Elevated Temperature and Carbon Dioxide alter the Tea Rhizosphere Soil Dynamics

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ABSTRACT

Aim: The aim of this work is to explore the impact of climate change on the tea rhizosphere soil dynamics. The experiment sought to study the tea rhizosphere Soil Organic Carbon (SOC), Total Nitrogen (TN), Microbial Biomass Carbon (MBC) and Microbial Biomass Nitrogen (MBN) under elevated levels of temperature and Carbon Dioxide (CO_2). **Materials and Methods:** The experimental design was set up with two Open Top Chamber (OTC) facilities, creating localized conditions with increased temperature and CO_2. Both temperature and CO_2 were elevated in OTC-1 (eTemp+eCO_2), with CO_2 concentration of 550 ppm, while in OTC-2 (eTemp), only the temperature was elevated. Temperature was 1.5-2°C higher than ambient. Four tea cultivars (TV1, TV20, TV22 and TV23) were placed inside the OTCs and rhizosphere soil samples were collected at regular intervals. **Results:** The results indicated that in eTemp+eCO_2 treatment, over the period of time, the rate of change in SOC showed significant increase ($p<0.01$) compared to control, while TN showed no significant variations, the rate of change in MBC showed significant increase ($p<0.05$) in eTemp treatment. No significant variation was observed in the rate of change in MBN. Overall, SOC and MBC, after 300 hr of treatments showed significant increase ($p<0.001$) in both eTemp and eTemp+eCO_2 treatments when compared to control. However, after 300 hr of treatment overall soil TN exhibited a significant increase ($p<0.05$) in eTemp+eCO_2 treatment. **Conclusion:** This study addresses the research gap through an evidence based experimental work that monitors the impact of temperature and CO_2 on tea rhizosphere. From the study, it can be concluded that elevated temperature and CO_2 alter the rate of change in soil organic carbon pool of tea rhizosphere soil along with the accumulation of biomass carbon, whereas no such alteration was found for microbial biomass nitrogen.

Keywords: Climate change, Tea, Rhizosphere, Soil organic carbon, Microbial biomass nitrogen.

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INTRODUCTION

Climate change is one of the emerging global issues, with far-reaching impacts on ecosystems, human health, and economic development. One of the main drivers of climate change is the increasing concentrations of Carbon Dioxide (CO_2) in atmosphere. CO_2 concentrations in the atmosphere have surged by 31%.in the last 150 years from 280 parts per million (ppm) to 380 ppm.[1] The rising atmospheric CO_2, with its greenhouse effects, results in an increase in global temperature. The trend of climate change has also been creeping into the picture of North East India resulting in declining rainfall, an increase of minimum temperature, and carbon dioxide.[2] As temperature rises by an average of 0.18°C each decade (2.52°C increase in overall global temperature) and precipitation patterns shift,[3] understanding the consequences of climate change on the world's most vital agricultural systems is becoming increasingly crucial. One such system is the tea rhizosphere, which provides the ideal environment for developing

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specialized bacteria that play a vital role in plant health and nutrient uptake.[4,5] In this research article, we sought to explore the influence of climate change on tea rhizosphere microflora and its subsequent effects. The rhizosphere and tea plant relation are critical in maintaining the growth-promoting effects of some bacteria[6] and can help mitigate diseases brought forth by the changes in the climate.[7]

In this research, we will analyze the alteration in the Soil Organic Carbon (SOC), Total Nitrogen (TN), and Microbial Biomass Carbon (MBC) and Biomass Nitrogen (MBN) of tea rhizosphere bacteria in response to changes in climate. We assessed the response of these alterations on the dynamics of microbial population with respect to their carbon and nitrogen assimilation potential as a measure of their ability to support tea plants. By exploring the interplay between climate changes, the tea rhizosphere soil, we hope to provide insights into the potential consequences of climate change for the tea industry and the measures that can be taken to mitigate these effects.

MATERIALS AND METHODS

Experimental site

The study was steered in the experimental area of Tocklai Tea Research Institute, Assam, India (26º43"N and 94º13"E). The experimental design was set up as factorial RBD with three treatments and four clonal types with five replications each. Two Open-Top Chamber facilities (OTC) set up in the institute in 2012 were utilized for creating localized conditions with increased temperature and CO_2. Both temperature and CO_2 were injected in OTC1 (eTemp+eCO_2) at a concentration of 550±13 ppm, while in OTC2 (eTemp), only the temperature was elevated. Temperature was 1.5-2ºC higher than the ambient temperature. An open region with ambient temperature and CO_2 was set up as the control.

