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Growth and biomass partitioning in mungbean with elevated carbon dioxide, phosphorus levels and cyanobacteria inoculation

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ABSTRACT

Mungbean is an important leguminous crop providing protein for the rural and urban poor in South and Southeast Asia. An experiment was conducted in free air carbon dioxide enrichment facility (FACE) ring to study the impact of increased CO_2 level on growth and biomass partitioning in mungbean crop. The crop was grown under ambient (400 µmol mol^{-1}) and elevated CO_2 concentration (550 µmol mol^{-1}) with 5 doses of P with and without cyanobacterial inoculation. Elevated CO_2 significantly increased biomass accumulation in mungbean crop which was further increased by P and cyanobacteria application. Leaf biomass increased by 34.4% at increased CO_2 level. Maximum biomass allocation to seeds was observed with P dose of 16 mg kg^{-1} soil in both ambient and elevated CO_2 conditions. Allocation was more in high CO_2 treatment. The study concludes that mungbean crop grown under elevated CO_2 condition accumulates more biomass which gets further improved by application of P nutrient and cyanobacteria inoculation.

Key wards: Mungbean, elevated CO_2, phosphorus, cyanobacteria, biomass partitioning.

Mungbean [*Vignaradiata* (L.)Wilczek] is an important legume providing protein for the rural and urban poor in South and Southeast Asia. Due to its rapid growth and early maturity it is adopted in multiple cropping systems in the drier and warmer climates of tropics and sub-tropics. Mungbean seed has a high nutritive value containing about 22.5% protein, 1.3 % fat and 60.4 % of carbohydrate (Hussain *et al.,* 2011). The legume crops like mungbean may be affected by the changing climate, caused due to increasing concentration of atmospheric carbon dioxide (CO_2) and other greenhouse gases (GHGs). Since 1750, CO_2 concentration has increased from 278 ppm (Pearson and Palmer, 2000) to currently 400 ppm (IPCC, 2014).

Due to their ability to fix atmospheric nitrogen, the legumes will get an added advantage over non-leguminous plants under elevated CO_2 condition (Zanetti *et al.,* 1996; Ross *et al.,* 2004). Growth response of different crop species to varying CO_2 concentration is different (Hunt *et al.,* 1991; Poorter *et al.,* 1996). There are reports of increased root length, root mass, and root diameter in soybean crop grown at increased CO_2 level. According to Jin *et al.,* (2012), elevated CO_2 increased the biomass and length of root in legume on an average by 16% and 14% respectively. Impact of atmospheric CO_2 enrichment on increase in peak leaf area index was also reported in leguminous crops like pigeon pea (Saha *et al.,* 2012).

In order to get all the benefits of enriched CO_2, adequate supply of other essential nutrients is required. Since phosphorus is an essential nutrient influencing N fixation in legumes hence P application may have significant effect on legume crops in changing climatic conditions. Cyanobacteria also known as blue green algae (BGA) have a unique potential to contribute to the productivity of different agro-ecosystem under different ecological situations. Cyanobacteria are widely known to exist in rice ecosystem. But their beneficial effects were also observed on other crops. In wheat, application of cyanobacterial strains resulted in enhanced plant growth and yields (Karthikeyan *et al.,* 2007; Nain *et al.,* 2010). The objectives of the present study were to study the partitioning of biomass in mungbean crop under elevated CO_2 condition with different P levels and cyanobacteria inoculation.

MATERIALS AND METHODS

An experiment was conducted during the kharif season (July to October) of 2014 in Indian Agricultural

Research Institute farm, New Delhi, India (28°35'N and 77°12'E). Mungbean crop (variety Pusa Vishal) was grown in pots both inside and outside the Free Air Carbon dioxide Enrichment (FACE) facility. The FACE ring is made up of eight horizontal pipes which releases CO_2 enriched air at the crop canopy level. Diameter of the ring is 8 m Carbon dioxide (CO_2) concentration inside the ring was measured by non-dispersive infrared gas analyzer (IRGA) and data was logged automatically in the computer at every 5 minutes interval. Carbon dioxide (CO_2) concentration inside the FACE ring was maintained at around 550 ± 20 µmol.mol^{-1} while crops grown outside the FACE ring was subjected to ambient CO_2 concentration i.e. 400 µmol mol^{-1}. Recommended doses of nitrogen and potassium were applied through urea and murate of potash (MOP) as basal dose. In the study there were five different levels of phosphorus (P) like control P (No phosphatic fertilizer applied), 75 % of recommended dose of P, 100 % of recommended dose of P, 125% of recommended dose of P and 150% of recommended dose of P. P was applied through single super phosphate (SSP) fertilizer. Mungbean seeds were inoculated with crop specific *Rhizobium* inoculants available in the Division of Microbiology and cyanobacterium - Calothrix sp. inoculation was done in soil.

