



Investigation Effects of Carbon Dioxide (CO₂) Concentrations Produced from a CO₂ Enrichment Design Process on the Growth and Physiological Properties of Corn and Soybean

Hananeh Rabiee¹, Hooman Rowshanaie², Omid Rowshanaie^{2*}, Hadis Safari Nezhad Fard¹, Mehdi Rabiee³

¹ B.Sc. Student, Biotechnology, Fatemiyeh Institute of Higher Education, Shiraz, Fars Province, Iran

² Assistant Professor, Fatemiyeh Institute of Higher Education, Shiraz, Fars Province, Iran

³ Shiraz University of Medical Sciences, Shiraz, Fars Province, Iran

Received: 12/06/2024

Accepted: 22/08/2024

Published: 20/09/2024

Abstract

Climate change is one of the most debatable reasons for changing plant performance. Meanwhile, a higher amount of carbon dioxide (CO₂) directly or indirectly affects the growth and development of corn and soybean oil plants; CO₂ concentration changes also affect the dry weight characteristics of the plant. In this experiment, three effective levels of CO₂ concentration have been evaluated on corn and soybean plants. Increasing the effectiveness of CO₂ concentration from 400 PPM to 800 PPM depicted a significant increase in the plant's physiological traits, a 54% and 8.5% increase in Soybean biomass and Corn biomass, respectively. Furthermore, based on leaf area, the results reveal a 4% and 5% increase in Soybean and Corn, respectively. In contrast, increasing the CO₂ concentration to 1000 PPM resulted in a decrease in plant performance and physiological traits, both directly and indirectly. With increasing CO₂ concentration, a direct increase in physiological traits is observed.

Keywords: CO₂, plant physiological traits, corn, soybean

1 Introduction

Climate is one of the most important reasons for annual changes in the yield and production of agricultural plants. In high-performance environments and modern technology, the source and pattern of solar radiation, temperature, and rainfall are the main determinants of plant growth through several direct and indirect mechanisms. Other factors, such as wind speed and the occurrence of storms, also have a significant effect on the growth of plants [1,2]. One of the effects of climate change is the increase in the concentration of greenhouse gases [3]. Changes in the concentration of the main gases that absorb atmospheric radiation by humans have created radiation pressure on the climate system. These gases are primarily released into the air from domestic activities and, to a lesser extent, from agricultural activities and land use change [4,5].

Higher amounts of carbon dioxide (CO₂) directly affect the physiology of water relations, growth, and development of plants. Because the temperature will increase, and as a result, the supply and consumption of water for the plant will change. Such climate changes can lead to changes in the type and composition of vegetation and the amount of production in natural and agricultural systems. Drought is the most important environmental factor limiting the growth and production of plants worldwide. One of the environmental aspects that has attracted the attention of many researchers, as mentioned, is the

issue of climate change due to the increase in the rate of CO₂ [6,7]. Significant increases in yield were a general worldwide phenomenon for oily plants, occurring in many crops and countries. Soybean has crucial role as vegetable oil, protein for animal and human consumption, and industrial uses. Corn also is a pivotal worldwide crop utilized as animal feed and consumer products [8].

Different ambient stresses can affect the physiological and morphological traits of plants. Rowshanaie et al. [9] revealed that ambient stresses have an opposite linear relationship with these traits. Among ambient stresses, CO₂ dramatically influences plants' growth and development. To investigate the change of CO₂ increase in the physiological factors of plants, especially oil plants, Miri and Rastegar [10] showed the effect of CO₂ increase from 350 PPM to 700 PPM on soybean. The results showed an increase in chlorophyll index by 6.6% and 7.8%. Also, the effect of increasing carbon dioxide on root dry weight, leaf dry weight, and shoot dry weight showed a significant increase. In another research, Miri [11] investigated the effect of increasing CO₂ on corn as an oil seed plant and evaluated physiological traits such as total dry weight, shoot dry weight, leaf dry weight, root dry weight, and leaf surface. The results revealed that increasing the amount of carbon dioxide from 400 PPM to 800 PPM showed a direct increase in the physiological traits of corn under the conditions of salinity stress and sufficient water. Also, in another research, Ikhlas

***Corresponding Author:** Omid Rowshanaie, Assistant Professor, Fatemiyeh Institute of Higher Education, Shiraz, Fars Province, Iran, Email: omid.rowshanaie@gmail.com

Pour et al. [12] investigated the physiological traits of sesame plants under the influence of increasing CO₂ concentration, and their results revealed that with the increase in carbon dioxide concentration from 380 PPM to 700 PPM, the physiological traits of sesame increased significantly under salinity stress conditions. Also, Mir Seyyed Hosseini et al. [13], by examining the physiological characteristics of wheat plants due to CO₂ increase, the aerial part of the plant showed an acceptable increase in growth.