Plant and soil experimental setup

Nursery plants of four Tocklai released cultivars were collected and planted in pots in September 2017. The cultivars were TV1, TV20, TV22, and TV23. Sixty pots of 17cm height and 23 cm diameter were filled with ~7.5 kg of sieved soil collected from Borbhetta Tea Estate of Jorhat, Assam, and an initial soil analysis was done to get the soil status. The parameters analyzed were SOC, TN, MBC and MBN.

Carbon dioxide and temperature elevation

The OTC facility was calibrated with a carbon dioxide analyzer before shifting the potted plants to the chambers. Potted plants of different cultivars were placed inside the two chambers (OTC1 and OTC2) and in ambient conditions in January 2020. After acclimatizing the plants inside the OTCs, one of the chambers (OTC1) was enriched with carbon dioxide from March 2020 at 550±13 ppm from a carbon dioxide cylinder. Carbon dioxide, humidity, and temperature data of the OTCs and ambient conditions were recorded daily. Enrichment was done for 5 hr per day up to 375 hr.

Soil sampling

The rhizospheric soil samples were collected from each cultivar, TV1, TV20, TV22, and TV23 at an interval of 0, 75, 150, 225, 300, and 375 hr of treatments. Sixty (60) soil samples were obtained from OTC1. OTC1 (eTemp+eCO_2), OTC2 (eTemp), and control at each interval; i.e., 20 samples from each treatment. Soil samples were gathered with the aid of an auger from a depth of 0-15 cm. Soil adhering tenaciously to the tea roots was obtained for the study following the method described by Pandey and Palni.[8]

Analysis of SOC, TN, MBC and MBN

Soil Organic Carbon (SOC) was determined using Walkey and Black oxidation method.[9] Sieved 0.5 g of air dried soil was weighed, and the organic C in the soil was oxidized by 10 mL 1N $K_2Cr_2O_7$ solution in concentrated H_2SO_4. The SOC was then estimated by back titrating with 0.5 N ferrous ammonium sulphate and diphenylamine (indicator). Total Nitrogen (TN) was calculated using Kjeldahl distillation method.[10] 5 g air dried soil was digested with H_2SO_4, followed by distillation where ammonia released was absorbed in 10 mL 2% boric acid solution for 10 min. Concentration was quantified by titrating against done against 0.005 N H_2SO_4 solution.

While MBC and MBN were determined by the fumigation incubation method.[11] 10 g of fresh soil was fumigated for 24 hr in a desiccator. 2 mL of 0.2N K_2SO_4 was added to the fumigated and unfumigated soil and shaken for 30 min.

Statistical analysis

Factorial ANOVA was used to ascertain the significance of the observed changes. All the statistical analysis was done using GraphPad Prism ver. 9.3.1.

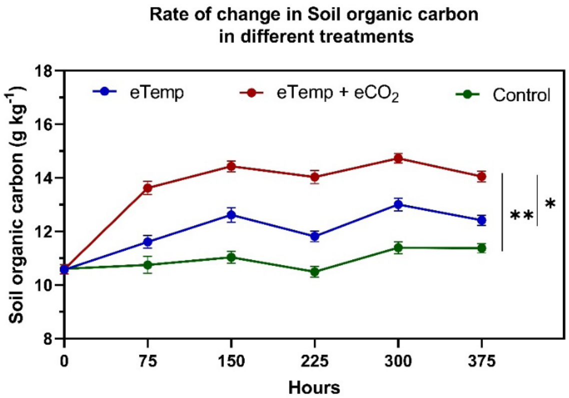

Figure 1: Rate of change in soil organic carbon (SOC) in tea rhizosphere soil after 375 hr of treatment in the three different treatments.

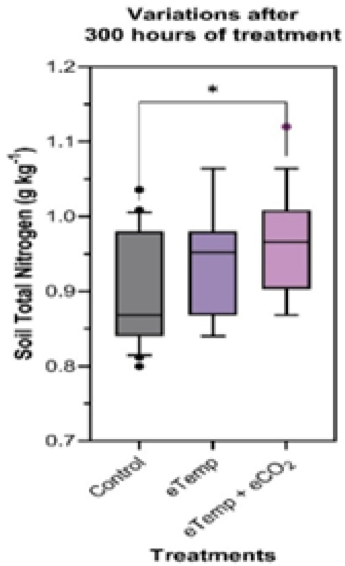

Figure 3: Boxplot showing overall variations in soil Total Nitrogen (TN) content in different treatments after 300 hr.

