations appeared for olivine additions for the *Prochlorococcus sp.* strain EQPAC1-C, where TA concentrations increased from 3015 µmol kg^{-1} at day 0 to 3373 µmol kg^{-1} at day 2 before subsequently declining (Fig. 1. e). For *Scrippsiella trochoidea*, CCMP1331, TA concentrations increased for the olivine treatment and the control group during the experimental period, with olivine additions reaching the maximum peak at 3087 µmol kg^{-1} (Fig. 1. f). For *Micromonas commoda*, the fluctuations in the olivine treatments closely resembled those in the control group from day 3 until the end of the cultivation period (Fig. 1. g). For diatoms, the control displayed the highest values on TA concentrations, both in terms of general concentrations and maximum values, as exemplified by CCMP1051's 3880 µmol kg^{-1} (Fig. 1. h) and CCMP2092's 3827 µmol kg^{-1} (Fig. 1. i). For the *Emiliania huxleyi* strain,

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ACC448, the influence of olivine additions on TA concentrations varied, similarly did the control. For ACC448, olivine reached its maximum peak of 2742 µmol kg^{-1} on day 3 (Fig. 1. j).

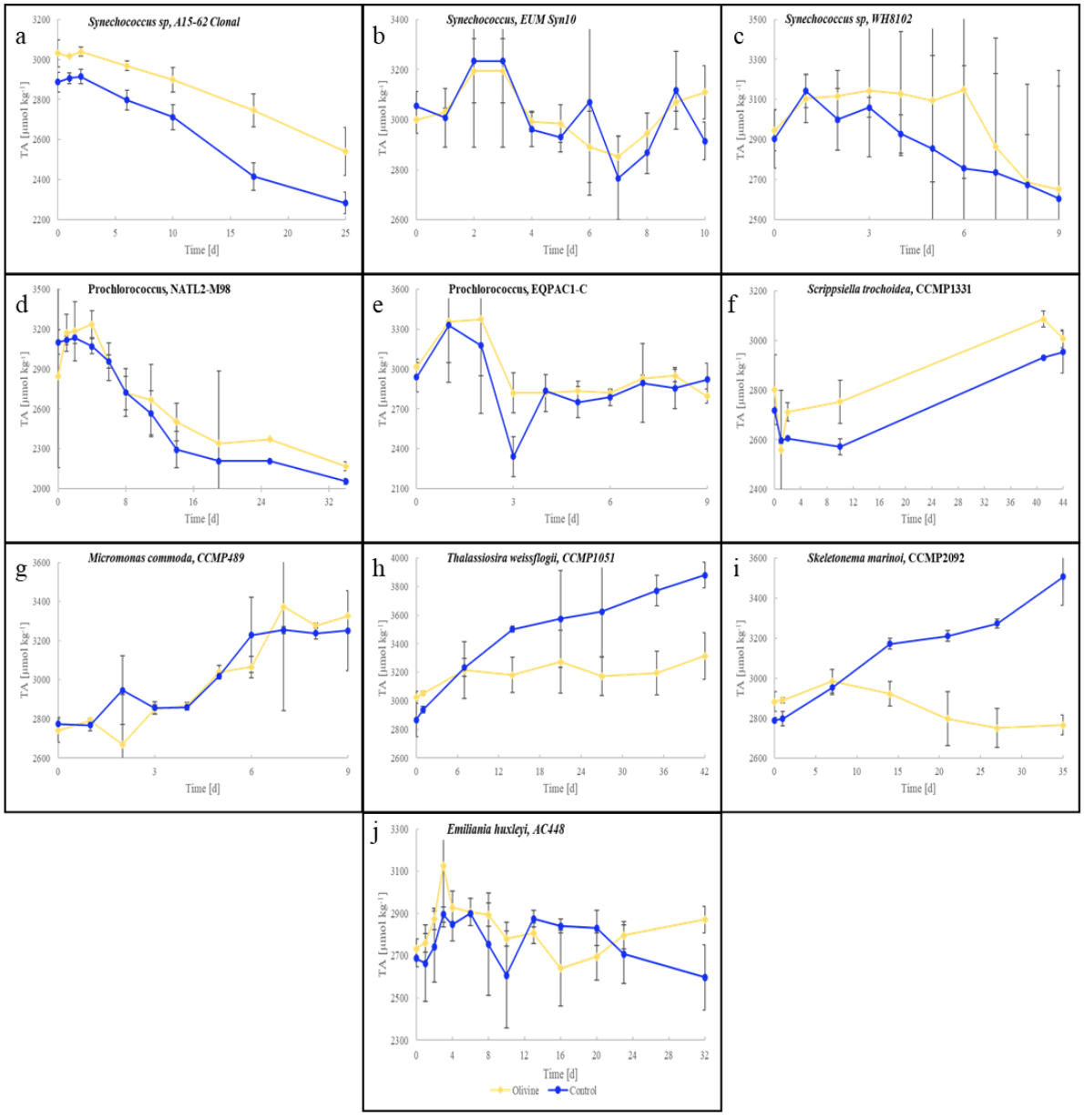

Figure 1. Total alkalinity concentrations (µmol kg^{-1}) in cultures exposed to olivine (yellow, diamond) additions and the control without any olivine supplement (blue, circle). The standard deviation is represented with grey bars.

3.1.2 pH changes over the course of the experiments

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Within our study, we observed pH across the primary producers examined. Despite the addition of olivine being predicted to be the primary driver of pH changes, some cultures also exhibited changes parallel with these in their control groups. The strains showing such analogous pH patterns include *Synechococcus sp.* EUM Syn10 (Fig. 2. b), *Scrippsiella trochoidea* CCMP1331 (Fig. 2. f), *Micromonas commoda* CCMP489 (Fig. 2. g), and the diatom strains CCMP1051 (Fig. 2. h), and CCMP2092 (Fig. 2. i). In multiple but not all cultivations, we noted an initial elevation in pH resulting from olivine additions compared to the control group (Fig. 2. a, d, e, f, and j). Certain incubations displayed an initial pH increase for specific olivine additions surpassing 0.2 relative to the control (Fig. 2. a and j). The *Synechococcus sp*. strain A15-62 Clonal displayed a pH increase from the introduction of olivine until the end of the cultivation period, ranging from 7.83 to 8.05 pH (Fig. 2. a). Similarly, in the *Prochlorococcus sp*. strain, EQPAC1-C pH increased from 7.8 to 7.92 (Fig. 2. e) after olivine additions. In contrast, pH levels declined from the initial incubation to the end of the cultivation period in response to olivine additions for the two diatom strains. For the diatom strain CCMP1051, pH decreased from 8.36 to 8.09 (Fig. 2. h), and strain CCMP2092 decreased from 8.60 to 7.76 pH (Fig. 2. i) during an incubation period of 35 days. *Emiliania Huxleyi*, AC448 showed a pH decline from 7.99 to 7.86 in response to the olivine additions during the experimental period (Fig. 2. j), the control strain maintained a pH range between 7.77 and 7.81 throughout the experimental period, with fluctuations occurring intermittently during the cultivation (Fig. 2. j). In the case of the pH changes for both the olivine additions and the control strain of *Synechococcus sp*. WH8102 varied during the 9-day cultivation period, fluctuating between 7.68 and 8.04 pH for the olivine additions and between 7.68 and 8.08 for the control strain (Fig. 2. c).