This research focuses on two oil plants (Soybean and Corn) based on the effects of increasing CO₂ concentration through a process designed to produce carbon dioxide on the physiological traits, consisting of dry weights, chlorophyll index (Chl Index), leaf area, and specific leaf area of these two plants was investigated, since mentioned traits have an integral role on propagation and adaptation of these plants indigenous ambient stresses.

2 Methodology

2.1 CO₂ Enrichment Design

CO₂ is the main effective parameter for photosynthesis, but the ambient CO₂ concentration is insufficient to yield C3 crops. Hence, the methods for CO₂ enrichment have an impact on crop photosynthesis and biomass improvement [14]. The design of a CO₂ enrichment process is shown in Figure 1. As illustrated in Figure 1, first, CO₂ in delivery state condition (ambient temperature and 45-65 bar) and liquid phase (vapor fraction=0) is increasing the pressure by a pump, then enters into the heat exchanger to change the phase of CO₂ into a gas (vapor fraction=1) and adjust the temperature to a suitable temperature that is not harmful to crops (below 37°C). Finally, using a CO₂ sensor (with ±30 PPM accuracy) enrichment for monitoring the

CO₂ production and a splitter, the evaporated CO₂ was divided into three concentrations: 400 PPM, 800 PPM, and 1000 PPM.

2.2 The Experiment Conducted on Corn and Soybean Plant

This experiment was conducted to investigate the increasing effect of three levels of CO₂ concentration, 400 PPM, 800 PPM, and 1000 PPM, on the growth rate and physiological traits of two oil plants, corn and soybean, under greenhouse conditions. The characteristics of the studied soil are also shown in Table 1.

In this experiment, according to Table 1, plants were cultivated in 15 kg pots with soil texture (top soil, washed sand, and vermiculite). under greenhouse conditions including 27 °C, 65-75% humidity, and 800 PAR light intensity with 1.5 month duration of CO₂ treatment that released from CO₂ enrichment process into greenhouse air. The seeds of corn and soybean crops were grown in pots with the appropriate density for the plant. During the growth period, in addition to proper watering of the plants based on FC measured for each plant, three CO₂ concentration factors of 400 PPM, 800 PPM, and 1000 PPM were investigated for each of the corn and soybean plants separately in the performance of physiological traits such as total dry weight, leaf dry weight, shoot dry weight, root dry weight, leaf area, leaf specific area, and chlorophyll. This experiment was done in factorial form in a completely randomized design with 20 treatments and 4 replications. The fertilizers used included ammonium phosphate in the amount of 0.3 gr in the pot before planting and 0.45 gr in the form of urea in two stages. Due to its ability to fix nitrogen, urea fertilizer of one-third of corn has been used for soybean plants.

