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SUNFLOWER AND BIOMASS SORGHUM PHOTOSYNTHESIS RESPONSE TO CO_2 ENRICHMENT

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ABSTRACT

Carbon dioxide concentration is expected to increase to 500-1000 ppm by 2100 according to the ecological and social scenarios of IPCC. In this context, the aim of the study was twofold, namely to assess and to compare the sunflower and biomass sorghum photosynthesis responses to CO_2 enrichment at leaf scale, as well as the related interactions with other physiological determinants. We wanted to test if an enrichment of $[CO_2]$ led to a better exploitation of resources (water and radiation) and if there were any differences as regards the performance of the two crops.

In order to evaluate the effect of the CO_2 enrichment at leaf scale, both crops (sunflower and biomass sorghum) were cultivated in Southern Italy under the same conditions. The results obtained showed that an increased concentration from 370 (ambient) to 760 µmol mol^{-1} of CO_2 led to: (i) an improvement in the net photosynthesis rate (e.g. more than 60% in the case of sunflower and more than 110% in the case of sorghum); (ii) lower values for the stomatal conductance, up to 7% in the case of sunflower and 12% for sorghum compared to the ambient carbon dioxide concentration; (iii) water saving (due to the transpiration loss) equal to 0.074 L of water per m^{-2} of leaf surface h^{-1} for sunflower and 0.063 L of water per m^{-2} of leaf surface h^{-1} for sorghum.

In addition, a higher photosynthesis activity coupled with a lower water loss, led to improved instantaneous water use efficiency (WUE). Specifically, doubling the ambient CO_2 concentration, the WUE reached 10.56 mg CO_2 g H_2O^{-1} in the case of sunflower (4.36 mg CO_2 g H_2O^{-1} at ambient CO_2) and 14.07 mg CO_2 g H_2O^{-1} for biomass sorghum (7.67 mg CO_2 g H_2O^{-1} at ambient CO_2).

Key words: sorghum, sunflower, instantaneous water use efficiency, net assimilation rate, leaf gas exchange, CO_2 enrichment**.**

INTRODUCTION

According to the projections of the Intergovernmental Panel on Climate Change (IPCC, 2007) the atmospheric carbon dioxide concentration $[CO_2]$ will increase to 500-1000 ppm by 2100.

Carbon dioxide has a central role for the plant metabolism, affecting growth and implicitly the chemical and physiological processes. Therefore, an increase of CO_2 availability for the photosynthesis leads to an overall improvement of plant growth and biomass accumulation.

In the case of C_3 plants, the fixation of CO_2 occurs through the Calvin cycle, or C_3 cycle, which provides the link with the ribulose-1,5-diphosphate and is catalyzed by the Rubisco enzyme; this enzyme can add to ribulose CO_2 or O_2, which compete for the active site for the enzyme.

C_3 plants, both under hot and drought conditions, close their stomata to limit the water loss. When stomata are closed, CO_2 concentration in the leaves air spaces falls. This triggers the phenomenon of photorespiration, with substrate oxidation without energy gain, decreasing in the same time the efficiency for the fixation of C.

The C_4 plants overcome the photorespiration process through a particular mechanism; the leaves of these plants have the Kranz anatomy with two types of cells: mesophill and bundle sheath around the vascular bundles. In the mesophill, CO_2 is added to the phosphoenolpyruvate, reaction catalyzed by PEP carboxylase, which has a greater affinity for CO_2 than Rubisco. In the

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bundle sheath the oxaloacetate releases the CO_2, starting the Calvin cycle. In this site the concentration of CO_2 is much greater than that of O_2, reducing the photorespiration and increasing the efficiency for the fixation of C.

Under elevated [CO_2] the rates of photosynthesis of C_3 plants are enhanced (Acock and Allen, 1985; Chen et al., 1995; Ainsworth, 2008), whereas contrasting results are reported for C_4 plants. Under well watered conditions, some studies reported that elevated [CO_2], affected positively leaf photosynthesis (Le Cain and Morgan, 1998; Ziska et al., 1999) of C_4 plants, while other researches have indicated the increase in photosynthesis activity only in dry conditions (Adam et al., 2000). Other authors underlined that photosynthesis rate P_N was not affected significantly (Chen et al., 1995; Rogers et al., 1983) when [CO_2] increased.

