Project title:	Increasing crop yield and resource use efficiency via root-zone CO2 enrichment
Project number:	CP 143
Project leader:	Ian Dodd
Report:	09/2016
Previous report:	None
Key staff:	Estibaliz Leibar-Porcel , PhD student Martin McAinsh, co-supervisor
Location of project:	Lancaster University
Industry Representative:	Philip Morley, British Tomatoes Growers' Association
Date project commenced:	1/10/2015
Date project completed (or expected completion date):	30/12/2018

DISCLAIMER

The results and conclusions in this report are based on an investigation conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.

AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

Professor Ian Dodd

Project leader

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Date………………. 2/2/17..

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CONTENTS

GROWER SUMMARY

Headline

CO_2 enrichment of the root-zone applied in the form of gas or bicarbonate could increase shoot growth of lettuce and pepper by 10-20%. In lettuce, this could decrease the time for the crop to reach marketable weight.

Background

Photosynthesis uses light energy to convert CO_2 and water into sugars, which are required for growth and respiration. Biomass accumulation is the difference between the photosynthesis rate and respiration rate. Greenhouse operators often inject extra CO_2 into the aerial environment to increase photosynthesis and dry-matter accumulation. However, when the humidity or the temperature is very high, the greenhouse is vented and CO_2 is released into the atmosphere (Fig. 1), which is economically wasteful and releases a greenhouse gas to the atmosphere.

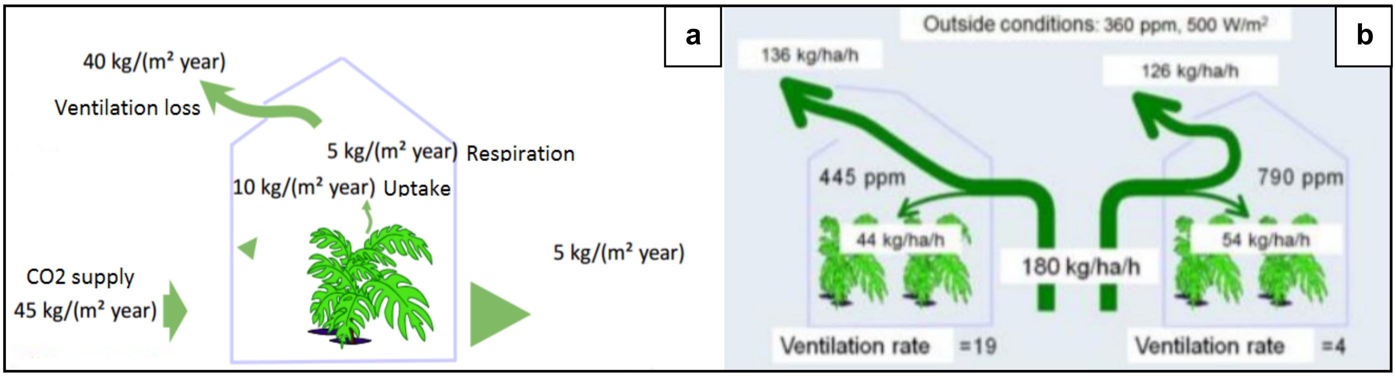

Figure 1. CO_2 balance model. a) General balance model when supplying 45 kg/ (m2 year). b) CO2 balance model when supplying 180Kg/ha/h CO2 and different ventilation rates are applied with same outside conditions. *Wageningen University & Research, Business Unit Greenhouse Horticulture.*

Sources of CO_2 for enrichment include boiler, combined heat and power (CHP) and burner exhaust gases and liquefied pure gas. Flue gases from natural gas boilers are widely used in the UK as a source of CO_2 for enrichment. This practice has high energy costs of £200.000

per annum for a 5 Ha glasshouse (HDC 2011;http://www.hdc.org.uk/sites/default/files/research_papers/PE%20003%20Final%202011_0.pdf). CO_2 gas is a “greenhouse gas” that contributes to global warming and climate change. Despite the efforts of growers to minimize spending and maximize production through technical improvements, it is necessary to consider other systems such as localized root-zone CO_2 enrichment, to improve the production without harming the environment.

This project focused on improving resource use efficiency, the cost-effectiveness and the environmental performance of tomato, lettuce and pepper production, by testing whether rootzone CO_2 enrichment with soilless culture systems provided a viable alternative to aerial CO_2 enrichment.