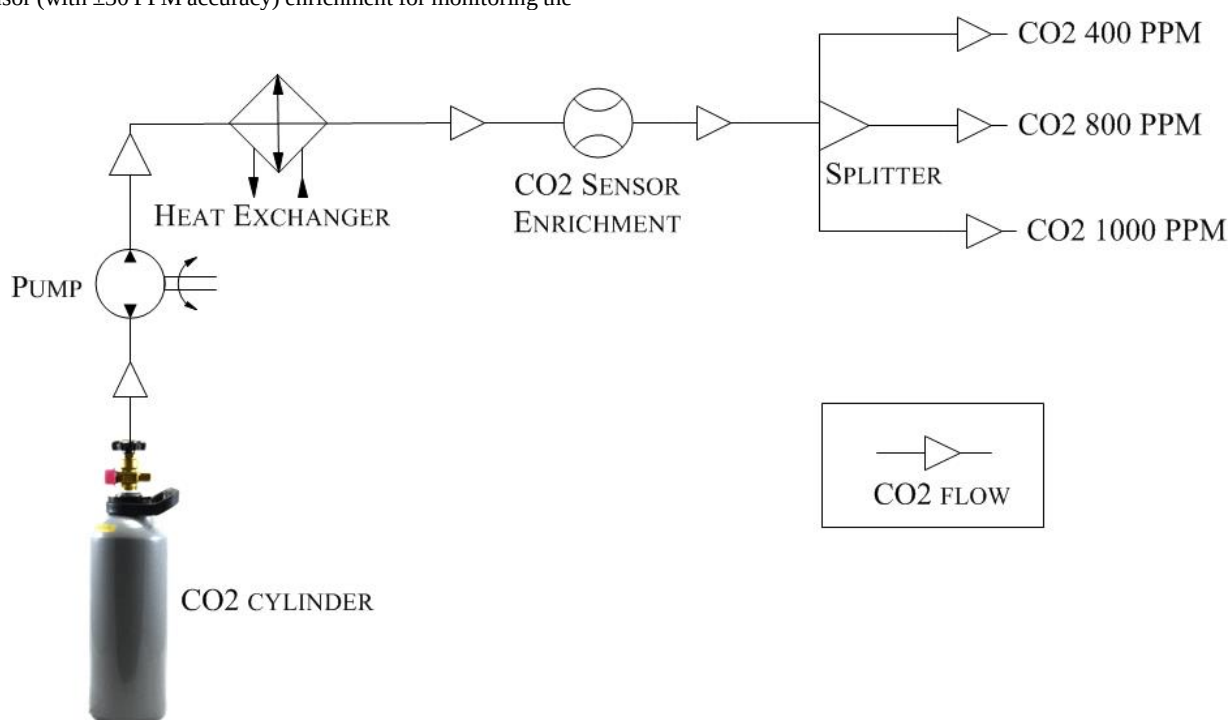


Figure 1. Designing CO₂ Enrichment Process

Table 1. Soil chemical and physical analysis

Sample	EC	PH	K	Na	Fe	Cu	Clay	Silt	Sand
	dS/m		meg/L		PPM			%	
Clay Soil	2.5	7.6	-	8.5	1.33	-	16.2	18.5	65.3
Sandy	0.86	7.4	-	1.8	0.77	-	2.7	1.1	96.2

3 Results and Discussion

3.1 Root Dry Weight

In soybean, the root dry weight was significantly changed. The effect of increasing CO₂ concentration on the dry weight of soybean and corn roots can be seen in Table 2. At a concentration of 400 PPM, the root dry weight of soybeans is 3.77. And at the concentration of 800 PPM, the root dry weight of soybean enhanced and reached 4.68, and at 1000 PPM, the root dry weight of soybean declined and reached 4.00 (Table 2). As an important reason, it can be said that the reaction of the roots to the increase of CO₂ depends on the soil moisture. In corn, the root dry weight has changed significantly, and at a concentration of 400 PPM, the root dry weight of corn is 29.40, and at a concentration of 800 PPM, the root dry weight of soybean has increased and reached 31.20, and at 1000 PPM, it was associated with a significant decrease and reached 30.60, because of reducing of photosynthesis rate (Table 2).

Table 2 The effect of CO₂ concentration on root dry weight of Soybean and Corn

Plant	Root Dry Weight (DWR) (gr)		
	400 PPM	800 PPM	1000 PPM
Soybean	3.77	4.68	4.00
Corn	29.40	31.20	30.60

3.2 Shoot Dry Weight

With the increase of CO₂ concentration from 400PPM to 800PPM, the shoot dry weight of soybean improved from 5.41 to 8.21, and a significant decline was observed in the shoot dry weight of soybean at 1000 PPM. At 400 PPM, the shoot dry weight of soybean was 46.00, and at 800 PPM, the shoot dry weight of corn increased and reached 52.00, and at 1000 PPM, it decreased and reached 49.00. The similar results revealed for Wheat and Sesame shoot dry weight [12,13]. Furthermore, with the increase in CO₂ concentration, significant changes were also observed between the shoot and the root.

Table 3. The effect of CO₂ concentration on shoot dry weight of Soybean and Corn

Plant	Shoot Dry Weight (DWS) (gr)		
	400 PPM	800 PPM	1000 PPM
Soybean	5.41	8.21	7.63
Corn	46.00	52.00	49.00

3.3 Leaf Dry Weight

The effects of increasing CO₂ concentration on the leaf dry weight of corn and soybean can be checked in Table 4. In soybean culture, with enhancing CO₂ concentration, total dry weight improved. At 400 PPM, the leaf dry weight of soybean was 6.36, and with the rise in CO₂ concentration to 800PPM, the leaf dry weight of soybean reached 11.02, and at 1000 PPM, the leaf dry weight of this plant decreased to 10.41. In the meantime, the vegetative growth that occurs in C₃ plant species is caused by the initial increase in carbon dioxide concentration. Regarding corn, with the increase in concentration from 400 PPM to 800 PPM, the leaf dry weight of corn increased from 60.40 to 64.20, and at 1000 PPM, the leaf dry weight of corn decreased to 63.60. An increase in CO₂ under stress conditions causes a significant increase in corn weight, and leaf weight is more sensitive to changes in CO₂ and stress than stem weight and plant weight. In severe ambient stresses, the stomatal conductance drops, and consequently, the leaf dry weight decreases.