Plants are collected by destructive sampling at 25, 45 and 65 days after showing (DAS)and above ground biomass were recorded in those stages. Leaf, stem, root and seeds were collected during harvesting stage of the crop (65 DAS) and their weight was recorded. Nodules were extracted from roots at 25 and 45 DAS and their biomass was recorded.

The design of the experiment was factorial completely randomized design (CRD) with 20 treatments. Statistical analysis of the data was performed using SAS (ver. 9.3) statistical package developed by SAS Institute Inc.

RESULTS AND DISCUSSION

Biomass Partitioning

Leaf biomass : Elevated CO_2 level, cyanobacterial inoculation as well as P application had significant effect on the leaf biomass of the crop. Elevated CO_2 increased leaf biomass by 34.4% at 65 DAS. Cyanobacterial inoculation also increased leaf biomass of the crop. Leaf biomass was 4.36 g plant^{-1} in without cyanobactria treatment while in treatment with cyanobacterial inoculation leaf biomass was 4.83 g plant^{-1}. Application of P significantly increased leaf biomass over control. The interactive effect of CO_2 and phosphorus significantly increased leaf biomass of the crop throughout the crop growth period.

Similar results were reported by Sharma and Sengupta (1990) who reported that the extra carbon fixed by mungbean crop in elevated CO_2 condition gets translocated to the growing organs resulting in higher biomass of the crop.

Stem biomass : Stem weight of mungbean crop ranged from 2.90 to 7.74 g plant^{-1} in different treatments (Fig. 1). Elevated CO_2 level also increased stem biomass of the crop. Stem weight of mungbean crop was 6.50 g plant^{-1} under high CO_2 level while in ambient CO_2 treatment stem biomass was 5.12 g plant^{-1}. Cyanobacterial inoculation also increased stem biomass of the crop by 10.5%. Application of P significantly increased stem weight of the crop over control. Highest stem biomass was recorded in treatment with 20 mg kg^{-1} P. Stem weight was 6.78 g plant^{-1} with P dose of 20 mg kg^{-1} soil.

Root biomass : Increased CO_2 level had significant improved root growth of the plant. Root weight in ambient CO_2 condition was 0.41 g plant^{-1} while in elevated CO_2 treatment root weight was 0.61 g plant^{-1}. Biomass allocation to mungbean roots also increased under high CO_2 condition. At harvesting stage of the crop 3.4% of total biomass was allocated to roots under elevated CO_2 condition while in ambient treatment 3% of the total biomass was partitioned to roots in mungbean crop. Several studies have shown increase in root weight under elevated CO_2 condition in several crops including wheat, black gram (Vanaja et al., 2007), and soybean (Del Castillo et al., 1989). Cyanobacterial inoculation significantly increased root weight of the crop. A study by Liu-Shi Ming and Liang-Shi Zhong (1998) revealed that cyanobacterial extracts increased root growth and number of roots in mungbean crop.

Increase in P dose significantly increased root growth of the crop which was evident from significantly higher root biomass at higher P levels throughout the crop growth period. Maximum root weight (0.60 g plant^{-1}) of the crop was observed with P dose of 20 mg kg^{-1} soil.

Above ground biomass : Increased CO_2 level has resulted in significant rise in above ground biomass of the crop under high CO_2 condition as compared to ambient one. Cyanobacterial inoculation also showed high above ground biomass in all the crop growth stages. At harvesting stage of the crop i.e. at 65 DAS, above ground biomass was 17.13 g plant^{-1} in high CO_2 treatments while in ambient condition above ground biomass was 13.05 g plant^{-1} (Table 1). Increased CO_2 concentration enhanced above ground biomass by

Table 1: Impact of elevated CO_2, cyanobacteral inoculation and phosphorus doses on above ground biomass plant^{-1}(g plant^{-1}) at 25, 45 and 65 DAS in mungbean.