Summary

Previous studies have shown that applying either bicarbonate to the roots at low concentrations (5 mM HCO_3^-) or gaseous CO_2 at high concentrations (2000 -50.000 ppm) increased growth of some crops such as tomatoes or lettuce. Also, initial studies carried out at Lancaster University by a previous AHDB-funded PhD student indicated that applying 700 ppm CO_2 to the rootzone of semi-aeroponically grown lettuce (without altering the aerial CO_2 concentration) increased biomass by 10%. Therefore, rootzone CO_2 enrichment in greenhouses may provide an alternative technique to increase yield.

Initial studies within this project identified that applying low concentrations of bicarbonate (1-5 mM) to the nutrient solution of hydroponically grown pepper and lettuce increased shoot biomass by 10%. Also, hydroponically grown tomato plants enriched with 1500 ppm root zone CO2 increased dry biomass by 11%.

Although gaseous rootzone CO_2 enrichment is still undergoing additional research, some experiments showed greater biomass (7-10%) in aeroponically grown lettuce. However, these experiments need to be repeated to reach a final conclusion.

Financial Benefits

Developing techniques to more effectively apply CO_2 will decrease the cost of supplying liquefied CO_2 or energy consumption (natural gas boilers) in a commercial scale greenhouses.

Action Points

1. Understand that there are potential alternatives to the current practice of aerial CO_2 enrichment in greenhouses that decrease CO_2 usage and reduce pollution, while maintaining crop yields.

SCIENCE SECTION

Introduction

Generally, soil CO_2 concentration greatly exceeds that of the atmosphere (400 ppm). Root respiration and microbial respiration, including decomposition of organic material, are major contributors to the soil inorganic carbon pool. Concentrations of CO_2 in the soil vary with depth (Johnson *et al.* 1994, Duenas *et al.* 1995), soil water content (Bouma *et al.* 1997), soil type (Duenas *et al.* 1995) and time of the year (Johnson *et al.* 1994) and range from 2000 to 5000 ppm but may become as great as 200.000 ppm when soils are poorly aerated (De Jong and Shappter, 1972; Norstadt and Porter, 1984).

In most higher plants, leaf stomata are the principal means of gas exchange, including the capture of CO_2. Although some aquatic plants assimilate large amounts of CO_2 from the sediments via roots, terrestrial plants are thought to capture insignificant amounts of CO_2 through their roots. However, the terrestrial plant *Stylites andicola*, which lacks stomata, captures almost all of the CO_2 via its roots (Keeley, Osmond et al. 1984), suggesting that some or perhaps all plants can obtain CO_2 from their roots.

In previous studies, several systems have exposed the roots to different CO_2 concentrations, most of them based on hydroponic and aeroponic systems. Hydroponics is a method where plants are grown without soil using a mixture of water and nutrient salts, called a nutrient solution. Aeroponics is a similar technique except that plant roots are suspended in air and sprayed with nutrient solution. In both systems, studies have applied either carbonate (HCO3-) ions (Bialczyk, *et al*. 1992, 1994, 2004, 2005; Alhendawi, *et al*.1997; Al mansouri, *et al.* 2014; Wolfgang Wanek *et al.* 2000 ; Parra Terraza *et al.* 2012 ; X.Yang *et al.* 1994 ; Siddiqi, *et al.* 2002) or gaseous CO_2 (Gao, *et al.* 1997; Bouma, *et al*. 1997; Cramer and Richard, *et al*. 1999 ; Cramer, *et al.* 1999; Van der Merwe, *et al.* 2000; Cramer, *et al.* 2001, Boru, *et al.* 2003; Viktor, *et al.* 2003; Cramer, *et al*. 2005; Viktor, *et al*. 2005; He, *et al.* 2007; X.Zhao *et al*. 2010; He, *et al* 2010; He, *et al.* 2016) (Table1).