RESULTS

Rate of change in SOC from the tea rhizosphere soil after 375 hr enrichment was observed. SOC showed significant increase in eTemp+eCO_2 ($p<0.05$ and $p<0.01$) treatment when compared to both eTemp treatment and control respectively (Figure 1). After 300 hr of treatment, overall, SOC showed significant increase ($p<0.001$) in eTemp as well in eTemp+eCO_2 treatment when compared to the control. The highest value was recorded in eTemp+eCO_2 treatment with an average of 14.73 ± 0.76 g kg^{-1} soil followed by the eTemp treatment with 13.01**±1.04** g kg^{-1} soil (Figure 2). After 375 hr of treatment, the rate of change in soil Total Nitrogen (TN), showed no significant increase in both eTemp and eTemp+eCO_2 treatments when compared to control. However, after 300 hr of treatment overall soil TN exhibited a significant increase ($p<0.05$) in eTemp+eCO_2 treatment in comparison to control (Figure 3). Similarly, the rate of change in the soil MBC of the tea rhizosphere after 375 hr was observed. The rate of change in MBC showed significant increase in eTemp in comparision to control ($p<0.05$). No significant variation was observed in eTemp+eCO_2 treatment when compared to the control (Figure 4). Whereas, after 300 hr of treatment, overall MBC exhibited a significant

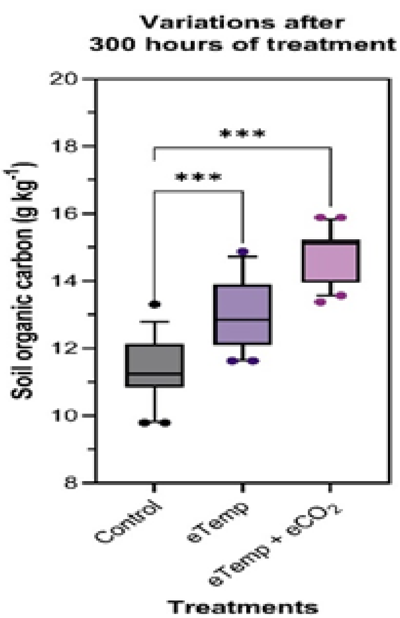

Figure 2: Boxplot showing overall variations in Soil Organic Carbon content (SOC) in different treatments after 300 hr.

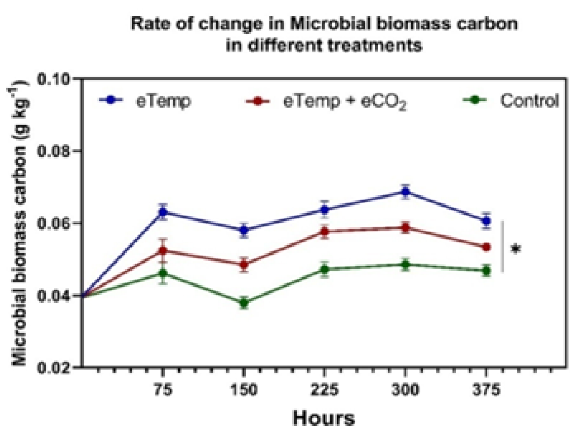

Figure 4: Rate of change in Microbial Biomass Carbon (MBC) in tea rhizosphere soil after 375 hr of treatment in the three different treatments.

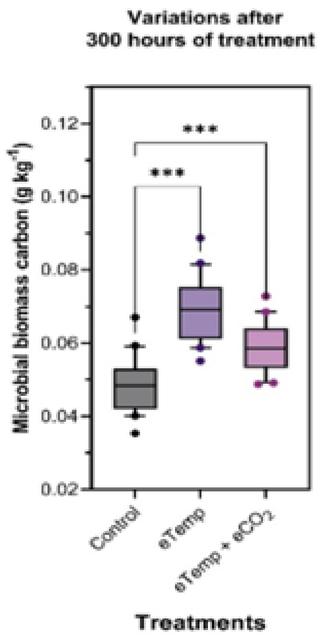

Figure 5: Boxplot showing overall variations in Microbial Biomass Carbon (MBC) content in different treatments after 300 hr.