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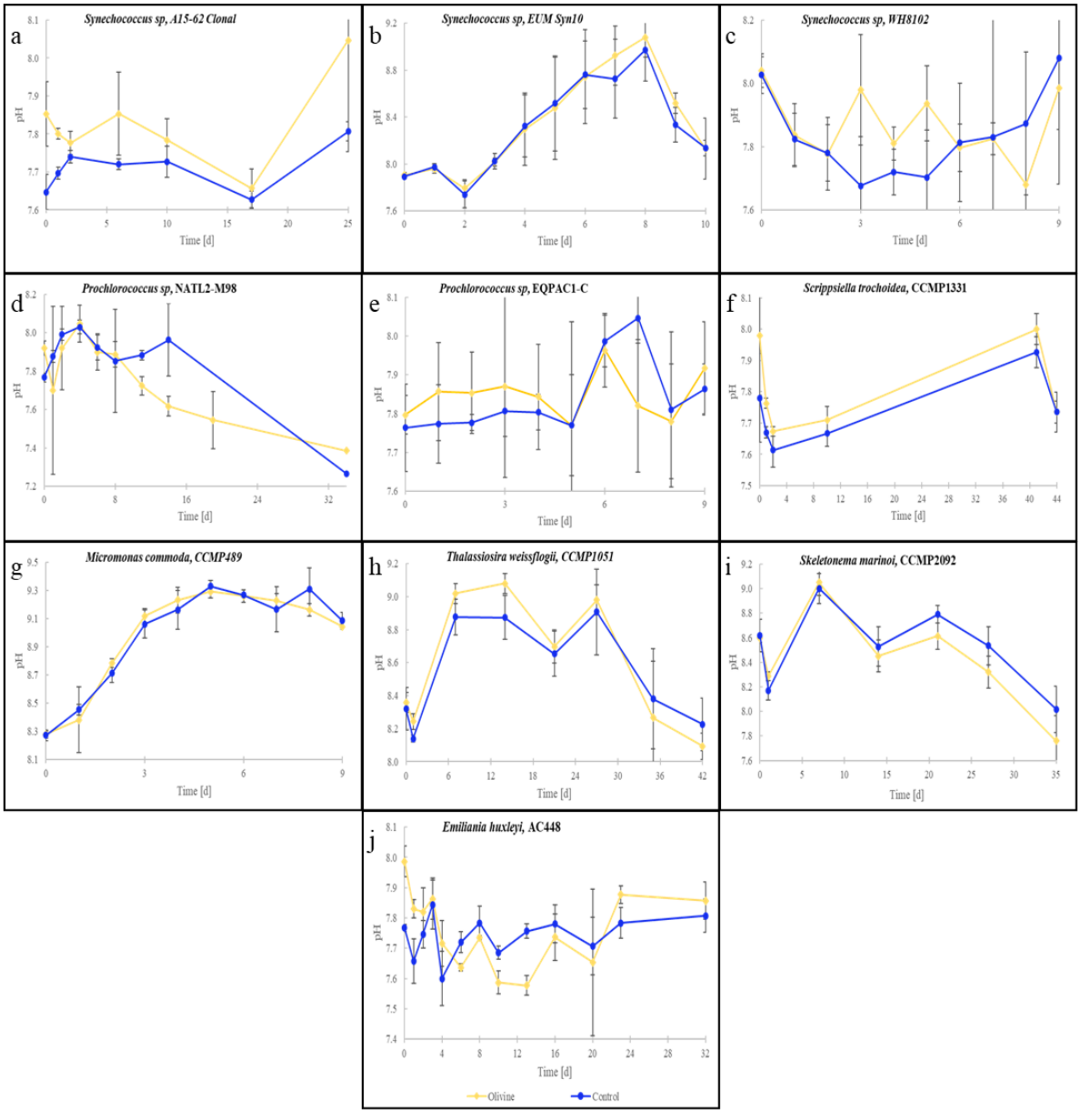

Figure 2. pH in cultures exposed to olivine (yellow, diamond) additions and the control without any olivine supplement (blue, circle). The standard deviation is represented with grey bars.

3.1.3 Dissolved inorganic carbon changes over the course of the experiments

The influence of olivine supplements on DIC concentrations within enclosed systems demonstrated substantial variability (Fig. 3). Throughout most of the experimental duration, the