The variation in photosynthesis rate is not the only effect due to [CO_2] enrichment. Some other effects can be mentioned, namely: (i) the stomatal closure in response to the elevated level of [CO_2] (Mott, 1990) which reduces the stomatal conductance (g_s; Knapp et al., 1994); ii) transpiration (E) decreases for both C_3 and C_4 plants (Mott, 1988; Assmann, 1999; Wand et al., 1999), although with different magnitude between species with different photosynthetic carbon pathways.

In response to [CO_2] enrichment, different paths for C_3 and C_4 plants have been noticed; the differences in photosynthetic activity have an impact on the final biomass accumulation and, on average, the growth stimulation is about 40-44% in the case of C_3 plants and 22-33% for C_4 plants, doubling the values for current [CO_2] (Poorter, 1993; Wand et al., 1999).

The analyzed data from previous research studies showed that photosynthetic behaviours of both C_3 and C_4 species are driven by different variables other than [CO_2]. This cannot furnish univocal response according to climatic condition, soil type and crop management. For instance in sunflower, Garofalo and Rinaldi (2015) indicated as the water stress lowered P_N up to 61 % compared to the well watered plants, according to the drought timing. Therefore, impacts of climate change in stimulating photosynthetic activity could change according to the environmental conditions and, on the same site, to crop management. The reduction of the erratic effect of these variables, lead to a more accurate evaluation of different photosynthesis patterns between C_3 and C_4 species.

The study of the effects of [CO_2] enrichment on plants, may involve morphological and productive traits, and/or physiological aspects at leaf scale. Research regarding the changes of vegetative habitus of crop has been carried out through the free air CO_2 enrichment (FACE) or open top chamber (OTC). In FACE system, a given perimeter of cultivated field is bounded by a set of pipes emitting a continuous flow of CO_2 to maintain a steady prefixed value of [CO_2]. Being an open system, the demand for CO_2 is high, affecting the operating costs of the system and under extreme wind conditions, [CO_2] on plants can undergo substantial changes. To overcome these issues, in OTC system, the crop usually is grown in pots and placed in a chamber with specific size and covering material as fibreglass, polycarbonate or Plexiglas. OTC system has the advantage of simplicity, cheapness, easiness of installation and a better control of [CO_2]. However the covering layer of the OTC system also has some disadvantages, such as: the higher temperature than the outside, altered radiation and humidity that affects the crop physiology and growth (D'Andrea and Rinaldi, 2010). Finally, in the OTC systems, the [CO_2] is established at the beginning of the experiment, thus the crop response is univocal; for this reason the "crop response curve" at different levels of [CO_2] is not achievable.

To partially overcome these issues, the infrared gas analyzer (IRGA) on crop grown under normal field conditions can be used. With this portable and friendly instrument, the physiological aspects of the plant at leaf level, such as transpiration, photosynthesis and stomatal conductance can be analyzed, by imposing different [CO_2] concentrations. Obviously, with this system, any direct assessment of morphological changes in response to different [CO_2] at field scale cannot be performed. However, the physiological data obtained with IRGA can be implemented in the crop growth model, photosynthetically-based (de Reffye et al., 1999; Fournier et al., 2003; Prusinkiewicz, 2004; Gayler et al., 2008; Génard et al., 2008; Evers et al., 2010), in order to check trends of crop growth in response to CO_2 changing.

In this paper we present the experimental results of a study that has considered sunflower (a C_3 plant) and biomass sorghum (C_4 plant). Both crops were grown under the same conditions. The main aim of the study was to assess and to compare the response of photosynthesis and the corresponding interactions to future carbon dioxide enrichment at leaf scale; more specifically we wanted to test if there are different performances of the two crops, as regards a better exploitation of [CO_2] availability and improvement in future resources use efficiency.

MATERIAL AND METHODS

The research was carried out at the experimental farm of the Council for Agricultural Research and Economics - Research Unit for Cropping Systems in Dry Environments, in Foggia (lat. 41° 8’ 7’’ N; long. 15° 83’ 5’’ E, alt. 90 m a.s.l.), Southern Italy, in 2010.