P Levels (mg kg^{-1} soil)	Phosphorus	Ambient CO_2	Mean elevated CO_2	Without Cyanobacteria	With Cyanobacteria
			25 DAS		
Control P	1.48 ^{E}	1.32 ^{I}	1.65 ^{G}	1.38 ^{H}	1.59 ^{G}
8	1.71 ^{D}	1.51 ^{H}	1.91 ^{F}	1.60 ^{G}	1.81 ^{F}
12	2.18 ^{C}	1.90 ^{F}	2.46 ^{C}	2.15 ^{E}	2.22 ^{D}
16	2.39 ^{B}	2.03 ^{E}	2.75 ^{B}	2.32 ^{C}	2.46 ^{B}
20	2.68 ^{A}	2.21 ^{D}	3.14 ^{A}	2.51 ^{B}	2.85 ^{A}
Mean	1.79^{B}	2.38 ^{A}	1.99 ^{B}	2.19 ^{A}	
			45 DAS		
Control P	6.12 ^{E}	5.32 ^{G}	6.92 ^{FG}	5.69 ^{A}	6.55 ^{A}
8	9.24 ^{D}	8.11 ^{F}	10.38 ^{E}	8.34 ^{A}	10.15 ^{A}
12	12.46 ^{C}	10.88 ^{E}	14.05 ^{BC}	11.49 ^{A}	13.43 ^{A}
16	13.56 ^{B}	11.98 ^{DE}	15.14 ^{B}	12.57 ^{A}	14.55 ^{A}
20	15.24 ^{A}	13.18 ^{DC}	17.30 ^{A}	14.49 ^{A}	15.98 ^{A}
Mean	9.89 ^{B}	12.76 ^{A}	10.52 ^{B}	12.13 ^{A}	
			65 DAS		
Control P	9.78 ^{D}	7.98 ^{A}	11.59 ^{A}	8.72 ^{A}	10.85 ^{A}
8	14.94 ^{C}	12.80 ^{A}	17.07 ^{A}	14.03 ^{A}	15.84 ^{A}
12	16.03 ^{B}	13.90 ^{A}	18.17 ^{A}	15.24 ^{A}	16.82 ^{A}
16	16.76 ^{B}	14.76 ^{A}	18.75 ^{A}	15.97 ^{A}	17.55 ^{A}
20	17.95 ^{A}	15.82 ^{A}	20.08 ^{A}	17.11 ^{A}	18.78 ^{A}
Mean	13.05 ^{B}	17.13 ^{A}	14.21 ^{B}	15.97 ^{A}	

Same letters are not significantly different.

Table 2: Impact of elevated CO_2, cyanobacteral inoculation and phosphorus doses on fresh weight of nodule plant^{-1} (mg plant^{-1}) at 25, 45 and 65 DAS in mungbean.

P Levels (mg kg^{-1} soil)	Phosphorus	Ambient CO_2	Mean elevated CO_2	Without Cyanobacteria	With Cyanobacteria
			25 DAS		
Control P	180.3^{E}	125.3^{G}	235.3^{DE}	161.2 ^{A}	199.4 ^{A}
8	230.5^{D}	159.9^{FG}	301.1^{C}	211.7 ^{A}	249.3 ^{A}
12	290.1^{C}	198.6^{FE}	381.7^{B}	274.0 ^{A}	306.2 ^{A}
16	321.2^{B}	228.3^{DE}	414.0^{B}	298.7 ^{A}	343.6 ^{A}
20	371.2 ^{A}	265.4^{DC}	477.0 ^{A}	353.5 ^{A}	388.9 ^{A}
Mean	195.5^{B}	361.8 ^{A}	259.8^{B}	297.5 ^{A}	
			45 DAS		
Control P	339.5^{D}	250.8^{G}	428.2^{DFE}	311.6 ^{A}	367.4 ^{A}
8	449.3^{C}	338.4^{GF}	560.1^{DC}	428.5 ^{A}	470.0 ^{A}
12	541.1^{B}	412.1^{FE}	670.1^{BC}	551.3 ^{A}	530.9 ^{A}
16	644.4 ^{A}	499.6^{DE}	789.2^{BA}	587.7 ^{A}	701.1 ^{A}
20	718.9 ^{A}	543.3^{DCE}	894.5 ^{A}	694.7 ^{A}	743.1 ^{A}
Mean	408.8^{B}	668.4 ^{A}	514.8^{B}	562.5 ^{A}	

Same letters are not significantly different.