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highest DIC concentrations were consistently observed in the control of the diatom strains CCMP1051 and CCMP2092 (Fig. 3. h and i). Among the *Synechococcus sp*. strains investigated, variations in carbonate chemistry were observed, with the strain A15-62 Clonal displaying the most visible changes from the beginning to the end of the experiment (Fig. 3. a). The addition of olivine influenced the DIC concentration by peaking at 2038.39 µmol kg^{-1}, whereas the DIC concentration of the control strain remained consistently lower throughout the experiment (Fig. 3. a). For the EUM Syn10 strain, the control alongside olivine additions had comparable developments in DIC concentrations (Fig. 3. b). In the case of *Synechococcus sp*. strain WH8102, olivine additions yielded a higher effect than the control group over the experimental period. The olivine addition reached a maximum peak of 3096 µmol kg^{-1} on day 5 (Fig. 3. c). Across the three *Synechococcus sp.* strains, the addition of olivine induced more pronounced changes in DIC concentrations in strains A15-62 Clonal and WH8102 compared to the control group, while strain EUM Syn10 exhibited similar changes to the control (Fig. 3. a-c). For the *Prochlorococcus sp.* strains, DIC concentrations have similarities between the olivine addition and the control. Nevertheless, the olivine additions resulted in the highest maximum DIC concentrations for both *Prochlorococcus sp.* strains NATL2-M98 at 2707.75 µmol kg^{-1} on day 1 (Fig. 3. d), and EQPAC1-C at 2782.60 µmol kg^{-1} on day 2 (Fig. 3. e), as well as for *Scrippsiella trochoidea* CCMP1331 at 3420.12 µmol kg^{-1} on day 44 (Fig. 3. f) compared to the control strain. Olivine additions seemed to have a negative impact on the DIC concentrations on the *Micromonas commoda*, CCMP489 strain throughout the experimental duration, compared to the control strain (Fig. 3. g.). We observed that olivine additions had a negative impact on DIC concentrations in the diatom strains compared to the control strain. Explicitly, both the overall increase and the maximum concentrations increased more in the control strain than in the olivine-treated strains (CCMP1051, 3501.42 µmol kg^{-1}; Fig. 3. h; and CCMP2092, 3734.43 µmol kg^{-1}; Fig. 3. i). Consistent with the patterns observed in TA concentrations (Fig. 1). The DIC concentrations for the *Emiliania huxleyi* ACC448 strain, olivine additions, had a considerable influence, with olivine having a pronounced effect until day 12 compared to the control group (Fig. 3. j).

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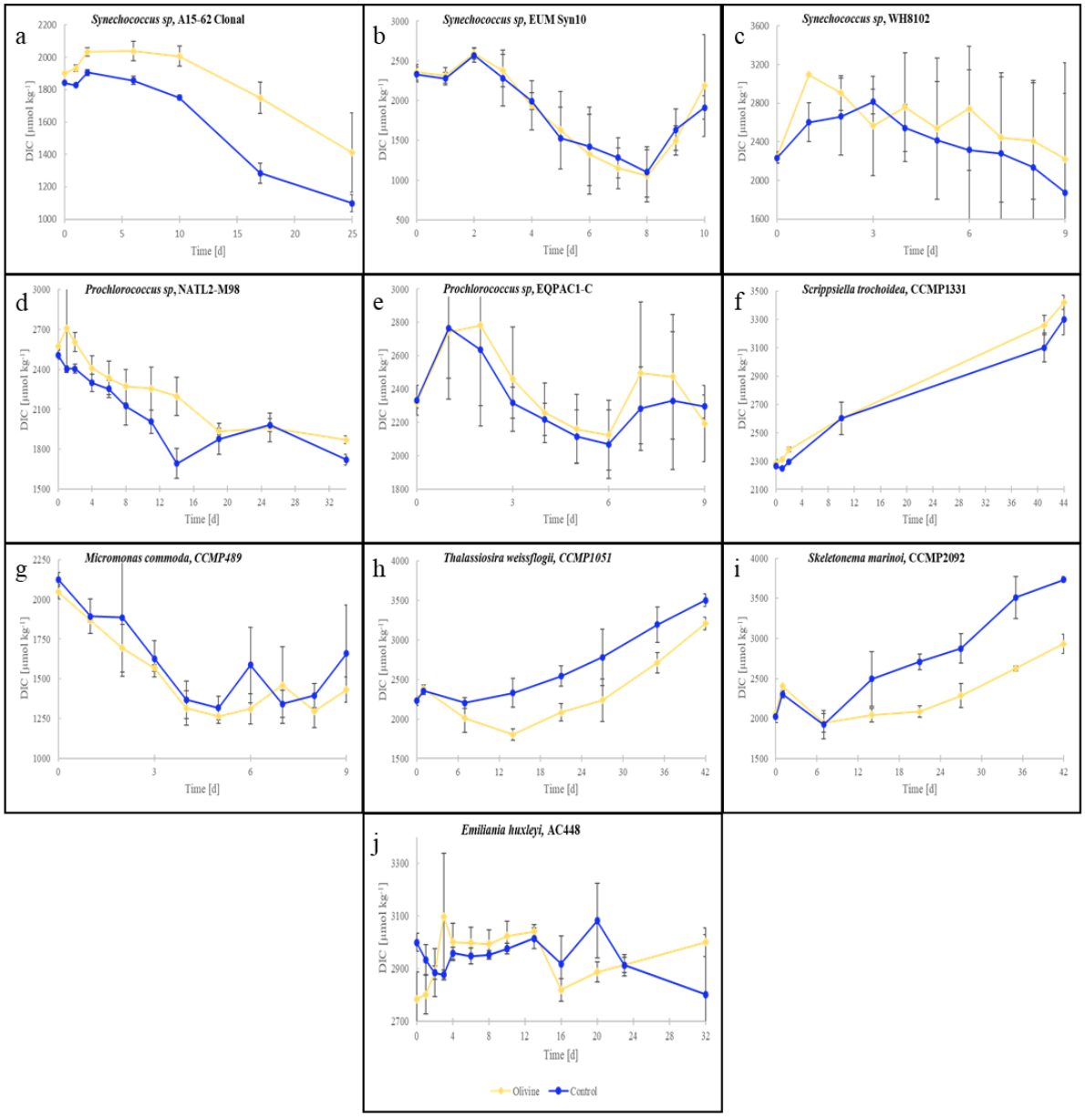

Figure 3. Dissolved inorganic carbon concentrations (µmol kg^{-1}) in cultures exposed to olivine (yellow, diamond) additions and the control without any olivine supplement (blue, circle). The standard deviation is represented with grey bars.