The soil is a vertisol of alluvial origin, Typic Calcixeret, (Soil Taxonomy 10th ed., USDA 2010), silty-clay with the following characteristics: organic matter, 2.1%; total N, 0.122%; $NaHCO_3$ extractable P, 41 ppm; NH_4O Ac-extractable K_2O, 1598 ppm; pH (water), 8.3; field capacity water content, 0.396 m^3 m^{-3}; permanent wilting point water content, 0.195 m^3 m^{-3}, available soil water, 202 mm m^{-1}. The local climate is „accentuated thermo-Mediterranean as classified by FAO-UNESCO Bioclimatic Maps (1962), with daily temperatures below 0 °C in the winter and above 40 °C in the summer. Annual rainfall (average 550 mm) is mostly concentrated during the winter months, while only 101 mm of precipitation/of rainfall is recorded, on average, during sorghum crop cycle (1st May - 15th August).

Biomass sorghum (*Sorghum bicolor* L., hybrid BIOMASS 133) and sunflower (*Helianthus annuus* L., hybrid SANBRO) were sown at first ten-day of May (25 and 7.5 plants per m^{-2} for sorghum and sunflower, respectively) and the harvest occurred at flowering for sorghum (first ten-day of August) and at maturity for sunflower (beginning of September). As pre-sowing fertilization 2 q ha^{-1} of diammonium phosphate was supplied to the crops. Both crops were cultivated in well watered conditions, supplying the water consumed by the crop, each time the crop evapotranspiration (ETc), calculated multiplying the reference evapotranspiration with FAO–Penman–Monteith method (ET_0; Allen et al., 1998) by the crop coefficient (kc; Garofalo et al., 2011), reached 60 mm.

Rainfall recorded during the crop growing cycle was 59.2 mm; total water supplied to the crops resulted equal to 452 and 338 mm for sorghum and sunflower, respectively. ETc was 706 mm for sorghum and 446 mm for sunflower.

Leaf gas exchange rates and associated parameters were measured on two new fully expanded leaves during the vegetative phase of three plants randomly chosen in the field, in the upper part of the canopy exposed to full sunlight, using a Photosynthesis System LCpro+, Portable Infrared Gas Analyzer (ADC, BioScientific Ltd., Hoddesdon, Herts, UK). Three measurements for each leaf were recorded automatically every 2 min., after changing the [CO_2] in the leaf chamber, to ensure a steady-state condition for the gas exchange flow.

The [CO_2] in the leaf chamber (c_{ref}) was increased step by step, starting with 370 (ambient), followed by: 450, 580, 680 and 760 μmol mol^{-1}.

The light units (the diode array contains blue LED’s at 470 nm and red ones at 660 nm) upper the jaw enclosing the leaf, were used to ensure a constant irradiance replicating the sunlight during the midday (PAR; 1566 μmol m^{-2} sec^{-1}).

The chamber cooler was set to avoid the increase of the temperature in the jaw up to 10°C from the ambient.

The measurements were carried out in the same three days for both crops: 30 days (juvenile phase; Jph), 60 days (full canopy expansion; Fph) and 90 days (beginning of senescence; Sph) after sowing. Parameters measured by the gas-exchange system were: leaf photosynthetic rate (P_N; µmol m^{-2} s^{-1}), sub-stomatal [CO_2] (C_i; µmol mol^{-1}), stomatal conductance (g_s; mol m^{-2} s^{-1}) and leaf transpiration rate (E; mmol m^{-2} s^{-1}).

Instantaneous water use efficiency (WUE; µmol CO_2 $mmol^{-1}$ H_2O) was calculated as ratio between P_N and E, and the instantaneous quantum yield as ratio between P_N and PAR (Q_y; µmol $mmol^{-1}$).

Analysis of variance of the data was carried out using a "randomized block" design model, and least significant difference (LSD) was used to compare mean values.

RESULTS AND DISCUSSION

1. Comparison between C_3 and C_4: ambient [CO_2]

The growth phase had effect on P_N and the other measured parameters, with different pathway for C_3 and C_4 plants, as shown in Table 1. For **sunflower**, P_N resulted higher in the first and second sampling, whereas in Sph the photosynthetic activity was almost halved. After the maximum leaf expansion, the sink activity decreased, and therefore, the demand for the photosynthetates was lower, hence for the net photosynthesis rate. In Fph, g_s decreased by 22%, this may be explained by the high temperatures of this period (July), compared to the coldest period during the juvenile phase (May - June). As a consequence of the decline of g_s, E also decreased by 24%, compared to the values recorded during juvenile period and initial senescence period.

According to the results, C_i cannot be linked to the stomatal conductance, resulting statistically similar during all the measurements. This confirms that as plant matures, the capability of leaves to fully exploit the intercellular [CO_2] is reduced. This leads to lower values for P_N and in the same time keeps similar concentration for C_i, despite the reduction of flow of CO_2 into the leaves, driven by the stomatal closure.