CO_2 Gas Experiments	**Gao *et al* (1997)**	**Cramer & Richard *et al* (1999)**	**Cramer, Gao & Lips(1999)**	**Van der Merwe & Cramer (2000)**	**Viktor & Cramer (2003)**	**Viktor & Cramer (2005)**	**Jie He *et al* (2007)**	**Jie He *et al* (2010)**	**Bouma *et al* (1997)**	**Cramer *et al.* (2001)**	**Boru *et al.* (2003)**	**Cramer *et al.* (2005)**	**X.Zhao, T.L. *et al* *(2010)***	**Li *et al.* (2009)**
Crop	Tomato Cv. F144	Tomato Cv. F144	Tomato Cv. F144	Tomato. Cv. F144	Tomato Cv. F144	Tomato. Cv. F144	Lettuce cv. Wintergreen	Lettuce	Citrus cv. Volcamer lemon Bean cv.	Tomato.cv. Daniella	Soybean cv. Williams	White Lupin	Tomato (China variety)	Muskmelon
Treatment	0.2mM NO3-, 0 or 100 NaCl and 4800 ppm CO2 0.2Mm NH4+, 0 or 100NaCl and 360ppm CO2	0,360,5000ppm CO2 + 0 ,70,100,125,150 mM NaCl	360,5000ppm CO2	0,360,5000,10000, 20000ppm CO2	0, 0.5 and 1% CO2	380, 5000ppm CO2	360,2000,10 000,50000pp m CO2	360, 2000, 10000, 50000ppm CO2	600, 20000ppm CO2	5000ppm CO2	1)15% CO2 +85%N2 2)30%CO2+ 70%N2 3)50%CO2+ 50%N2	0,100,360, 6000ppm CO2	370ppm, 2500ppm, 5000ppm, 10000ppm CO2	2500ppm, 5000ppm CO2
Bicarbonate Experiments	**Bialczyk (1992)**	**Bialczyk *et al* (1994)**	**X.Yang *et al* (1994)**	**Bialczyk *et al* (2004)**	**Bialczyk *et al* (2005)**	**Alhendawi *et al.* (1997)**	**Wolfgang Wanek *et al.* (2000)**	**Parra Terraza *et al.* (2012)**	**Hamza Massoud Al mansouri *et al* . (2014)**					
Crop	Tomato cv Torena F1	Tomato cv Torena F1	Rice (zn-inefficient zn-efficient)	Tomato cv Perkoz F1	Tomato cv Perkoz F1	Barley, sorghum and maize	Poplar	Tomato cv Slolly F1	Maize					
Treatment	$KHCO_3^-$ 22.72 mM (0.1% CO2)	$KHCO_3^-$ 0 , 5.68 , 22.72mM	$NaHCO_3^-$ 0,5,10,20 mM	$NaHCO_3^-$ 5mM	$NaHCO_3$ 0,5,10,20 mM $NaHCO_3$ 5mM + NO3:NH4+ (1:1 / 1:4 / 4:1)	$NaHCO_3^-$ 0,5,10,20 mM	$KHCO_3$ 0, 0.5, 1 mM	0, 0.5 and 5 mM HCO_3^- with NO3-/NH4+:100/0, 70/30, 85/15	$NaHCO_3^-$ 0,5,10,20 mM					

Table 1. Previous CO_2 and bicarbonate experiments

1.1 Dissolved inorganic carbon

CO_2 dissolves in water to form dissolved inorganic carbon (DIC) through the following reaction:

$$CO_2 + H_2O = H_2CO_3 \leftrightarrow H^+ + HCO_3^- \leftrightarrow 2H^+ + CO_3^{2-}$$

Dissolved inorganic carbon (DIC) is the sum of dissolved CO_2 gas (CO_2), carbonic acid (H_2CO_3), bicarbonate (HCO_3^-), and carbonate (CO_3^{2-}) (Karberg, 2005).
Solution pH determines the reaction direction of carbonates, and thus the proportion of the carbonate species present in the solution (Figure.1). The prevalent form of carbonates at pH ≤6.36 is H_2CO_3, at pH between 6.36 and 10.33 is HCO_3^-, and CO_3^{2-} is predominant at pH >10.33 (Lindsay, 1979). The solubility of CO_2 increases from pH 5, because at this given pH a proportion of DIC exists as HCO_3^-, and CO_3^{2-} (Golterman and Clymo, 1969).