3.2 Impact of olivine additions on primary producer growth

Chl-a concentrations were utilized as the primary indicator for assessing the growth patterns of the primary producers. The chl-a concentrations reflected the primary production activity during the

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experiments. Fluorescence measurements were taken to obtain data supporting chl-a concentrations, including RFU and OD (S1-supplemental material). Overall, we did not find a solid response to olivine additions compared to untreated controls concerning chl-a development over the course of the experiment, except *Emiliania huxleyi* AC448, which did not show indications of an increase after adding olivine (Fig. 4, j). Strains generally and expectedly showed species-specific growth patterns with chl-a maxima ranging from 0.55 µg L^{-1} to 3222.99 µg L^{-1} between the cultivations (Fig. 4). The maximum chl-a concentration for *Synechococcus sp.* strains under olivine additions ranged from 84.10 µg L^{-1} on day 25 (Fig. 4. a), 14.71 µg L^{-1} on day 8 (Fig. 4. b), and 18.25 µg L^{-1} max (Fig. 4. c) on day 6. The control group demonstrated a higher chl-a concentration compared to the olivine additions for the *Synechococcus sp*. strain WH8102 throughout the experiment, except on day 6, the control group reached a maximum of 21.70 µg L^{-1} (Fig. 4. c). For *Prochlorococcus sp.* strains, the chl-a concentrations for olivine additions were between 614.91 on day 8 (Fig. 4. d), whereas the control group reached a maximum on 1430.00 µg L^{-1} and were generally at higher chl-a concentration than the olivine additions (Fig. 4. d). for the strain EQPAC1-C it reached a max on day 9 at 216.15 µg L^{-1} (Fig. 4. e). *Scrippsiella trochoidea*, CCMP1331, reached a peak chl-a concentration of 69.27 µg L^{-1} on day 41 following the olivine addition (Fig. 4. f). *Micromonas commoda*, CCMP489, exhibited a maximum of the olivine addition at 2947.73 µg L^{-1} on day 6, whereas the untreated control reached a peak at 2860.40 on day 6 (Fig. 4. g). The diatom strains' chl-a concentration peaked with maxima in the untreated controls between 3222.99 on day 21 for strain CCMP1051 (Fig. 4. h) and 3031.09 µg L^{-1} on day 21 for strain CCMP2092 (Fig. 4. i). The maximum chl-a concentrations of the *Emiliania huxleyi* strain were at 0.55 µg L^{-1} on day 23, reached in the control group (Fig. 4. j). Olivine addition was associated with low chl-a concentrations for the *Emiliania huxleyi* AC448 at 0.04 µg L^{-1} (Fig. 4. j).

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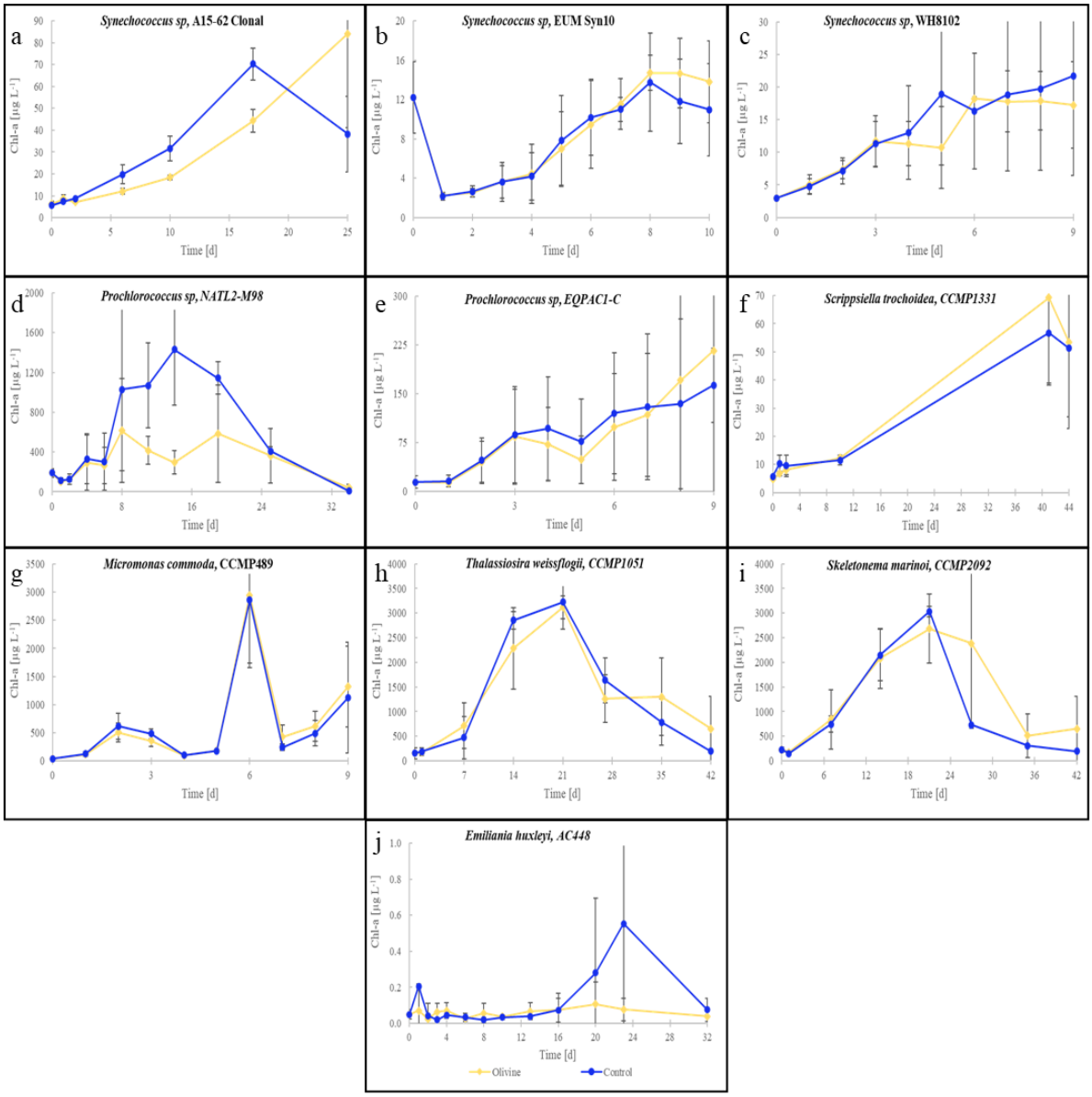

Figure 4. Chl-a concentration (µg L^{-1}) in cultures exposed to olivine (yellow, diamond) additions and the control experiments without any olivine supplement (blue, circle). The standard deviation is represented with grey bars.