WUE was influenced by the trend of P_N and E. Indeed, the highest value for WUE was recorded at full canopy expansion, due to the greatest net photosynthetic rate, while the lowest WUE was observed at Jph, because of the greatest transpiration rate of the young leaves.

The pathway of P_N influenced the Q_Y, the latter showing comparable values during the active growth of plants (16.7 µmol $mmol^{-1}$) as well a lower value at the beginning of senescence.

Table 1. Net photosynthesis rate and associated parameters for sunflower and sorghum during the crop growing cycle at ambient [CO_2]

Different letters for the same crop, indicate significant differences between means among growth phases, and different letters between averages indicate significant differences between crops at P<0.05 level (LSD test)

Crop	Phase	P_N µmol m^{-2} s^{-1}	g_s mol m^{-2} s^{-1}	E mmol m^{-2} s^{-1}	C_i µmol mol^{-1}	WUE µmol CO_2 $mmol^{-1}$ H_2O	Q_Y µmol $mmol^{-1}$
Sunflower	Jph	24.11 a	0.93 a	6.78 a	236	3.56 b	15.6 a
	Fph	27.69 a	0.73 ab	6.37 a	218	4.36 a	17.7 a
	Sph	17.92 b	0.48 b	5.21 b	247	3.45 ab	11.6 b
	Avg.	23.24 a	0.71 a	6.12 a	234 a	3.79 b	14.8 a
Sorghum	Jph	25.84 b	0.29 b	4.20 ab	134 b	6.07 b	16.5 b
	Fph	35.98 a	0.38 a	4.97 a	113 b	7.67 a	23.0 a
	Sph	19.62 b	0.23 ab	3.71 b	161 a	5.63 b	12.5 b
	Avg.	26.57 a	0.30 b	4.25 b	136 b	6.23 a	17.3 a

Abbreviations: Jph = juvenile phase; Fph = full canopy expansion; Sph = beginning of senescence;
P_N = leaf photosynthetic rate; g_s = stomatal conductance; E = leaf transpiration rate; C_i = sub-stomatal [CO_2]; WUE = instantaneous water use efficiency; Q_Y = instantaneous quantum yield.

Different behaviour was observed for **sorghum**, more specifically the best performances of the analyzed parameters (excepting C_i) were observed at Fph. In this phase the higher temperatures and drought conditions, led to defensive mechanisms of plants by reducing the transpiration. The C_4 crops due to the lower C_i, stomatal conductance and consequently leaf transpiration, suffer less under the heat stress or drought condition (Vitkauskaitè and Venskaitytè, 2011) showing an advantage in term of P_N and WUE over C_3 plants. Again, the advantage of sorghum is referring to the full canopy expansion period.

The highest value for P_N, recorded at maximum canopy expansion, could be explained by the characteristics of C_4 leaves at different development stages. Indeed, Ghannoum et al. (2000), reported that young C_4 leaves have C_3-like characteristics (similar value for P_N during Jph, both for sorghum and sunflower) and the cell-specific accumulation of photosynthetates begins later (with the initiation of veins in monocotiledones and dicotiledones). As consequence, in the fully expanded leaves, the photosynthetic rate resulted greater than 39% as compared to the young leaves (and higher than sunflower), whereas in Sph the photosynthetic rate was halved compared to Fph. Indeed, in the last part of the growing cycle, suberization of leaf tissue reduced the g_s and, consequently, the transpiration of leaves (Ghannoum et al., 2000). This led to a lower capability of the leaf mesophill for the photosynthesis process, explaining a higher $[CO_2]$ in the sub-stomatal cavity despite a lower CO_2 flow into the leaf (consequent to the stomatal closure).

As for P_N, Q_Y also resulted higher in Fph and lower during the last part of the growing cycle, with a maximum difference of 44% between the two phases.

At ambient $[CO_2]$, the overall path for P_N resulted statistically similar for C_3 and C_4 plants, although the intercellular CO_2 to be allocated to the photosynthesis process was lower in sorghum than in the sunflower. Indeed, in the case of C_3 plants, Rubisco enzyme involved in photosynthesis is also implicated in the uptake of CO_2, whereas for C_4 plants, the delivery of CO_2 to the Rubisco is assigned to the PEP Carboxylase. This means a faster and more efficient processing of carbon dioxide for C_4 than C_3 crops, allowing a lower CO_2 accumulation in the leaf tissues before to being processed by the photosynthesis. This entails also a shorter time for the stomatal opening of C_4 compared to C_3 species, for the same amount of CO_2 gain for photosynthesis.