increase (p<0.001) in both eTemp and eTemp+eCO_2 treatments when compared to control. The highest MBC value was recorded in eTemp treatment with an average of 0.069**±0.01** g kg^{-1} soil followed by the eTemp+eCO_2 with 0.059±0.01 g kg^{-1} soil (Figure 5). The rate of change in Microbial Biomass Nitrogen (MBN) showed no significant increase in both eTemp and eTemp+eCO_2 treatments. Likewise, after 300 hr of treatment, overall MBN showed no significant increase in eTemp+eCO_2 treatment compared to the control.

DISCUSSION

Climate change is an inevitable aspect of modern developmental society, which has led to an increase in temperature accompanied by CO_2 concentrations all over the globe. Nowhere is this more evident than in tropical regions, which have resulted in biodiversity loss and increased arid-humid transitions.[12] Soils and soil microbiomes are different everywhere. Soils differ substantially in terms of their biotic as well as abiotic characteristics. As a result, generalising the impacts of climate change on soil microbiomes across diverse soil ecosystems is difficult.[13] Soil carbon helps to restore degraded soils which can improve tea yield and productivity. Healthier soils make tea plants more resilient against harsh environmental conditions. Grace in 2006 have predicted a 6.4 % (518 metric tons) increase in topsoil C concentration by 2100 under high CO_2 emissions in parts of Australia,[14] similarly, our research results indicated parallel trend with increase in SOC in elevated temperature and carbon dioxide. Our findings were likewise aligned with those of Allen and his co-authors. According to their findings, SOC accumulation in peanut and bahiagrass plots increased significantly (p<0.01) by 0.94 g kg^{-1} and 1.20 g kg^{-1} at different levels (ppm) of CO_2 enrichment, respectively. Similarly, at increasing temperatures of +1.5ºC and +3.0ºC , the total SOC increased by 1.21 g kg^{-1} and 0.97 g kg^{-1}, respectively.[15] Similarly, Pendall and co-workers reported in 2004, that in grassland ecosystems, rhizodeposition of carbon to SOC was doubled under higher CO_2 conditions.[16] Our data also revealed that prolonging the treatments from 300 to 375 hr resulted in a steady decrease in the rate of SOC formation. This might be owing to the treatments' prolonged exposure, which could lead to a saturation point in the carbon cycle, resulting in a reduction in SOC accumulation. Another research found that rice rhizosphere soil acquired from increased CO_2 chambers accumulated more carbon than soil collected from ambient CO_2 chambers.[17] In contrast, Oelbermann and colleagues discovered that the rhizosphere soil of cereal legumes collected at increased temperatures and CO_2 levels resulted in a significant drop (p=0.001) in SOC concentration.[18] Thus, the effect of increased temperature and CO_2 on SOC accumulation in the rhizosphere is plant specific. Rate of change in soil Total Nitrogen (TN) in both treatments exhibited no significant increase; however, variations were noted after 300 hr i.e., TN exhibited a significant increase in elevated temperature and CO_2 treatment when compared against control. Our study's overall findings were comparable to those of Allen and his colleagues. Long-term exposure to both temperature and CO_2 can enhance soil nitrogen, according to their research. The influence of temperature on soil nitrogen was significant (p<0.01), but the effect of higher CO_2 increased it by 0.09 g kg^{-1}.[15] Xu and Yuan, on the other hand, reported in 2017 that the indirect warming approach employing the OTC methodology dramatically reduced the TN content of the soil.[19] Similarly, warming without the addition of CO_2 had almost no effect on nitrogen content.[20] Another report by Figueiredo and his team found that the accessible nitrogen forms did not respond to increased temperature.[21] As a result, raising the temperature or CO_2 can cause specific differences in SOC and TN. The biotic activity of the tea rhizosphere population is of great concern for the health of the tea plant, as the activities of the tea rhizosphere bacteria are an essential factor in promoting the plant's growth. Both Diaz (1993) and Zak (1993) found increase in microbial biomass with elevated CO_2.[22,23] In our results we also observed a higher rate of incorporation of C into the soil and a significant increase in the portion of Microbial Biomass Carbon (MBC) under elevated temperature treatment. A similar pattern was reported in the study of Xu and Yuan, 2017, who discovered a 3.61% increase in MBC in higher temperature compared to ambient temperature.[19] Belay-Tedla and his team investigated that experimental warming increased soil MBC in the United States' Great Plains.[24] Manna and his co-workers reported in 2013 that MBC of rice rhizosphere soil samples stored at elevated CO_2 levels increased around two folds compared to controls. This increase might be attributed to higher soil exudates in rice grown in high CO_2 conditions.[25] Another study on the effect of increasing CO_2 on rice crops discovered that MBC in rice grown in an increased CO_2 environment was much higher than rice soil grown in ambient CO_2.[26] Whereas, in contrast to our findings, it was pragmatic that under high temperatures the MBC in an alpine meadow on the Tibetan Plateau dropped.[27] As a result, the MBC proportion in the rhizosphere soil might be regarded as plant-specific, potentially leading to increased biomass carbon uptake. The MBN displayed a huge spatial heterogeneity within the soil, CO_2 enrichment had no discernible effect on the MBN within its soil profile.[28] According to the works of Allen, no significant difference was observed in microbial biomass nitrogen due to CO_2 treatment on any sample.[15] Similarly, in our study, it was observed that the microbial biomass nitrogen showed no significant variations when compared to control. Earlier research revealed that enhanced nitrogen availability in the rhizosphere soil of coniferous plants increased MBN. This meant that the net increase in total nitrogen was reflected in the quantity of nitrogen absorbed into biomass by microorganisms.[29] The rise in nitrogen mineralization and nitrification is evident in the soil nitrogen pool and its microbial biomass.[30] These outcomes are consistent with our findings, in which the total nitrogen rate did not exhibit any significance in either treatments and therefore it was reflected in the soil MBN. However, it was shown that increasing the duration of the treatments from 300 to 375 hr resulted in a progressive decrease in the rate of MBN. This might be due to the treatments' prolonged exposure, which could have resulted in a saturation point among the bacteria engaged in nitrogen cycling, which incorporates nitrogen into its biomass. Nonetheless, we have to consider that the lack of CO_2 effect on MBN does not certainly imply that there is no influence on soil microflora, changes in microbial community might occur without changes in its MBN accumulation. The findings of our results are based on potted cultivars in earthen planters, but the same experimental study might be done within the field conditions considering our methods, which would emphasize our results more clearly. Thus, this study fills a research gap by monitoring the impact of temperature and CO_2 on the tea rhizosphere.