3.3 Impact of olivine additions on carbon fixation rates

In contrast to a relatively low impact of olivine additions on chl-a concentrations, as described above, we do not see a uniform response of carbon fixation rates to alkaline mineral additions. We identified species-specific responses in carbon fixation rates in response to the addition of olivine

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(Fig. 5). The *Synechococcus sp.* showed carbon fixation rates in our experiment, ranging between 61.86213 C L^{-1} d^{-1} reached by the olivine addition (Fig. 5. a), 188.47021 C L^{-1} d^{-1} reached by the olivine addition (Fig. 5. b), and 159.55055 µmol C L^{-1} d^{-1} reached by the untreated control (Fig. 5. c). The carbon fixation rates at the end of the experiment were substantially higher than those at T0 for all three *Synechococcus sp.* strains (Fig. 5. a-c). For the *prochlorococcus sp.* NATL2-M98 strain, the carbon fixation rate reached 27.23686 µmol C L^{-1} d^{-1} by the untreated control group and 20.84817 µmol C L^{-1} d^{-1} in the olivine addition treatment (Fig. 5. d). The EQPAC1-C strain reached the highest carbon fixation rate with olivine addition, reaching 195.51166 µmol C L^{-1} d^{-1} (Fig. 5. e). The carbon fixation rates at the end of the experiment were substantially higher than those at T0 for the two *prochlorococcus sp.* strains (Fig. 5. d-e). *Scrippsiella trochoidea* exhibited a carbon fixation rate of 0.69189 µmol C L^{-1} d^{-1} following olivine addition (Fig. 5. f), whereas the control group demonstrated a carbon fixation rate at 0.35906 µmol C L^{-1} d^{-1} after 44 days, marginally surpassing the carbon fixation rate measured at T0 0.31303 µmol C L^{-1} d^{-1} (Fig. 5. f). *Micromonas commoda* showed a carbon fixation rate of 453.97966 µmol C L^{-1} d^{-1} following olivine addition (Fig. 5. g), the carbon fixation rates of both the control and the olivine treatment considerably exceeded those at T0 (Fig. 5. g). The diatoms displayed carbon fixation rates following olivine additions, reaching 69.08472 µmol C L^{-1} d^{-1} for the CCMP1051 strain (Fig. 5. h) and 44.18524 µmol C L^{-1} d^{-1} for the CCMP2092 strain (Fig. 5. i). In comparison, the untreated control showed carbon fixation rates of 56.29830 µmol C L^{-1} d^{-1} for the strain CCMP1051 (Fig. 5. h) and the strain CCMP2092 at 41.36381 µmol C L^{-1} d^{-1} (Fig. 5. i). Both diatoms demonstrated substantially higher carbon fixation rates at the end of the experiment compared to those at T0 (Fig. 5. h-i). The *Emiliania huxleyi* strain AC448 exhibited a carbon fixation rate of 1.40959 µmol C L^{-1} d^{-1} following olivine addition (Fig. 5. j), whereas the untreated control had a carbon fixation rate of 0.15304 µmol C L^{-1} d^{-1}, a rate much closer to T0 of 0.02907 µmol C L^{-1} d^{-1} in contrast to the olivine addition (Fig. 5. j).

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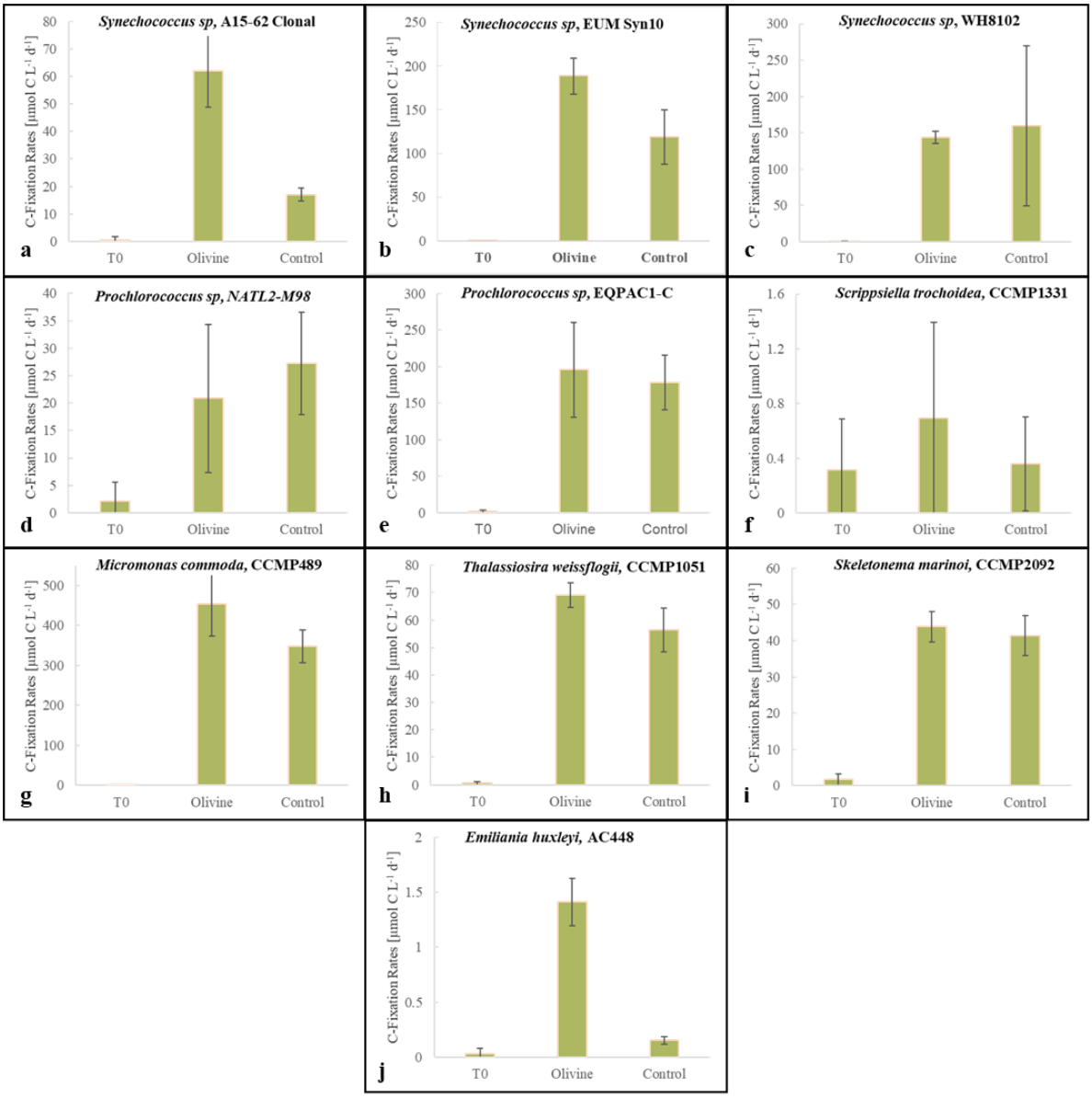

Figure 5. Carbon fixation rates (µmol C L^{-1} d^{-1}). T0 displays the initial biological triplicates at the beginning of the experiments. Olivine display the effect of ground alkaline minerals treatment at the end of the experimental period, while the control strain was cultivated without any alkaline mineral additions. The standard deviation is represented with grey bars.