As result, the lower g_s recorded for sorghum during all the measurements (0.41 mol m^{-2} s^{-1} less than sunflower), led to a water saving (due to transpiration), equal to 31% compared to sunflower.

From these measurements a better value for the WUE emerged in the case of C_4 compared to C_3 plants, with an overall improved performance of 64%.

The instantaneous quantum yield efficiency resulted slightly higher in sorghum than in sunflower, namely an increase of 13%.

2. Comparison between C_3 and C_4: enriched $[CO_2]$

The analysis of the behaviour of C_3 and C_4 plants in response to CO_2 enrichment, was carried out during the measurements at maximum canopy development (Fph), corresponding to the maximum P_N both for sorghum and sunflower.

The increase of $[CO_2]$ at leaf scale, determined significant increment of C_i, both for C_3 and C_4 plants. Indeed, doubling the c_{ref} from the current value, also the sub-stomatal $[CO_2]$ resulted almost double in the case of sorghum and more than double in the case of sunflower (Figure 1a), indicating the capability of plants to capture CO_2 to the atmosphere, proportionally to its increment.

Despite the rate of the carbon sap in the leaf tissue was similar for sorghum and sunflower, the final gain in term of P_N resulted quite dissimilar. With c_{ref} ranging between 370 and 580 μmol mol^{-1}, C_4 plants kept the advantage in term of P_N; more specifically the P_N oscillated between 0.097 and 0.09 μmol mol^{-1} per μmol mol^{-1} of c_{ref}, greater than 18% compared to C_3 plants (Figure 1b). With further increases in c_{ref}, the photosynthetic gap between C_3 and C_4 plants rapidly closed, to vanish completely at 760 μmol mol^{-1}.

At the highest level of c_{ref}, the values of P_N were comparable between sorghum and sunflower (58.57 μmol m^{-2} s^{-1}, on average). However, under different $[CO_2]$ (starting from ambient and then doubling the [CO_2]), the gain in term of P_N varies, resulting 76% greater in the case of C_3 compared to C_4 species.

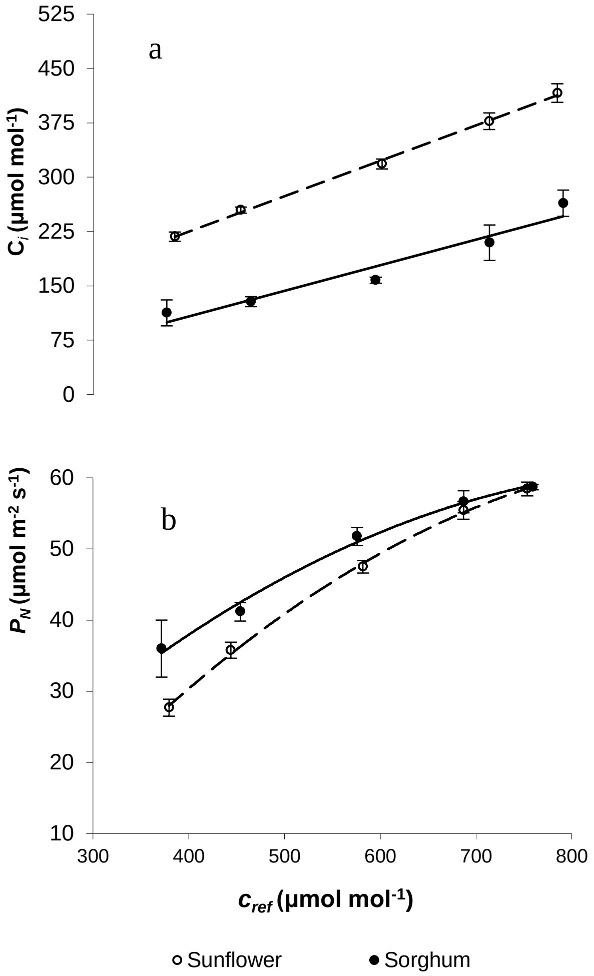

Figure 1. Intercellular [CO_2] (*C_i*; **a**) and net photosynthesis rate (*P_N*; **b**) for sunflower and sorghum, at different level of leaf [CO_2] (*c_{ref}*)
Bars indicate the standard error.