Table 3: Impact of elevated CO_2, cyanobacteral inoculation and phosphorus doses on seed yield (g plant^{-1}) of mungbean crop.

P Levels (mg kg^{-1} soil)	Phosphorus	Ambient CO_2	Mean elevated CO_2	Without Cyanobacteria	With Cyanobacteria
Control P	2.7^E	2.1^A	3.3^A	2.4^A	3.0^A
8	4.6^D	4.0^A	5.3^A	4.3^A	5.0^A
12	5.0^C	4.4^A	5.7^A	4.7^A	5.4^A
16	5.4^B	4.7^A	6.1^A	5.0^A	5.7^A
20	5.7^A	5.0^A	6.4^A	5.3^A	6.0^A
Mean		4.0^B	5.4^A	4.3^B	5.0^A

Same letters are not significantly different.

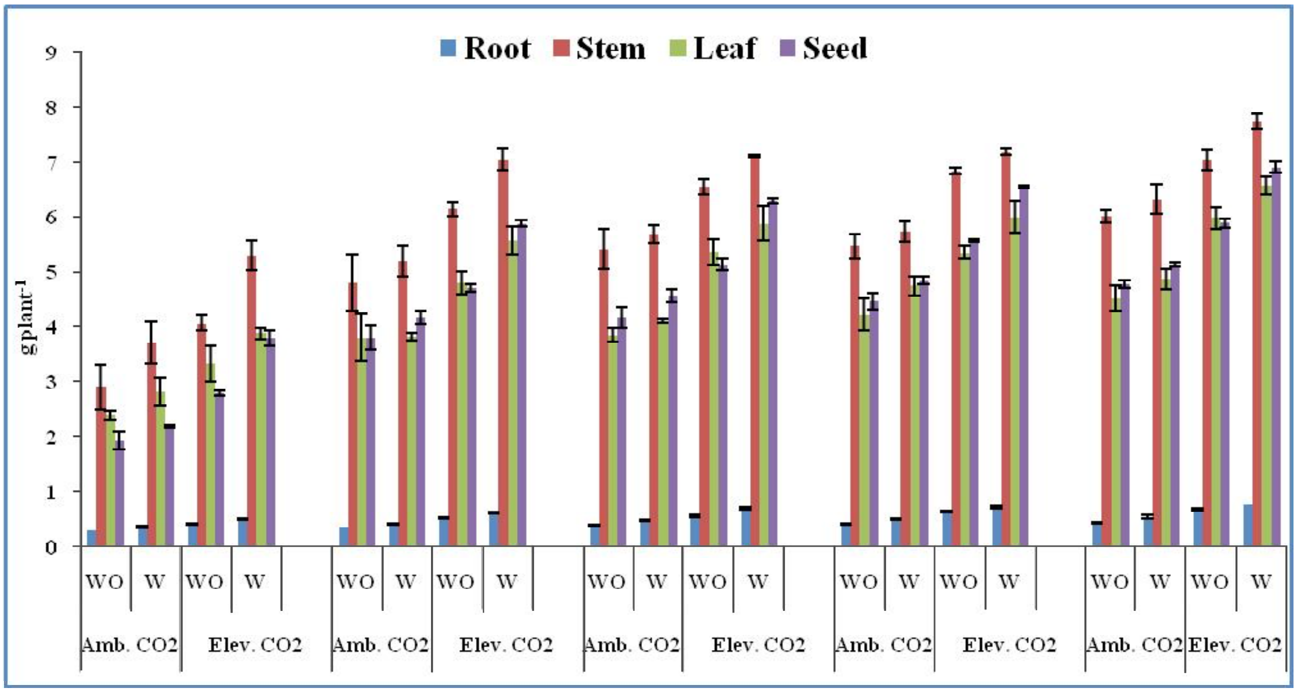

Fig 1: Interactive effect of elevated carbon dioxide, cyanobacterial inoculation and phosphorus doses on biomass of root, stem, leaf and seed per plant in Mungbean at 65 DAS (WO-Without Cyanobacteria, W-With Cyanobacteria).