3.4 Development of nickel content during the experimental period

Overall, the nickel concentrations observed in the various cultivation experiments were considerably higher in mineral addition experiments compared to the controls (Fig. 6). For

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instance, in both *Synechococcus sp.* strains exhibited nickel concentrations at the end of the experiment were observed. The strain A15-62 Clonal reached a nickel concentration of 1258.59 µg L^{-1} (Fig. 6. a), while strain WH8102 showed a concentration 536.63 µg L^{-1} (Fig. 6. b). However, in *Scrippsiella trochoidea* we found a decrease in nickel concentration since the start of the experiment, with nickel concentrations declining by 262.57 µg L^{-1} following the olivine addition reaching a nickel concentration of 36.96 µg L^{-1} at the end of the experiment (Fig. 6. c). The nickel concentration in the *Prochlorococcus sp.* NATL2-M98 experiment reached substantial concentrations, measuring 2347.08 µg L^{-1}, and experienced an increase of 1730.25 µg L^{-1} after the addition of olivine (Fig. 6. d). Conversely, the *Prochlorococcus sp.* EQPAC1-C, experienced a decrease in nickel concentration following the olivine addition at the beginning of the experiment with a decrease of 400.61 µg $L^{-1,}$ ending at a nickel concentration of 93.45 µg L^{-1} (Fig. 6. e). At the end point of the experiment, *Emiliania huxleyi* strain AC448 exhibited a nickel concentration of 583.38 µg L^{-1} following the addition of olivine. This concentration reflected a decrease of 325.78 µg L^{-1} from the initial olivine treatment (Fig. 6. f). The diatoms exhibited varying responses in nickel concentrations following olivine addition. For the CCMP1051 strain, the nickel concentration decreased by 208.33 µg L^{-1} to 972.07 µg L^{-1} towards the end of the experiment (Fig. 6. g), while the CCMP2092 strain experienced an increase in the nickel concentration by 115.60 µg L^{-1} increasing to 840.43 µg L^{-1} since the olivine treatment (Fig. 6. h).

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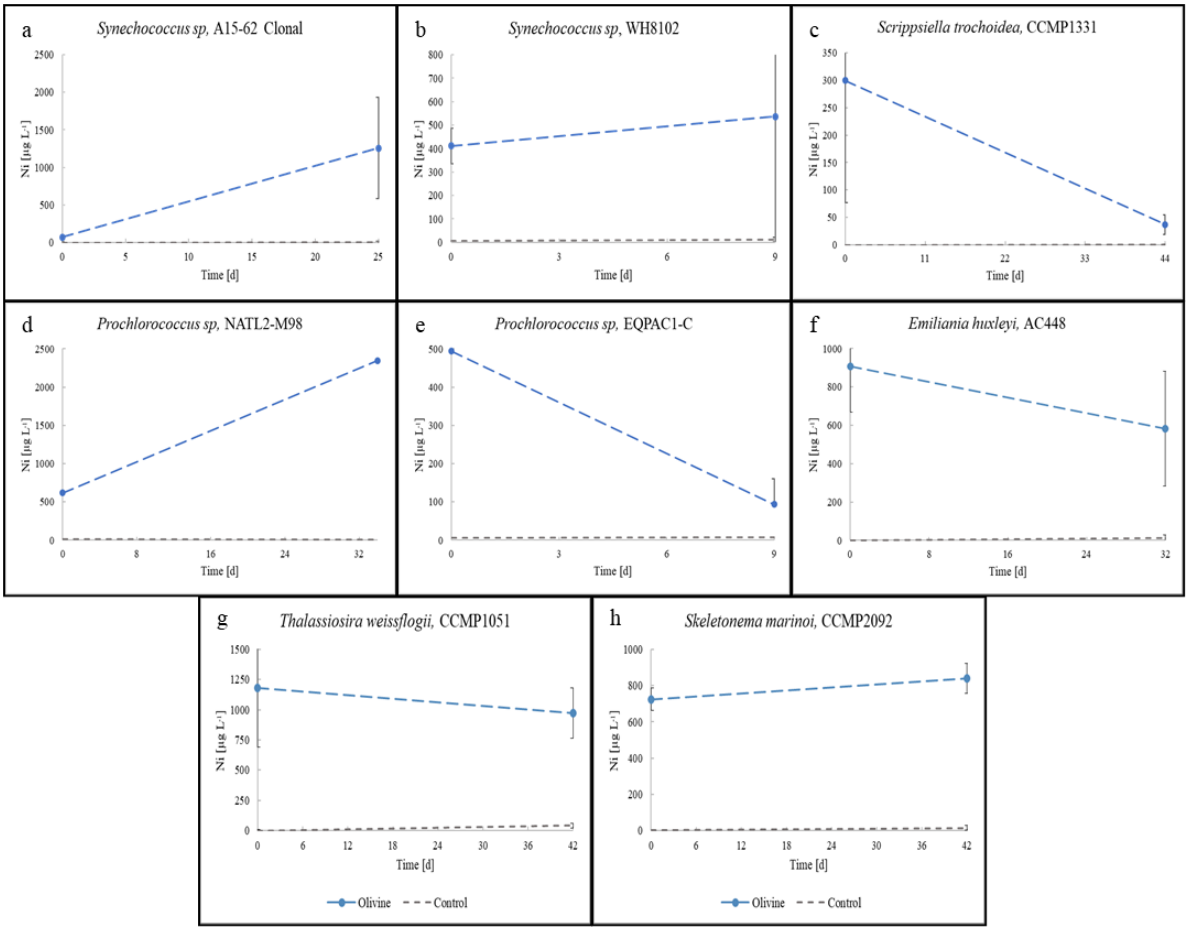

Figure 6. Nickel concentration (µg L^{-1}) in cultures exposed to olivine (blue, circle, long dash) additions and the control experiments without any olivine supplement (blue, small dash). The standard deviation is represented with grey bars.