Some authors (Taiz and Zeiger, 1991; von Caemmerer et al., 1997) reported that for C_4 plants, the saturation for P_N is reached near the current [CO_2] level, with a small improvement in term of net photosynthetic rate, as a consequence of the increase of [CO_2]. In the case of C_3 plants, the saturation for P_N (similar to the C_4 plants) is reached by doubling the current [CO_2].

On the other hand, several studies (Wand et al., 1999; Ziska et al., 1999) have shown that saturation of photosynthesis for C_4 species is achieved at a higher level of [CO_2] than the current one, especially when crops are in well watered regimes.

In our study, sorghum achieved the saturation for P_N (58.71 µmol m^{-2} s^{-1}), when the C_i doubled the initial sub-stomatal [CO_2] (Figure 2), confirming the results of Ziska and Bunce (1997) with saturation for P_N in sorghum, at 220 µmol mol^{-1} of C_i.

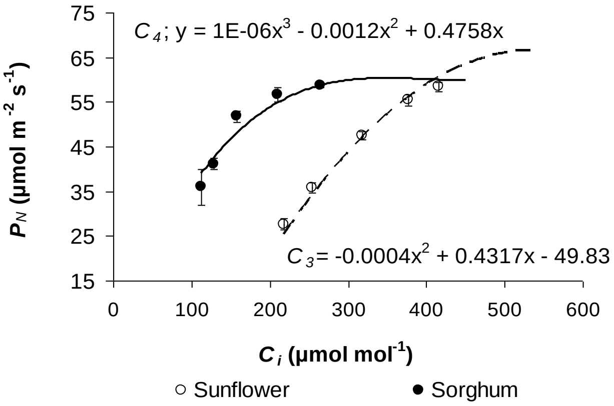

Figure 2. Net photosynthesis rate (*P_N*) at different level of intercellular [CO_2] (*C_i*) for sunflower and sorghum
Bars indicate the standard error.

For C_3 species, the saturation is reported to be achieved at levels of intercellular [CO_2] higher than in the case of C_4 plants. In soybean, the plateau of P_N is reached close to 500 µmol mol^{-1} of C_i (Sims et al., 1998). Also for rice, Makino et al. (1994) indicated a significant increment in P_N when the intercellular CO_2 pressure raises from 49 to 395 µmol mol^{-1} and then stabilized close to 444 µmol mol^{-1}.

Specifically for sunflower, in our study, we observed that P_N was saturated when C_i reached 500 µmol mol^{-1}, a lower value if compared with that (close to 800 µmol mol^{-1}) reported by Makino and Mae (1999) for the same crop.

So, both for C_3 and C_4 species, the saturation point for P_N, requires defined level of C_i, that in this research, resulted halved in sorghum compared to sunflower.

Figure 3a shows that a rise of the [CO_2] level in the leaf chamber, declines the g_s. Moreover, when c_{ref} equals 760 µmol mol^{-1}, g_s decreased by 7% compared to ambient [CO_2] in the case of sunflower, and by 12% compared to ambient [CO_2] for sorghum.

CO_2 enrichment leads to a decrement in g_s both for C_3 and C_4 plants, with a magnitude that varies according the studies reported in the literature. Thus, in a review of Ainsworth and Rogers (2007), it has been shown that in FACE experiments, the decrease of g_s could oscillate between 20 and 30% in the case of C_3 crops, as well between 19 and 39% for C_4 crops, when the [CO_2] passed from 366 to 567 µmol mol^{-1}.

As regards the open top chamber experiments, Chen et al. (1995) found that g_s passed from 1.73 to 0.89 mol m^{-2} s^{-1} in the case of sunflower and from 0.64 to 0.18 mol m^{-2} s^{-1} in corn, when the environmental [CO_2] rose from 350 ppm to 700 ppm.

For sunflower, Maddaluno et al. (2011) reported that the plants grown in water stress condition, reduced the g_s by 62% compared to well watered plants, when the [CO_2] was set to 760 µmol mol^{-1}.

However, climatic condition affects the physiological response of crops to [CO_2] enhancement, especially when vapour pressure deficit condition is low; the crop tends to emphasize the effect of enriched [CO_2] on stomatal closure (Wullschleger et al., 2002).

Moreover, the growth under low N and drought, exacerbates the decrease of g_s when the plants are submitted to the increase of [CO_2], with magnitude almost doubled on the stomatal closure, compared to optimal growth conditions (Ainsworth and Long, 2005).