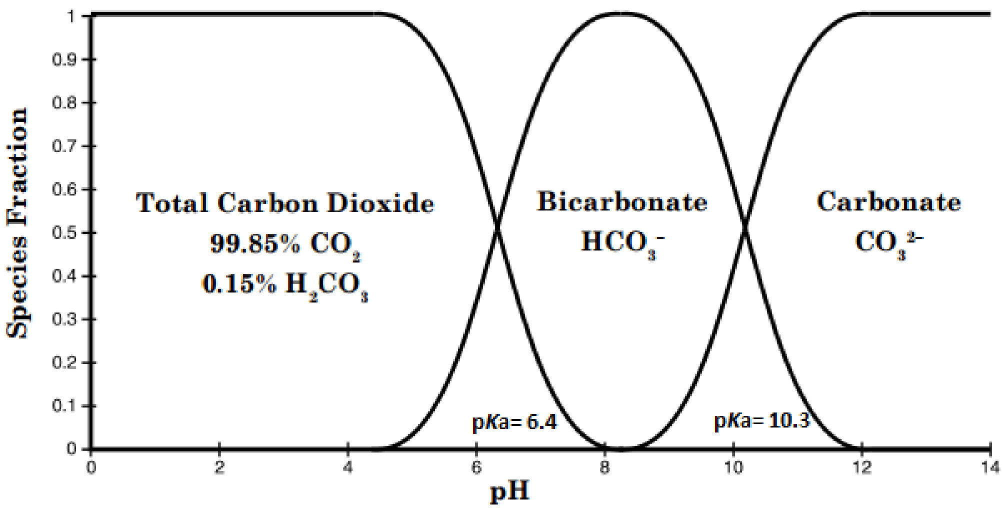

Figure 1. Distribution of total carbon dioxide, bicarbonate and carbonate vs. pH *(John A. Wojtowicz – Chapter 1.1)*

1.2 Incorporation of DIC into roots

In hydroponics, roots grown at 5000 ppm CO_2 took up 9 times more DIC than plants grown at 360 ppm CO_2 (Cramer *et al.,* 1999).This suggests that DIC is incorporated into the root cells. Root incorporation of DIC has been demonstrated using $H^{14}CO_3$ (Vuorinen *et al*., 1992; Cramer *et al*., 1993; Hibberd *et al.,* 2002), and is used for the synthesis of organic and amino acids which are transported by the xylem to the shoots (Bialczyk *et al*.,1992, 1995; Cramer *et al*.,1995, 1999).

Bicarbonate may be transported actively, co- transported with H+ or by exchange with OH-, or there could be a conversion of HCO_3^- to CO_2 or H_2CO_3 externally and subsequent assimilation of CO_2 or H_2CO_3 (Raven, 1984; Lucas *et al* 1985). Dissolved CO_2 gas may diffuse into the cells or may enter via aquaporins. Aquaporins are 23-31 kDa channel proteins present in the plasma and intracellular membranes of plant cells. They facilitate the transport of water, small solutes and gases. Some studies (Sade, 2010, Maurel, 2008) have shown that CO_2 gas can diffuse through these aquaporins located either in the leaves or in the roots.

1.3 RZ CO_2 enrichment affects growth and yield

According to previous studies, the effects of elevated RZ CO_2 on plant growth depend on plant species, pH, air temperature, irradiance, mineral nutrition, abiotic stresses such as high irradiance or salinity, the duration of root zone CO_2 enrichment, CO_2 concentration applied and the RZ CO_2 concentration.

In a review of 358 experiments, Enoch and Olesen (1993) reported a significant mean biomass increase of 2.9% when elevated RZ CO_2 was applied. Despite this low percentage, some authors have reported 1.8-fold more dry matter and leaf blade area in tomato plants, when 5.68 mM of HCO_3^- (0.0025% CO_2) was added to a standard nutrient solution at pH 6.5 (Bialczyk *et al.*1994) . Also, adding 5 mM of HCO_3^- to the nutrient solution containing modified nitrogen concentrations at an optimum ratio (NO3- 4: NH4+ 1) and at pH 6.8 increased biomass of tomato by about 1.8-fold (Bialczyk *et al.* 2005). Cramer and Richards (1999) found that the biomass of both control and salinized (100 mM NaCl) tomato plants increased when the hydroponic solution was aerated with 5000 ppm CO_2 under high irradiance (1500 μmol m^{-2} s^{-1}) and high air temperatures (37/19 °C) at pH 5.8. However, the effect of DIC was 40% greater in non- salinized than in salinized plants. When plants were grown at irradiances less than 1000 μmol m^{-2} s^{-1}, elevated rhizosphere DIC increased growth rates only of control plants grown at high temperatures (35ºC) or salinized plants at more moderate temperature (28ºC). Two weeks' treatment of elevated RZ CO_2 (50 000 ppm) in aeroponically grown crisphead type lettuce increased the growth (~1.6 fold) under 36/30°C and irradiance of 650 μmol $m^{-2}s^{-1}$ at pH 6.5 compared to plants aerated with ambient (360 ppm) CO2 (He *et al*. 2010). Moreover, increasing RZ CO_2 in aeroponically grown lettuce alleviated midday depression of photosynthesis and therefore increased leaf area, shoot and root production (He *et al*. 2007).

