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Effect of Nanoclay polymer loaded urea and nitrification inhibitor on root parameters in rice and wheat under elevated CO_2 and temperature condition

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Abstract
An experiment was conducted to study the effect of NCPCs, urea and nitrification inhibitor on root parameters in rice and wheat crops under elevated CO_2 and temperature condition. In general the elevation in atmospheric CO_2 concentration caused significant increase in root parameters. In wheat crop, under both elevated CO_2 and temperature conditions the root length was found to be highest under (NCPC+ Efficient NI-1) treatment with 3013 and 2603 cm, respectively. Root length of wheat crop under treatment applied NCPC with nitrification Schiff base -1 and NCPC with nitrification Schiff base-2 were at par with each other in both ambient and elevated condition. The studies showed that at elevated CO_2 concentration in the rhizosphere environment is supposed to be modified due to higher C- fluxes through exudates, mucilages, mucigel etc. Similar results are observed for root surface area. The maximum root volume was found under treatment receiving NCPC with nitrification (Schiff base 1) under ambient (5.12 cm^3) and elevated CO_2 (5.45 cm^3) and temperature (4.52 cm^3) conditions in rice crop. In all, the root growth parameters increased when NCPCs applied along with nitrification inhibitor (Schiff base-2).

Keywords: Root parameter, NCPCs, nitrification inhibitors, elevated CO_2

Introduction
There are various ways to improve the nutrient efficiency considerably by leaf application, by avoiding leaching, immobilization, etc., which occurs in the soil. The most commonly applied plant nutrient i.e. nitrogen (N), has been considered to be the major factor in limiting the yield (Bockman and Olfs 1998) [2]. The urea is most widely used as a source of N but its utilization efficiency is about 30-40 percent due to surface runoff, leaching, and volatilization losses (Ladha *et al*., 2005) [4]. One of the key approach for lowering the nutrient losses is by use of controlled nutrient release which reduces leaching after application that ultimately minimize the economic costs as well as environmental problems (Noppakundilograt *et al*., 2015) [5]. The combination of nitrification inhibitors and superabsorbent clay-polymers may enhance plant nutrition and also at the same time mitigate the impact on environment caused by water-soluble mineral fertilizers (Li *et al*., 2005; Zhang *et al*., 2005) [5, 10]. Also it may happen that at elevated CO_2 more nanoparticles enter into the root system and cause undesirable effects. Similarly, the effect of nanoparticles on soil microbes at elevated CO_2 is not known. Keeping in view the above knowledge gaps this investigation was undertaken to study the effect of NCPC, urea and nitrification inhibitor on root parameters in rice and wheat crops under elevated CO_2 and temperature condition.

Materials and method
For the preparation of NCPCs, acrylamide (Am) and acrylic acid (AA) for polymer, crosslinker as N,N-methylene bis-acryamide and free radical initiator as ammonium persulphate (APS) procured from SRL Pvt. Limited, Mumbai, India were used. The nitrification inhibitors loading with urea (1 and 3 wt % of N, respectively) were done through adding of pre weighed dry gel with compounds solution for 20 h to reach swelling equilibrium. The swollen gels were dried at 60ºC for 6 days, milledand stored for further analysis and use. In National Phytotron Facility, ICAR-Indian Agriculture Research Institute, New Delhi, anexperiment was conducted on Rice (*Oryza sativa*) (PUSA Basmati-1509) and wheat (HD-2932).

These crops were taken at two levels of atmospheric carbon dioxide (390 ± 10 ppm & 610 ± 10 ppm). In the phytotron growth chamber variation in CO_2 attentiveness was ± ten ppm. Recommended dose of fertilizer was applied (90:60, P_2O_5:K_2O kg ha^{-1}, respectively) by solution. Three doses of N were selected (i.e., 90%, 75% and 60% of recommended N dose) which were applied in three split doses, 50% as basal, 25% at 30 DAS and left over 25% on 60 DAS. The treatments taken in this experiment included: two CO_2 level: (390 ± 10 mmol mol^{-1} and 600 ± 10 mmol mol^{-1}); two temperature level: 2 (Ambient and Elevated which is 2.5°C more than that of ambient throughout the life cycle of crop). The treatment combinations are given below-: T_1-Control, T_2-NCPC, T_3-Urea (50%), T_4-Urea (100%), T_5-Neem Coated Urea (NCU), T_6- NCPC + Urea (50%), T_7- Urea + DCD (100%), T_8- NCPC + dicyandiamide (DCD) (50%), T_9- NCPC + (Schiff base-1, T_{10}- NCPC + Schiff base-2).