4 Discussion

4.1 Experimental considerations and concerns

Our methodology in this experiment warrants consideration for its potential impact on the overall results. The use of different media for the various algae species may have influenced TA development (Fig. 1), particularly in the case of cyanobacteria cultured in PCR S11 Red Sea Medium (Tab. 3). The inclusion of HEPES buffer in PCR S11 Red Sea Medium could have mitigated the alkalization effect on pH. HEPES acts as a pH buffer, thereby stabilizing the aquatic cultivation media and promoting consistency in experimental conditions (McFadden and Melkonian, 1986). Additionally, the use of silica in the L1 medium, which was incorporated on

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for the diatom to enhance their growth (Tab. 3), might have resulted in co-precipitation within the cultivation medium following the addition of the alkaline mineral.

The addition of sodium bicarbonate, known to act as a buffer to maintain alkalinity in the solution, introduces an additional layer of concern. Sodium bicarbonate is recognized as a compound that effectively increases alkalinity (Furtado et al., 2011). Its inclusion prior to the olivine additions likely influenced the experimental conditions across all cultivations for the alkalinity. Therefore, the presence of sodium bicarbonate needs to be carefully considered when interpreting the results. Another apparent consideration is the potential impact of nutrient concentrations on alkalinity within the different media used in our experiments. The medias contain varying concentrations of nitrate (NO_3^-) and nitrite (NO_2^-) and ammonia (NH_4^+) (Tab. 3). Furthermore, the k and L1 media consist of seawater from the North Sea, potentially containing higher nutrient concentrations from land sources, while the PCR S11 Red Sea Medium is based on deionized water. This variability in nutrient composition raises concerns about the extent to which these nutrients have affected alkalinity concentrations, as a release of NO_3^- and NO_2^- decreases alkalinity and a release of NH_4^+ increases alkalinity (Wolf-Gladrow et al., 2007). Therefore, the differing nutrient compositions of the medias may have contributed to variations in alkalinity concentrations detected during our experiments.

The shaking or no shaking could potentially impact the growth of the primary producers in relation to the nickel concentrations (Tab. 3). However, it appears that this variable did not have a noticeable effect on either the growth of the organisms or the concentration of nickel (Fig. 4 and 6), as for example *Scrippsiella trochoidea* was not mixed and it does show good growth compared to most of the other primary producers.

This study is one of the first investigations directly examining the response of naturally occurring olivine additions on both primary producers and general carbonate chemistry (S2-supplemental material). In contrast to other, earlier studies (Delacroix et al., 2023; Hutchins et al., 2023), we worked directly with olivine, rather than other forms of alkalinity. This decision was made to mimic the process of OAE through the addition of mineral-based substances in aquatic conditions. By doing so, we aimed to replicate the complex interactions and potential impurities present in naturally occurring olivine, which may be challenging to explore when using artificial alkaline products.The presence of sand and other non-alkaline components in the olivine might further

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impact organisms and aquatic ecosystems. Moreover, the smaller grain size in this study, approximately ~63µm, distinguishes it from and earlier study Fuhr et al. (2022) in which grain sizes ranging from 100-125 µm and additions ranging between 25g and 100g per L were applied. While our additions were significantly lower compared to other studies, we used smaller grain sizes, thus increasing the reactive surface of the particles.

4.2 Potential effects of Ni on phytoplankton cultures

Our study could not identify fatal effects or growth inhibition on the cultivated primary producers exposed to olivine minerals despite increases in Ni concentrations for several species observed in the media following the olivine additions (Fig. 6). Neither chl-a concentrations nor carbon fixation rates were impacted compared to the other olivine addition and the control group. Interestingly, recent studies investigating the toxic impact of Ni on phytoplankton species yielded similar findings (Guo et al., 2022; Hutchins et al., 2023). Still, we observed an immediate and continuous increase in Ni concentrations from ground olivine. A recent study by Xin et al. (2023) directly investigated the impact of various concentrations of Ni on algae after instantaneous exposure (Xin et al., 2023). The study showed varying responses of phytoplankton strains to changes in Ni concentrations, with some strains being more impacted than others. Specifically, the coccolithophore *Emiliania huxleyi* and the dinoflagellate *Amphidinium carterae* were only mildly impacted. The investigated strain of *Emiliania huxleyi* did not show many signs of growth for either the control or the olivine addition (Fig. 4 and 5. j), but it did show a decline of nickel concentration in the aquatic media (Fig. 6. f). In our study on a dinoflagellate, carbon fixation rates in *Scrippsiella trochoidea* were observed to be the lowest compared to T0 (Fig. 5f). Interestingly, while nickel concentrations declined during the experiment (Fig. 6. c), chl-a concentrations increased (Fig. 4. f). This suggests that *Scrippsiella trochoidea* potentially were not impacted by the nickel concentrations in the media. Instead, it is possible that the *Scrippsiella trochoidea* had an alternative metabolic pathway to support its growth and biomass production.

In contrast, the diatom *Thalassiosira weissflogii* displayed a high inhibition in growth rate with increasing Ni concentrations, specifically at the three highest concentrations at 20 µmol L^{-1}, 50 µmol $L^{-1,}$ and 100 µmol L^{-1}. These findings imply that the diatom *Thalassiosira weissflogii* has a lower tolerance to Ni than the other cultures tested (Xin et al., 2023). Endpoint concentrations during our experiments were towards the lower end of the range tested in that study, and ranged

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between 0.6 – 40.0 µmol L^{-1} (Figure 6), depending on cultivation media and primary producers. These concentrations are still several orders of magnitude higher than ocean average Ni concentration in surface waters at 2.1 nmol kg^{-1}, with a maximum of 11 nmol kg^{-1} in the deep sea (Bruland, 1980). Still, our study can confirm the results by Xin et al. (2023), with even high concentrations of Ni not significantly impacting the growth of the primary producers resulting from the olivine additions.