The positive effects of increased DIC concentration in the rhizosphere on plant growth can be due to increased DIC incorporation in root cells, enhanced NO_3^- uptake, decreased CO_2 release during root respiration or from changes in shoot gas exchange (Cramer and Richard

1999; J. Qi *et al* 1994). However, negative effects also have been reported. Enrichment with 5, 10 and 20 mM bicarbonate markedly decreased shoot and root dry weight of hydroponically grown barley, sorghum and maize maintained at pH 8 (Alhendawi *et al*., 1997). Aerating semi-hydroponically grown white lupin with 6000ppm RZ CO_2 decreased growth by ~27% compared to control plants grown at 360 ppm CO2 (Cramer et al. 2005). These negative effects were related to decreased root elongation and nutrient uptake and diminished ion transport to aerial organs. However, some of these studies used pH levels as high as 7 or 8 (Alhendawi *et al.* 1997, Wolfgang Wanek *et al.* 2000, Hamza Massoud Al mansouri *et al*. 2014) where the nutrient availability was likely suboptimal. Also, variability of different studies may be due to the different experimental conditions and plant species.

1.4 Objectives

Due to the variable impacts of rootzone CO_2 enrichment in previous studies, the experiments conducted this year aimed to establish different cultural systems to study the effects of rootzone CO_2 enrichment:

- bicarbonate enrichment of hydroponics
- gaseous CO_2 enrichment of hydroponics
- gaseous CO_2 enrichment of aeroponics

In growing 3 horticultural species (lettuce, pepper and tomato) in all growing systems, we hypothesised that impacts of rootzone CO2 enrichment on crop growth were independent of the species and the growing system.

Materials and methods

Experiment 1: Direct bicarbonate enrichment of hydroponics

Aim: Determine the effects of various nutrient solution HCO_3^- concentrations (1, 5, 10 and 20 mM) on the vegetative growth and biomass accumulation of hydroponically-grown tomato, pepper and lettuce plants.

Experimental procedures:

Three hydroponic systems were built, one for each crop. Seeds of tomato (*Lycopersicon esculentum* (L.) Mill. cv. Alisa Craig), pepper (*Capsicum annuum* (L.) cv. Bellboy F1) and lettuce (*Lactuca sativa* (L.) cv. Sunstar), were grown in vermiculite and transferred to hydroponic culture 23 days post germination, after rinsing the roots in water. Pepper and tomatoes were grown in the glasshouse at maximum and minimum temperatures of 25°C/16°C and lettuce in a controlled environment room at 20°C/16°C for 10 days after transferring them to the hydroponics.

The tanks were completely opaque and contained 14L of half-strength Hoagland solution. Bicarbonate was applied in the form of NaHCO3 at 0, 1,5,10 and 20 mM. The medium was changed every 3-4 days and the pH was maintained at 6.4 (at this pH, CO2 and bicarbonate concentrations are equivalent) by adjusting the pH every day with H_3PO_4 or NaOH.

Picture 1. Bicarbonate enriched hydroponic lettuces grown in the CE room.

Experiment 2: Direct gaseous CO_2 enrichment of hydroponics

Aim: Determine the effects of 1500 ppm CO_2 applied in the nutrient solution on the vegetative growth and biomass accumulation of tomato and pepper.

Experimental procedures:

Tomato seedlings (cv. Ailsa Craig) grown in Grodan rockwool were transferred to hydroponic culture 14 days after germination. The hypocotyls of the plants were inserted through neoprene collars in the lids of 20 L hydroponic tanks with 4 plants per tank and two tanks per treatment. The tanks were opaque and contained 16 L of half-strength Hoagland nutrient solution. The medium was changed every 3-4 days and the pH was maintained at 6 by adjusting the pH every day with HCl or NaOH.