Root samples were collected from harvesting stage of rice and wheat crops. Roots were washed twice with double distilled water to avoid root detritus. After staining the roots with methyl violet solution, root area, the root length, average root density and root volume were determined with help of a root length scanner (Aggarwal *et al.*, 2009) [1]. The ratio of the total root length to the soil core volume was used to calculation of the root length density (cm cm^{-3})

Result and Discussion

Effect of NCPC, urea and nitrification inhibitor on root length (cm)

The root length increased significantly over the control when NCPC was applied with urea and DCD. The maximum root length (2800 cm) was observed under treatment NCPC with nitrification inhibitor-1 (T9) in wheat under ambient condition (Table 1). Root length of wheat crop under treatment NCPC with nitrification inhibitor-1 and NCPC with nitrification inhibitor-2 were at par with each other in both ambient and elevated condition. Statistically non-significant difference in root length was observed among the treatments T2, T3, T5, T7, T8, T9 and T10 under ambient condition (Table 1). Root length of wheat crop was also highest under elevated CO_2 and temperature condition in T9 in wheat crop 3013 and 2603 cm respectively. The lowest root length was observed in control (T1) under investigated environmental conditions in both crops (Table 1). Root length of rice crop also increased significantly over the control with different NCPCs, urea and nitrification inhibitor treatments. The maximum root length under ambient (3002 cm), elevated CO_2 (3100 cm) and temperature (2712cm) conditions was observed under treatment applied NCPC with nitrification Schiff base-1(Table 3).

Effect of NCPC, urea and nitrification inhibitor on root volume (cm^3)

The root volume was observed significantly lower in control (T_1) as compare to all fertilized treatments under all investigated environmental conditions in both wheat and rice crop. The maximum root volume was observed under (T_9) NCPC with nitrification inhibitor-1 under ambient (4.86cm^3), elevated CO_2 (5.28 cm^3) and temperature (4.27 cm^3) conditions in wheat crop (Table 2). Root volume of wheat crop under treatment receiving NCPC with nitrification inhibitor-1 and NCPC with nitrification inhibitor-2 were at par with each other in both ambient and elevated condition. The statistically non-significant difference was found in root volume between treatments 50% urea and 100% urea under ambient and elevated conditions in wheat crop (Table 2). Root volume of rice crop also increased significantly over the control with different NCPC, urea and nitrification inhibitor treatments. The maximum root volume was found under treatment receiving NCPC with nitrification inhibitor-1 under ambient (5.12 cm^3) and elevated CO_2 (5.45 cm^3) and temperature (4.52 cm^3) conditions in rice crop (Table 2). The significant difference in root volume was observed between T_9 and T_{10} under ambient and elevated temperature condition whereas, non-significant difference was observed under elevated CO_2 in rice crop (Table 2).

Root surface area (cm^2) as influenced by different treatments

The root surface area was increased significantly over the control when NCPC applied with urea, DCD and nitrification inhibitor in both wheat and rice crop under all investigated environmental conditions. The maximum root surface area was observed in NCPC with nitrification inhibitor-1 treatment (T_9) under ambient (431 cm^2) and elevated CO_2 (463 cm^2) and temperature (390 cm^2) conditions in wheat crop (Table 3). There was no significant difference in root surface area of wheat between treatments 50% urea and 100% urea under ambient and elevated conditions. Almost similar trend was found in rice crop under investigated environmental conditions. Root surface area of rice crop also increased significantly over the control with different NCPC, urea and nitrification inhibitor treatments under ambient and elevated CO_2 and temperature. The maximum root surface area was found in T_9 under ambient conditions (508 cm^2), elevated CO_2 (529 cm^2) and temperature (533 cm^2) in rice crop (Table 3). Statistical significant difference in root surface area was observed between T_9 and T_{10} under both ambient and elevated conditions. There was no significant difference in root surface area of rice crop between treatments 50% urea and 100% urea in ambient and elevated conditions (Table 3). Elevated CO_2 concentration led to substantial increase in the root length. It was also observed that the rate of plants growth with moderately insufficient nutrient increased by higher CO_2 in atmosphere, however under sufficient nutrient supply does not show the same (Ziska *et al*, 2003) [11]. The root surface area, root length and root volume in NCPC + urea + Schiff base-1 were significantly lower at ambient and elevated temperature conditions related to elevated CO_2. Yield-boosting effect of elevated CO_2 often diminishes when not supported by sufficient nutrient supply in soil (Campbell and Sage 2006). This implies that the requirement of plant nutrients, which are not increasing in soil in accordance with the rising atmosphere CO_2, will also increase the carbon fertilisation effect (Sinclair 1992; Manoj-Kumar *et al.*, 2011) [9, 6]. Rising temperature which affects the plant growth and also have impact on availability as well as uptake of nutrients by plants. Climate change has potential to influence the supply and demand of nutrient dynamics by interactive effect with increasing CO_2 (Manoj-Kumar *et al*. 2012) [7].