33%, 29% and 31.3% than ambient CO_2 level at 25, 45 and 65 DAS respectively. P application also significantly affected aboveground biomass of the crop. At 25, 45 and 65 DAS highest above ground biomass (2.68, 15.24 and 17.95 g plant^{-1}) was observed with P dose of 20 mg kg^{-1} soil (Table 1). The interactive effect of CO_2 and phosphorus levels on above ground biomass was observed at 25 and 45 DAS.

Increased crop biomass at elevated CO_2 level was primarily attributed to the fact that more amounts of carbon assimilated by the crop under high CO_2 concentration was partitioned towards the growing organs leading to more leaf, stem, root, seed and ultimately higher above ground biomass of the crop. According to Saha *et al.,*(2012) increased partitioning of photosynthate led to higher biomass and seed yield in pigeonpea crop under elevated CO_2 condition. Partitioning of accumulated above ground biomass to seeds was higher (31.5%) in elevated CO_2 condition than ambient treatment (30.7%).

Nodule biomass : Elevated CO_2 as well as cyanobacterial inoculation significantly increased nodule weight of plants throughout the crop growth period. Nodule growth was maximum at 45 DAS (Table 2). At this stage nodule weight was 668.4 mg plant^{-1} under increased CO_2 concentration while in ambient condition nodule weight was 408.8 mg plant^{-1} (Table 2). Earlier workers also reported that nodule biomass in legumes increased by 46 % in response to enriched CO_2 (Jin *et al.,* 2012). In cyanobacteria inoculated treatments nodule biomass was 562.5 mg plant^{-1} while in without cyanobacteria treatment nodule weight was 514.8 mg plant^{-1} (Table 2). Increasing doses of P also significantly increased nodule growth in mungbean plants (Table 2). At 25 DAS nodule weight increased significantly with increased P dose with maximum nodule weight (371.2 mg $plant^{-1}$) recorded at P dose of 20 mg kg^{-1} soil (Table 2). But at 45 DAS, application of P upto 16 mg kg^{-1} soil significantly increased nodule biomass of the crop. The interactive effect of elevated CO_2 along with high doses of P further improved the nodule growth in mungbean crop. According to Israel (1987), phosphorus has specific roles in nodule initiation, growth, and functioning in soybean crop.

Yield

Elevated CO_2 significantly increased seed yield of mungbeancrop.Seed yield ranged from 1.9 to 6.9 g $plant^{-1}$ in different treatments (Fig. 1). Vanaja *et al* (2010) also found that increased level of CO_2 increased both biomass and seed yield of pigeonpea crop as compared to ambient condition. Application of cyanobacterial inoculation also increased seed yield with 5 g $plant^{-1}$ seed yield in cyanobacteria applied treatments and 4.3 g $plant^{-1}$ seed yield in without cyanobacteria treatment (Table 3). Increase in P dose improved seed yield in both ambient and elevated CO_2 treatments. Maximum seed yield (5.4 g $plant^{-1}$) was observed with P dose of 20 mg kg^{-1} soil (Table 3). Partitioning of accumulated biomass to seeds also improved with P application. Maximum biomass allocation to mungbean seeds was observed with P dose of 16 mg kg^{-1} soil in both ambient and elevated CO_2 condition. But the allocation was more in high CO_2 treatment. High CO_2 concentration along with cyanobacterial inoculation further improved seed yield of mungbean crop. Jagannath *et al.* (2002) also showed that use of BGA in chickpea crop has resulted in higher biomass yield and seed yield of the crop.

CONCLUSIONS

Elevated CO_2 condition significantly increased biomass accumulation in mungbean crop. Higher amount of carbon assimilated by the crop under high CO_2 concentration has increased partitioning of more assimilates to the growing organs like leaf, stem, root and seed. Cyanobacterial inoculation as well as P application also improved growth of leaf, stem, roots and ultimately seed yield of the crop. Partitioning of accumulated biomass to seeds was also found to be more in elevated CO_2 treatment than ambient treatment. The study concludes that mungbean crop grown under elevated CO_2 condition accumulates more biomass which gets further improved by application of P nutrient and cyanobacteria inoculation. Partitioning of accumulated assimilates to seeds was also higher in high CO_2 concentration. The P application caused more biomass translocation to mungbean seeds under elevated CO_2 condition.

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