Picture 2. CO_2 enriched hydroponic tomatoes grown in the CE room.

After transplanting, two different [CO2] treatments were applied into the nutrient solution. The system consisted of an enriched channel supplemented with CO_2 (1500 ppm) and a non-enriched channel supplied only with compressed air (400 ppm). The air from the enriched channel was completely mixed in a mixing box before feeding the hydroponic tanks. The [CO_2] in the mixing box was monitored continuously using a CO_2 gas analyser (PP Systems, WMA-4). To prevent leakages, the lid was sealed with self-adhesive rubber foam around the rim. The air above the lid and at the shoot base was routinely sampled with a LI-COR 6400 with no significant difference compared to the ambient air.

Experiment 3: Direct gaseous CO2 enrichment of aeroponics

Butterhead lettuce type seedlings (*Lactuca sativa* (L.) cv. grown in Grodan rockwool were transferred to two aeroponic systems (Platinum aero pro-8) 23 days after germination. The hypocotyls of the plants were inserted thought neoprene collars in the lids of 11 L pots with one plant per pot and 8 plants per system. Microsprinklers (flow rate: 52-56 L h-1) misted roots with recirculated half-strength Hoagland's solution coming from a 60 L reservoir. The pH was maintained at 6 by adjusting the pH every second day with HCl or NaOH.
After transplanting, two different [CO2]: 400ppm and 1500ppm were applied into each bin. The system consisted of an enriched channel supplemented with CO_2 and a non-enriched channel supplied only with compressed air. The air from the enriched channel was completely mixed in a mixing box before entering the aeroponic bins. The [CO_2] in the mixing box was monitored continuously using a CO_2 gas analyser (PP Systems, WMA-4).

To prevent leakages, the lid was sealed with self-adhesive rubber foam around the rim. The air above the lid and at the shoot base was routinely sampled with a LI-COR 6400 with no significant difference compared to the ambient air.

Picture 3. CO_2 enrichment of aeroponically grown lettuces in the glasshouse**.**

Results

Experiment 1: Direct bicarbonate enrichment of hydroponics.

Vegetative growth and biomass accumulation of lettuce increased by 10% at 1 mM and 5 mM HCO_3^- (Fig.1) whereas in pepper, this increase was only visible at 1 mM HCO_3^- (Fig. 2).

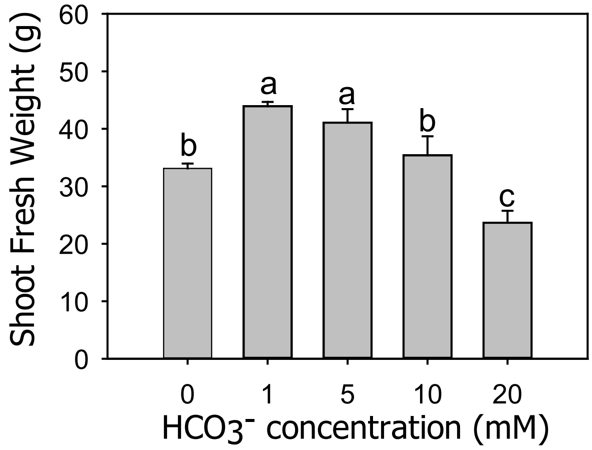

Figure 1. Lettuce shoot fresh weight after two weeks of growth under different HCO3- concentrations. Bars=mean ±SEM (n=8). Different letters indicate significant ($p < 0.05$) differences between treatments.

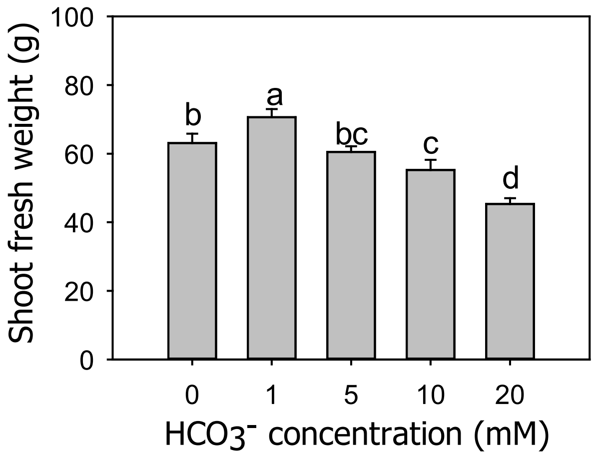

Figure 2. Pepper shoot fresh weight after two weeks of growth under different HCO3- concentrations. Bars=mean ±SEM (n=9). Different letters indicate significant ($p < 0.05$) differences between treatments.