Table 4. The effect of CO₂ concentration on leaf dry weight of Soybean and Corn

Plant	Leaf Dry Weight (DWL) (gr)		
	400 PPM	800 PPM	1000 PPM
Soybean	6.36	11.02	10.41
Corn	60.40	64.20	63.60

3.4 Biomass

The biomass of soybeans at the concentration of 400 PPM is equal to 15.54, and at the concentration of 800 PPM and 1000 PPM, it is 23.91 and 22.04, respectively. The biomass of corn enhanced from 400 PPM to 800 PPM, increased from 135.80 to 147.40, and reduced at 1000PPM (Table 5). Enhancing the amount of carbon dioxide concentration in both corn and soybean as oily plants cause the stomata to open and more exchange of photosynthetic gases, as a result of which the rate of photosynthesis increases and it shows a significant increase in the conversion of organic matter to minerals necessary for plant growth at 800 PPM CO₂ concentration. With the increase in CO₂ concentration, there are noticeable changes in the stress of two oil plants, corn and soybean.

Table 5. The effect of CO₂ concentration on biomass of Soybean and Corn

Plant	Biomass (gr)		
	400 PPM	800 PPM	1000 PPM
Soybean	15.54	23.91	22.04
Corn	135.80	147.40	143.20

3.5 Leaf Area (LA)

According to Table 6, the increase in CO₂ concentration had a significant effect on the Leaf area (LA) of soybean and corn. At a concentration of 400 PPM, the leaf area of soybean is 201, while at 800 PPM, the leaf area of soybean increases and reaches 209. At 1000 PPM, the leaf area of the soybean is reduced compared to 800 PPM and reaches 205. Between the concentration of 400PPM and 1000PPM, it is increasing. The leaf area of corn is 2115, 2220, and 2202, respectively, at the increasing rate of carbon dioxide concentration from 400 PPM to 800 PPM and finally at 1000 PPM. The leaf area of the corn is increased at 800 PPM and at 1000 PPM, the leaf area of the corn is increased at 800 PPM compared to 400 PPM.

Table 6. The effect of CO₂ concentration on leaf area of Soybean and Corn

Plant	Leaf Area (LA) (cm ²)		
	400 PPM	800 PPM	1000 PPM
Soybean	201	209	205
Corn	2115	2220	2202

3.6 Specific Leaf Area (SLA)

As shown in Table 7, at a concentration of 400 PPM, the specific leaf area (SLA) of soybean leaves is 31.60, and with a significant decrease at 800 PPM, it reaches 18.96. From 400 PPM to 1000 PPM, it decreases; from 800 PPM to 1000 PPM, the specific level of soybean leaves increases and increases from 18.96 to 19.69. In the SLA of the corn, the concentration of CO₂ is 400 PPM, the value is 35.02, and it decreased to 34.58 at 800 PPM and increased from 800 PPM to 1000 PPM; that is, it increased from 34.58 to 34.62.

3.7 Chlorophyll Index (Chl Index)

The chlorophyll index (Chl Index) is largely influenced by CO₂ concentration. The chlorophyll index increases with increasing CO₂ concentration in soybeans (Table 8). With the

increase of CO₂ concentration from 400 PPM to 800 PPM in soybeans, the chlorophyll index also increased and changed from 34.2 to 35.9, and CO₂ concentration decreased from 800 PPM to 1000 PPM in soybeans.

Table 7. The effect of CO₂ concentration on specific leaf area of Soybean and Corn

Plant	Specific Leaf Area (SLA)		
	400 PPM	800 PPM	1000 PPM
Soybean	31.60	18.96	19.69
Corn	35.02	34.58	34.62

In corn, with the increase of CO₂ concentration, the chlorophyll index is the same, and it has a significant increase from 400 PPM to 800 PPM, and at 1000 PPM, the chlorophyll index is decreasing. C₃ plants respond to increased or reduced CO₂ concentration due to biochemical and ultra-structural limitations or changes in the canopy level. Also, the increase of CO₂ level up to 800 PPM causes chlorophyll synthesis, which directly affects the rate of photosynthesis and increases the dry weight of plant organs.