From these data, we can emphasize that between g_s and the [CO_2] enrichment there is an inverse relationship; crop management and pedoclimatic conditions could also impact highly on the relative magnitude. The small variation for g_s observed in this study, can be explained by the crop management, targeted to reduce the water and nitrogen stress for plants.

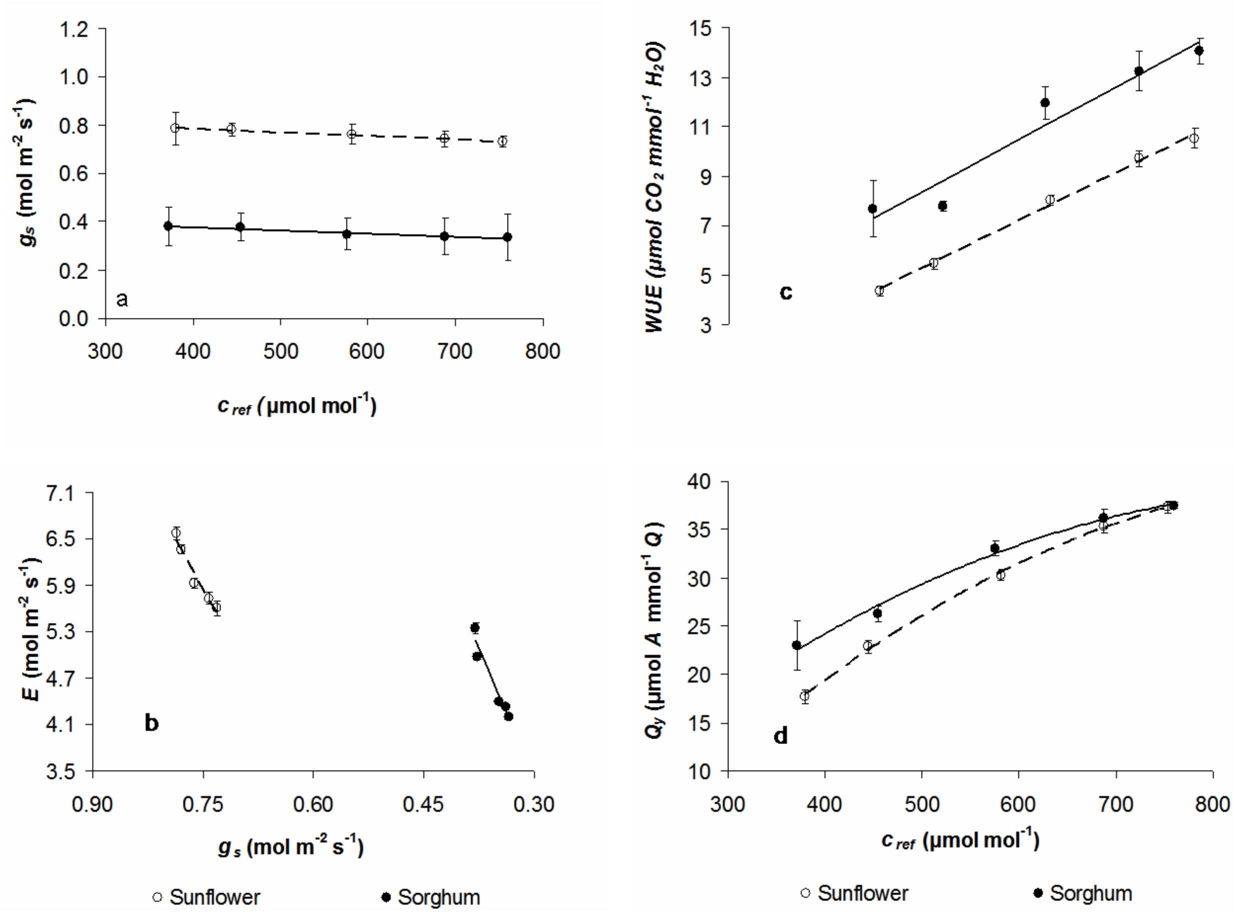

Figure 3. Stomatal conductance (g_s) at different level of [CO_2] (**a**) and response of leaf transpiration (*E*; **b**) to g_s changes. Instantaneous water use efficiency (*WUE*; **c**) and quantum yield (*Qy*; **d**) at different level of leaf [CO_2] Bars indicate the standard error.