Experiment 2: Direct gas CO_2 enrichment of hydroponics

Aeration with 1500 ppm CO_2 significantly increased dry mass accumulation by 11% compared to aeration with 400 ppm CO_2 (Fig. 3).

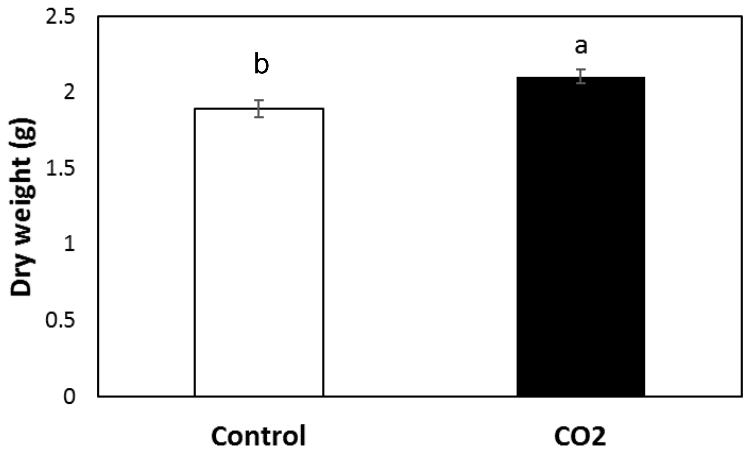

Figure 3. Tomato total dry weight after 11 days of growth under 400ppm and 2000ppm CO2. Bars=mean ±SEM (n=7). Different letters indicate significant ($p < 0.05$) differences between treatments.

Experiment 3: Direct gas CO_2 enrichment of aeroponics.

There was no significant difference in leaf area, shoot fresh and dry weight in tomato plants grown with 1500 ppm root-zone CO2.

CO2 (ppm)	Leaf area (cm^2)	Shoot fresh weight (g)	Shoot dry weight (g)
400	489 ± 37^{a}	13.5 ± 1.2^{a}	2.8 ± 0.2^{a}
2000	470 ± 15^{a}	13.7 ± 0.5^{a}	2.6 ± 0.1^{a}

Table 2. Tomato leaf area**,** shoot fresh weight and shoot dry weight. Bars=mean ±SEM (n=7). Different letters indicate significant ($p < 0.05$) differences between treatments.

Similar results were obtained for lettuce plants grown at 2000 ppm CO2. Although there were no significant differences, plants grown with elevated RZ CO2 had 8 % higher leaf area and 17% higher shoot dry weight (Table 2.)

CO_2 (ppm)	Leaf area (cm^2)	Shoot fresh weight (g)	Shoot dry weight (g)
400	539 ± 19[a]	26.6 ± 1.5[a]	1.31 ± 0.06[a]
2000	582 ± 36[a]	28.4 ± 2.0[a]	1.53 ± 0.09[a]

Table 3. Lettuce leaf area, shoot fresh weight and shoot dry weight. Bars=mean ±SEM (n=8). Letters indicate significant ($p < 0.05$) differences between treatments.

Discussion

Many studies have focused on the impact of increasing atmospheric CO_2 on plant metabolism and physiology, however relatively few studies have considered the impact of rhizosphere CO_2 concentrations. It is almost certain that plant roots are exposed to high CO_2 concentrations in the soil. Moreover, past studies are contradictory since some indicated benefits of enriching the roots with CO_2 (Gao *et al*.,1997; Cramer *et al*.,1999; Van der Merwe & Cramer., 2000; Viktor & Cramer., 2003, 2005; He *et al*., 2007, 2010, 2016), while others showed no significant effect (Cramer *et al*., 2001; Bouma *et al*., 1997) and some even pointed out negative effects of root zone CO_2 enrichment (Cramer *et al*., 2001, 2005; Boru *et al*., 2003; X. Zhao *et al*.,2010; Li *et al*., 2009) (Table 1). This project aims to investigate the physiological and metabolic impacts of enriching the root zone with CO_2 concentrations between 700-2000 ppm on tomato, pepper and lettuce, in trying to understand the mechanisms involved.