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CONTRIBUTION OF AUTHORS

ASR, JB and SRS designed the experiment; ASR performed the soil data analysis. ASR wrote the manuscript draft while JB and SRS reviewed it. The statistical analysis, along with the graphs, was prepared by ASR and RDB. ASR and SRS finalized the manuscript.

CONFLICT OF INTEREST

The authors find no conflict of interest and would like to report the same.

ETHICAL CLEARANCE

The authors want to declare that no animals were used for these experiments; hence, no ethical clearance is required.

ABBREVIATIONS

SOC: Soil organic carbon; **TN:** Total nitrogen; **MBC:** Microbial biomass carbon; **MBN:** Microbial biomass nitrogen; **OTC:** Open Top Chamber; **TV:** Tocklai Vegetative; **C:** Carbon.

SUMMARY

Tea is an important economic crop for the regions of North East India; however, the recent change in

climate has led to significant changes in the timings of tea pruning and the appearance of new leaves. In the last decade, tea cultivation has changed its practices. Thus, the tea-growing soil of North East India needs to be better understood regarding the impact of climate change. Tea soil demands optimum conditions to support tea cultivation. Changes in these conditions may need to be updated for the development of future tea plantations. With this intent, the study was designed to understand the consequences of increased CO_2 and temperature on the tea soil rhizosphere carbon, nitrogen and their incorporation into the microbial biomass. This study addresses the research gap through an evidence based experimental work that monitors the impact of temperature and CO_2 on tea rhizosphere. From the study, it can be concluded that elevated temperature and CO_2 alter the rate of change in soil organic carbon pool of tea rhizosphere soil along with the microbial biomass carbon, whereas no such alteration was found for soil total nitrogen and microbial biomass nitrogen. Further this work will also help in informing future researchers by identifying new research questions and also to study more such tea cultivars with new ideas and purposes, providing a picture in developing a better understanding of the invisible impacts of increasing temperature and CO_2 in the rhizosphere of tea soil, which is an important aspect for sustainable tea cultivation.

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