However, to a small decrement for the stomatal conductance, corresponds a significant reduction in the leaf transpiration, equal to 15 and 22% for sunflower and sorghum, respectively (Figure 3b). This means water saving due to reduced E, equal to 0.074 and 0.063 L of water per m^{-2} of leaf surface h^{-1} for sunflower and sorghum, respectively. For sorghum, it can be considered a mean value for the leaf area index from the juvenile to beginning of senescence phase, equal to 5.9 m^2 m^{-2} (Rinaldi and Garofalo, 2011) and 4.6 m^2 m^{-2} for sunflower (Goksoy et al., 2004), as well a day light length for both crops equal to 10 hours. This means a water saving during the whole crop grown equal to 393 and 261 mm for sorghum and sunflower, respectively.

The larger P_N and the lower transpiration rate, observed at higher level of [CO_2] resulted in a greater WUE (Figure 3c). The sunflower was the crop that mostly took advantage by [CO_2] enrichment, more specifically the WUE increased by 141%, doubling the current [CO_2], whereas the increment for sorghum was 83%.

For C_3 species, such as cotton, Reddy et al. (1995) reported an increment in WUE at 700 μmol mol^{-1} [CO_2] equal to 100%, whereas for sunflower, Chen et al. (1995) underlined that the improvement in instantaneous water use efficiency resulted of 90%, a percentage not so dissimilar to that for corn (97%) when [CO_2] passed from 350 to 700 μmol mol^{-1}.

However, the advantage in term of WUE seemed to be due mainly to a better exploitation of water availability (hence, the P_N) rather than a lower water requirement, or reciprocal of water use efficiency. The latter was 54% for sorghum and 41% in sunflower at doubled c_{ref}.

If improvement in P_N is reported for the well watered conditions, in water stress condition, especially for C_3 plants, different studies showed that drought condition negatively impacts on photosynthesis and accelerates the photorespiration rate by stomatal closure in C_3 plants, reducing the activity of key enzymes in C metabolism (Maroco et al., 2002) and down-regulation of photosynthetic apparatus by low C availability (Tourneux and Peltier, 1995).

However, even for C_4 species, water stress causes the reduction in plant transpiration through stomatal closure; transpiration may be lower up to 61% comparing well watered and stressed plants, but with no adverse effects on WUE (Garofalo and Rinaldi, 2013).

Finally, the path for Q_y, resulted quite similar for both species (Figure 3d). The gain in term of Q_y, was slightly higher in sorghum than sunflower up to 580 μmol mol^{-1} [CO_2], but above this concentration, the gap resulted null and the final gain was equal to 14.5 and 19.6 μmol $mmol^{-1}$ for sunflower and sorghum, respectively.

CONCLUSIONS

This research study assessed the gas exchanges and associated parameters at leaf scale, in response to [CO_2] enrichment for C_3 (sunflower) and C_4 (biomass sorghum) crops in well watered conditions. The main findings of the experiment were that for any increment of [CO_2] lower or equal to 300 μmol mol^{-1} to the ambient value, the P_N greatly improved in the case of C_4 crop. With further increase of carbon dioxide, the C_3 plants retrieved this gap or, pushing the [CO_2] beyond 760 μmol mol^{-1} may have even better performances.

However, the increase of [CO_2], as predicted by the future climate scenarios, will be accompanied by raise of temperature and less rainfall. In these conditions, the C_4 plants through the abatement of the stomatal conductance and leaf transpiration, combined with a faster intercellular CO_2 processing, could fit better to the changing environmental conditions than C_3 species, enhancing leaf P_N and growth via increases in C_i improvements of shoot water relations and increases in leaf temperature. Indeed for C_3 species, adverse climatic issues (high temperatures, drought) together with [CO_2] enrichment, may cause an excessive stomatal closure by increasing the rate of photorespiration.

However, as underlined from our study, in well watered conditions, the increases in carbon dioxide concentration led to an improvement in plant carbon assimilation and processing, by: (i) increasing the P_N, (ii) reducing the water transpired by the plants and (iii) improving the water use efficiency and quantum yield. These effects were more pronounced for the C_4 plants at the initial increases of [CO_2], indicating a better flexibility, compared to the C_3 species, in the environmental short term CO_2 enrichment. However, optimal cropping management could exploit the higher carbon availability also for C_3 plants and, as reported, these species may be more efficient compared to C_4 species, especially in the long term carbon dioxide enrichment.

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