Bicarbonate enrichment of hydroponic solutions (1 mM and 5 mM concentration of HCO_3^-) increased shoot growth of lettuce and pepper plants (Fig. 1 and 2). Previously, bicarbonate enrichment of hydroponically grown rice (X.Yang *et al.,* 1994) and tomato (Bialczyk *et al.,* 1994, 2005) stimulated growth. With the right proportions of bicarbonate (5 mM) and N (NO3-4: NH4+ 1) concentrations in the nutrient solution, xylem sap concentrations of amides and amino acids increase, thereby supplying carbon skeletons to NH4+ incorporation and regulating the activity of some enzymes of ammonium metabolism. Therefore, further work is needed to decipher if nitrogen uptake is the only process promoting the growth of bicarbonate enriched plants.

Gaseous CO_2 enrichment of hydroponic solution (1500 ppm CO_2) increased growth of hydroponically grown tomato plants (Fig. 3) but not pepper (data not shown). Previously, positive effects of gaseous CO_2 enrichment were detected when plants were stressed (salinity stress, high temperatures, high irradiance) or at higher rootzone CO_2 concentrations (5.000

ppm, 10.000 ppm, 500.000 ppm). Also, an increase in nitrogen concentration in the nutrient solution combined with elevated bicarbonate or CO_2 may influence the effect of DIC on growth. However, the experiments described herein aimed to study the direct DIC effect, independent of other interactive effects such as salinity, high temperatures, high irradiance or altered nitrogen content in the nutrient solution. Furthermore, the CO_2 concentrations used in this research were lower (700-2000 ppm). For this reason, the lack of significant results obtained in some of our experiments could result from an absence of stress which somehow elevated rhizospheric CO2 could alleviate. Alternatively, perhaps the low CO2 levels applied in our studies are not enough to promote growth even if the plant is absorbing the CO_2 from the roots. Future experiments will focus on determining the uptake of CO2 and nutrients from the roots.

Aeroponics are a good system to study the effect of CO_2 since there are no physical barriers when applying the gas to the root zone. Although CO_2 enriched lettuce plants had 17% higher dry weight (Table 3), 2000 ppm CO2 had no significant effects on tomato and lettuce growth (Table 2, 3). Previous aeroponic experiments with crisphead lettuce concluded that; i) increasing the root zone CO_2 could alleviate midday depression of photosynthesis (*A*) thus increasing the productivity (He *et al.,* 2007) ii) Increased *A* could be due to a higher shoot reduced N (He *et al*., 2010) iii) Growing lettuce at high CO_2 root-zone concentrations (2000, 10000, 50000 ppm) at 20°C-RZT enhance its productivity (He *et al*., 2016). The reason for the lack of significant results, could be that butterhead lettuce (used here) may have a lower root CO_2 uptake capacity than crisphead lettuce (used by He and colleagues). Therefore, future experiments should use different lettuce varieties to determine if there are genotypic differences in response.

Irrespective of treatment differences in biomass accumulation under the different forms of rootzone CO2 enrichment, experiments measuring leaf gas exchange and nitrogen content will help determine whether the possible physiological basis of the responses reported herein and in the literature.

Conclusions

- Bicarbonate enrichment of hydroponics enhanced growth of lettuce and pepper at low HCO_3-concentrations, perhaps by stimulating NO_3- uptake.
- Applying 1500 ppm RZ CO_2 to hydroponically grown tomato plants may stimulate growth although more research is needed to substantiate this conclusion.
- Although previous studies showed that 2000ppm RZ CO_2 stimulated growth of aeroponically-grown lettuce plants, we did not reach the same conclusion, thus further studies are needed.

Knowledge and Technology Transfer

Conferences:

Leibar-Porcel, E. Increasing crop yield and resource efficiency via root-zone CO_2 enrichment. The Great British Tomato Conference. Chesford Grange Hotel. 28-29th September 2016.

Leibar-Porcel, E. Increasing crop yield and resource efficiency via root-zone CO_2 enrichment. Plant & Crop Science Postgraduate Conference, Lancaster University, 4th October 2016.

Posters:

Leibar-Porcel, E. Increasing crop yield and resource efficiency via root-zone CO_2 enrichment. LEC PGR Conference, Lancaster University, 21-22th April 2016.

Glossary

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Appendices