Table 1: Root length (cm) of rice and wheat as influenced by different treatments

Treatment	Wheat			Rice		
	Ambient	Elevated		Ambient	Elevated	
		Temp.	CO_2		Temp.	CO_2
T1	1309^{D}	1208^{D}	1387^{C}	1341^{D}	1301^{D}	1456^{D}
T2	2517^{AB}	2404^{AB}	2593^{AB}	2750^{AB}	2517^{AB}	2813^{AB}
T3	2277^{ABC}	2052^{ABC}	2366^{BCD}	2150^{CD}	1966^{CD}	2233^{CD}
T4	2336^{B}	2116^{ABC}	2492^{ABCD}	2256^{C}	2040^{C}	2353^{C}
T5	2500^{AB}	2260^{AB}	2665^{ABC}	2730^{B}	2443^{B}	2823^{AB}
T6	2396^{C}	2201^{ABC}	2557^{ABCD}	2613^{B}	2350^{B}	2716^{B}
T7	2600^{AB}	2414^{AB}	2783^{AB}	2807^{AB}	2527^{AB}	2900^{AB}
T8	2474^{ABC}	2470^{AB}	2699^{ABC}	2865^{AB}	2581^{AB}	2960^{AB}
T9	2800^{A}	2603^{A}	3013^{A}	3002^{A}	2712^{A}	3100^{A}
T10	2752^{A}	2566^{A}	2958^{AB}	2716^{B}	2444^{B}	2818^{AB}

Table 2: Root volume (cm^3) of rice and wheat as influenced by different treatments

Treatment	Wheat			Rice		
	Ambient	Elevated		Ambient	Elevated	
		Temp.	CO_2		Temp.	CO_2
T1	1.96^{E}	1.68^{D}	2.30^{D}	1.85^{F}	1.61^{DE}	2.18^{C}
T2	4.05^{ABC}	3.48^{ABCD}	4.63^{ABC}	4.64^{B}	4.07^{A}	5.00^{AB}
T3	3.09^{DE}	2.91^{BCD}	3.36^{D}	3.13^{E}	2.82^{D}	3.46^{BC}
T4	3.46^{CDE}	3.05^{ABCD}	3.73^{CD}	3.77^{D}	3.40^{C}	4.18^{ABC}
T5	3.77^{BCD}	3.40^{ABCD}	4.04^{BCD}	4.36^{C}	3.93^{B}	5.05^{AB}
T6	4.43^{AB}	3.86^{ABC}	4.80^{AB}	4.17^{C}	3.75^{B}	4.53^{ABC}
T7	4.15^{ABC}	3.68^{ABCD}	4.53^{ABC}	4.74^{B}	4.27^{A}	5.07^{AB}
T8	4.21^{ABC}	3.84^{ABC}	4.58^{ABC}	4.84^{B}	4.34^{A}	5.21^{A}
T9	4.86^{A}	4.27^{A}	5.28^{A}	5.12^{A}	4.52^{A}	5.45^{A}
T10	4.42^{AB}	3.99^{AB}	4.88^{AB}	4.25^{C}	3.77^{B}	4.95^{AB}

Table 3: Root surface area (cm^2) of rice and wheat as influenced by different treatments

Treatment	Wheat			Rice		
	Ambient	Elevated		Ambient	Elevated	
		Temp.	CO_2		Temp.	CO_2
T1	153^{D}	126^{E}	176^{D}	179^{E}	120^{G}	212.3^{F}
T2	344^{BC}	310^{BC}	379^{BC}	452^{AB}	430^{C}	482^{BC}
T3	244^{C}	226^{D}	266^{C}	302^{D}	373^{EFG}	322^{EF}
T4	299^{BC}	270^{CD}	295^{BC}	323^{D}	393^{DEF}	339^{EF}
T5	320^{ABC}	280^{CD}	344^{ABC}	441^{B}	433^{CD}	454^{BCD}
T6	308^{BC}	275^{CD}	335^{ABC}	367^{C}	423^{CDE}	387^{DE}
T7	364^{ABC}	330^{ABC}	399^{ABC}	482^{A}	470^{BC}	502^{ABC}
T8	338^{ABC}	301^{BCD}	366^{ABC}	502^{A}	496^{AB}	521^{AB}
T9	431^{A}	390^{A}	463^{A}	508^{A}	533^{A}	529^{A}
T10	377^{AB}	370^{AB}	418^{AB}	411^{B}	437^{CD}	431^{CD}

Conclusion

The maximum root length (2800 cm) was observed under treatment applied NCPC with nitrification inhibitor-1 (T9) in wheat under ambient condition. The maximum root length under ambient (3002 cm), elevated CO_2 (3100 cm) and temperature (2712cm) conditions was observed under treatment applied NCPC with nitrification inhibitor-1. The Statistically non-significant difference was found in root volume between treatments 50% urea and 100% urea under ambient and elevated conditions in wheat crop. The maximum root surface area was observed in NCPC with nitrification inhibitor-1 treatment (T_9) under ambient (431 cm^2) and elevated CO_2 (463 cm^2) and temperature (390 cm^2) conditions in wheat crop.

Elevation in atmospheric CO_2 concentration caused significant increase in root growth. The root growth was highest under treatment T_9at ambient and elevated CO_2.Elevated CO_2 increases carbon supply below ground by secretion of organic acids, whereas warming is likely to increase respiration and decomposition rates, leading to speculation that these effects will alter nutrient supply and many other soil processes. Being a C_3 crop, higher grain yields of wheat exposed to elevated CO_2 due to reduction in photorespiratory losses of photosynthates and net increase in carbon assimilation rate.

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