Table 8. The effect of CO₂ concentration on Chlorophyll Index of Soybean and Corn

Plant	Chlorophyll Index (Chl Index)		
	400 PPM	800 PPM	1000 PPM
Soybean	34.2	35.9	35.3
Corn	46.0	49.8	47.5

4 Conclusion

The present research showed that increasing CO₂ from 400 PPM to 800 PPM significantly increased the physiological traits of corn and soybean plants. Still, the plant's yield decreased with the increased carbon dioxide concentration to 1000 PPM. This initial increase in the amount of carbon dioxide concentration, directly and indirectly, affects the amount of photosynthesis and the dry weight of plant organs. Current findings can contribute to farmers who plant these oily crops in industrial regions to monitor the yield and physiological traits of these crops. As a recommendation for future research, focusing on the effect of different CO₂ concentrations on medicinal plants will be addressed as well.

Ethical issue

Authors are aware of, and comply with, best practice in publication ethics specifically with regard to authorship (avoidance of guest authorship), dual submission, manipulation of figures, competing interests and compliance with policies on research ethics. Authors adhere to publication requirements that submitted work is original and has not been published elsewhere in any language.

Competing interests

The authors declare that there is no conflict of interest that would prejudice the impartiality of this scientific work.

Authors' contribution

All authors of this study have a complete contribution for data collection, data analyses and manuscript writing.

References

- 1- Nimma D, Devi OR, Laishram B, Ramesh JVN, Boddupalli S, Ayyasamy R, et al. Implications of climate change on freshwater ecosystems and their biodiversity. *Desalination and Water Treatment*. 2025;321:1-9.

- 2- Joshi J, Kumar SSH, Rout RK, Rao PS. Millet processing: prospects for climate-smart agriculture and transition from food security to nutritional security. *Journal of Future Foods*. 2025;5(5):470-479.
- 3- Rowshanaie O, Tohir MZM, Mustapha F, Effendy Ya'acob M, Rowshanaie H. Evaluation of Subcritical Organic Rankine Cycle by Pure and Zeotropic of Binary and Ternary Refrigerants. *Pertanika Journal of Science & Technology*. 2022;30(4):2333-2362.
- 4- Willaume M, Raynal H, Bergez J-E, Constantin J. Optimization of nitrogen management and greenhouse gas balance in agroecological cropping systems in a climate change context. *Agricultural Systems*. 2025;222:1-14.
- 5- Nalley L, Popp M, Fortin C. The Impact of Reducing Greenhouse Gas Emissions in Crop Agriculture: A Spatial and Production-Level Analysis. *Agricultural and Resource Economics Review*. 2011;40(1):63-80.
- 6- Wang A, Lv J, Wang J, Shi, K. CO₂ enrichment in greenhouse production: Towards a sustainable approach. *Frontiers in Plant Science*. 2022;13:1-10.
- 7- Zhang Y, Yasutake D, Hidaka K, Okayasu T, Kitano M. Crop-localised CO₂ enrichment improves the microclimate, photosynthetic distribution and energy utilisation efficiency in a greenhouse. *Journal of Cleaner Production*. 2022;371:1-12.
- 8- Heatherly LG, Ray JD. Soybean and Corn. *Irrigation of Agricultural Crops*. 2007;30(2nd Edition): 477-533.
- 9- Rowshanaie H, Jaafar HZ, Abd. Halim MR, Edaroyati Megat Wahab P, Rowshanaie O. Impact of Different Water Levels on Growth, Plant Water Relations and Leaf Characteristics in Seedling of Tongkat Ali (*EURYCOMA LONGIFOLIA JACK*). *Journal of Multidisciplinary Engineering Science and Technology (JMEST)*. 2014;1(4):197-201.
- 10- Miri HR, Rastegar A. The Effect of Increasing Carbon Dioxide Concentration on the Growth and Competitive Ability of Soybean and Millet with Weeds Such as Sedge and Cocksfoot. *Crop Production*. 2012;1(1):1-18.
- 11- Miri. Effect of CO₂ Enrichment under Drought Stress Condition on Growth and Weed Competitiveness against Corn. *Plant Ecophysiology*. 2013;5(14):59-73.
- 12- Ikhlas Pour M, Rahimi A, Madah Hosseini Sh, Taj Abadi Pour A. Physiological parameters of sesame plants under the influence of salinity stress and increased carbon dioxide concentration. 2018;26(2):115-126.
- 13- Mir Seyyed Hosseini H, Gerdliani AF, Kohestani M, Bihanta M. The effect of carbon dioxide concentration and soil nitrogen availability on wheat growth and physiological indices. *Iranian Journal of Soil and Water Research*. 2017;49(4):767-779.
- 14- Busch FA, Sage T, Cousins AB, Sage RF. C₃ plants enhance rates of photosynthesis by reassimilating photorespired and respired CO₂. *Plant, Cell & Environment*. 2012;36(1):200-212.