In contrast, Hutchins et al. (2023) observed similar patterns regarding the response of different phytoplankton taxa towards Ni exposure. Their study describes that most of the investigated primary producers were irresponsive to the exposure of the applied Ni concentrations. Interestingly, the synthetic olivine dissolution applied in Hutchins et al. (2023) had lower Ni concentrations compared to that in Xin et al. (2023); the experimental designs hinder a precise comparison between the studies discussed. This is in accordance with other recent observations studying OAE scenarios regarding trace metals (Guo et al., 2022). Guo et al. (2022) specifically focused on elevated Ni exposure in a broad range of phytoplankton groups; similar to our study, they did not observe any strong effects across the investigated taxa. It is important to note that we are unaware of the specific chemical species of dissolved Ni affecting the physiology of primary producers. However, evidence suggests that phytoplankton are primarily sensitive to free Ni^{2+} ions rather than the total concentration of dissolved Ni (Guo et al., 2022).

As Flipkens et al. (2021) proposed, establishing guidelines for trace metals, including Ni and Cr, would be highly advantageous to ensure the safe implementation of coastal enhanced weathering. Extending these guidelines to cover OAE would be equally beneficial, helping to mitigate uncertainties in marine-based CDR while considering various trophic levels (Flipkens et al., 2021). Additionally, to avoid the ecological concerns regarding potentially toxic trace metals from alkaline minerals in OAE processes, trace metals like Ni could be extracted, for instance, from olivine. (Santos et al., 2015).

4.3 Effects of olivine additions

Our experiments suggest that adding olivine is either slightly beneficial or did not negatively impact the tested primary producers, with no harm to growth parameters such as the carbon fixation rates, and chl-a concentrations (Fig. 4 and 5). Our primary producers were cultivated in specific

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growth media that provided all necessary nutrients for growth (Tab. 3), ensuring they had abundant essential components for optimal growth. This approach minimized bias and sensitivity, which can be more pronounced in nitrogen and phosphate-limiting marine environments, but it also added a layer of concerns and considerations to our experiments as mentioned above in the discussion section 4.1. Consequently, the addition of olivine is expected to have a lesser impact on growth in these conditions. Additionally, our primary producers were cultivated in closed systems that experience no natural mixing and blending with sea- or freshwater. Residence times of the olivine additions in the water column are relatively brief (He and Tyka, 2023), implying that dissolution in natural settings is expected to occur more rapidly than in our experiments. Our study deliberately introduced substantial quantities of pulverized olivine to conservatively explore their impact on primary producers.

Consequently, we anticipate that the olivine additions in our experiments likely encountered considerably higher concentrations of dissolved components from the olivine additions than under typical field or natural conditions. Our experiments primarily emphasized the responses of primary producers regarding a select set of physiological indicators and carbonate chemistry. Future investigations should broaden the scope to assess the effects on various trophic levels and encompass phytoplankton belonging to different central taxonomic groups.

4.4 Where could ocean alkalinity enhancement be applied?

Our study has revealed the potential use of olivine. To determine suitable regions for implementing OAE practices, it is crucial to comprehend the regional and local variations in primary producers and the aquatic chemistry.

In our experiments, we have demonstrated that regions characterized by abundant phytoplankton populations, particularly picophytoplankton (cyanobacteria and Micromonas), tended to exhibit either positive or neutral responses to olivine additions factors like growth rates, carbon fixation rates, and TA concentrations. The diatoms control group exhibited slightly higher increases in chlorophyll-a concentrations compared to the olivine additions (Fig. 4). However, olivine additions had a greater impact on carbon fixation rates for the diatoms (Fig. 5). Interestingly, physical processes such as eddies, coastal upwelling, wind, or convective mixing in oceanic mechanisms can promote the occurrence of diatom blooms or larger cells of primary producers as

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nutrient-rich waters enter the surface from the deeper ocean (Walsh et al., 1978; Olaizola et al., 1993; Falkowski et al., 1998) and most likely also.

However, satellite measurements indicate a global decline in diatoms between 1998 and 2012 within the surface mixed layers (Rousseaux and Gregg, 2015). This decline is critical since diatoms contribute approximately 40% to marine primary productivity and serve as a primary source of carbon export to the deep ocean (Tréguer et al., 2018). Consequently, an increase in nutrients from for example olivine in targeted regions, driven by the aforementioned physical processes, can enhance CO_2 export to the deeper ocean by promoting diatom growth. In contrast, nutrient-poor waters, mainly populated with small primary producers like picocyanobacteria, particularly in central ocean gyres (Falkowski et al., 1998), result in minimal carbon escape from remineralization (Azam, 1998).

With this, satellite data analysis can aid in identifying regions of interest (Righetti et al., 2019). Specifically, satellite imagery has proven instrumental in identifying four major groups of phytoplankton (Kramer and Siegel, 2019). Furthermore, these satellite-derived measurements can contribute to understanding net primary production in oceanic regions.

(Longhurst et al., 1995; Behrenfeld and Falkowski, 1997). Combined with our dataset, this is essential knowledge when considering additions olivine in coastal or open oceans. Based on our findings, we suggest considering locations such as the Labrador Sea, the North Atlantic, and the Eastern China Sea to explore OAE practices further. The following steps of OAE research should explore the underlying mechanisms of growth responses in phytoplankton, extend to ecosystem levels, and identify potential synergies and trade-offs between different alkaline minerals and primary producers to inform the progress of sustainable OAE strategies for marine ecosystems (Eisaman et al., 2023).

5. Conclusion

We aimed to assess the influence of olivine additions on various cosmopolitan primary producers, considering a moderate range of solid-liquid ratios. Our data suggests that olivine additions did not lead to growth inhibition across cultivated cultures, with the picoplankton benefiting the most.

Our findings further indicate that Ni exposure from olivine additions did not inhibit phytoplankton growth in our cultures. While our study provides valuable insights into the responses of certain

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cosmopolitan primary producers to olivine additions, it is important to recognize its limitations. Our study focused on phytoplankton responses in controlled laboratory conditions, which may not fully capture the complexities of natural ecosystems. Future research should aim to incorporate a community or ecosystem-level perspective to better understand the broader impacts of olivine additions in natural settings.

Additionally, it is essential for future studies to address potential pitfalls and concerns associated with our experimental setup, particularly regarding considerations of carbonate chemistry. By addressing these limitations, future research can provide more robust and comprehensive insights into the effects of olivine additions on marine ecosystems.**Data availability:** Data are presented in the supplement.

Author contribution: JR and CL designed the study. JR, ZK and MJ carried out laboratory experiments and analyses. JR analyzed the data and wrote the manuscript together with CRL, and input of all co-authors.

Competing interests: At least one of the (co-)authors is a member of the editorial board of Biogeosciences.

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