

Article

The Influence of Elevated CO₂ on Volatile Emissions, Photosynthetic Characteristics, and Pigment Content in *Brassicaceae* Plants Species and Varieties

Andreea Lupitu , Cristian Moisa , Simona Gavrilas , Mihaela Dochia, Dorina Chambre, Virgiliu Ciutină, Dana Maria Copolovici  and Lucian Copolovici * 

Faculty of Food Engineering, Tourism and Environmental Protection, Institute for Research, Development and Innovation in Technical and Natural Sciences, Aurel Vlaicu University of Arad, Elena Drăgoi St., No. 2, 310330 Arad, Romania; pag.andreea@yahoo.com (A.L.); moisa.cristian@yahoo.com (C.M.); simona2213@yahoo.com (S.G.); dochiamihaela@yahoo.com (M.D.); dorinachambree@yahoo.com (D.C.); virgiliutina@yahoo.com (V.C.); dana.copolovici@uav.ro (D.M.C.)

* Correspondence: lucian.copolovici@uav.ro; Tel.: +40-745259816

Abstract: Climate change will determine a sharp increase in carbon dioxide in the following years. To study the influence of elevated carbon dioxide on plants, we grew 13 different species and varieties from the *Brassicaceae* family at three carbon dioxide concentrations: 400, 800, and 1200 ppmv. The photosynthetic parameters (assimilation rate and stomatal conductance to water vapor) increase for all species. The emission of monoterpenes increases for plants grown at elevated carbon dioxide while the total polyphenols and flavonoids content decrease. The chlorophyll content is affected only for some species (such as *Lipidium sativum*), while the β -carotene concentrations in the leaves were not affected by carbon dioxide.

Keywords: green leaf volatiles; induced emissions; monoterpene emission; photosynthesis; quantitative responses; volatile organic compounds



Citation: Lupitu, A.; Moisa, C.; Gavrilas, S.; Dochia, M.; Chambre, D.; Ciutină, V.; Copolovici, D.M.; Copolovici, L. The Influence of Elevated CO₂ on Volatile Emissions, Photosynthetic Characteristics, and Pigment Content in *Brassicaceae* Plants Species and Varieties. *Plants* **2022**, *11*, 973. <https://doi.org/10.3390/plants11070973>

Academic Editor: Carmen Arena

Received: 20 February 2022

Accepted: 31 March 2022

Published: 2 April 2022

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

Carbon dioxide is the main greenhouse gas emitted into the atmosphere due to human activities. Global CO₂ concentration has increased more than ever in the last 20 years as the carbon dioxide amount has grown by 43.5 ppmv, increasing 12 percent. The actual concentration of carbon dioxide will exceed 420 ppmv at the end of 2021. The projected CO₂ concentrations in 2100, under a range of emissions scenarios developed for The Intergovernmental Panel on Climate Change (IPCC), vary from 500 ppmv to 1200 ppmv [1]. The critical impacts of projected climate change on plants are considered inevitable [2]. The increase in carbon dioxide emission could lead to more climate change, including different episodes of extreme heat, droughts, or flooding that stress terrestrial vegetation [3]. Plants can respond to rising atmospheric CO₂ concentrations by increasing the water-use efficiency and photosynthetic rates [4–6]. Free-air CO₂ enrichment (FACE) experiments, which use plants grown in open-air environments enriched with carbon dioxide, have been shown to increase the yield of food crops, even compared to chamber experiments [7].

Regarding photosynthetic parameters, increasing carbon dioxide determines an increase in photosynthesis for C3 plants [8,9]. On the other hand, different studies have shown elevated carbon dioxide downregulation of photosynthetic capacity [10]. Such limitation has been found for *Quercus ilex* L. leaves and has probably been due to the low capacity for ribulose-1,5-bisphosphate regeneration [11]. The net photosynthetic rate dropped for *Glycine max* (L.) plants grown at elevated carbon dioxide levels due to stomatal traits and mesophyll tissue size changes [12].

The emission of volatile organic compounds (VOC) from plants grown at high carbon dioxide is enhanced compared with plants grown at the actual CO₂ concentration

(see [13] for a review). For example, the isoprene emission increases in *Phragmites australis*, *Platanus × acerfolia*, and *Populus nigra × maximowiczii* NM6 when grown in high carbon dioxide [14]. The abundance and diversity of plant volatile organic compounds emission from *Hordeum vulgare* L. seedlings, including aromatics, terpenes, and green leaves volatiles, were also changed by elevated CO₂ compared with the actual carbon dioxide concentration [15]. In contrast, foliar VOC concentrations are unaffected by the growth conditions (700 ppmv CO₂ compared to 400 ppmv CO₂) for *Artemisia annua* plants [16].

It has been found that elevated CO₂ increases the foliar contents of polyphenols of tea seedlings but decreases in free amino acids and caffeine [17]. Carbon dioxide fertilization has not affected the total phenols determined in chickpea leaves [18]. Leaf polyphenol concentrations found in two Cerrado native species, *Baccharis dracunculifolia* and *B. platypoda*, grown at 400 ppmv and 800 ppmv carbon dioxide, have not been statistically significant [19].

The concentration of chlorophylls in the leaves of plants grown at elevated carbon dioxide decreases despite the total phenols concentration. The rate of decline in the chlorophyll contents of *Oryza sativa* L. leaves was faster in plants grown under high carbon dioxide, mainly in the later growth period [20]. Exposing tomato leaves to various CO₂ treatments revealed a decrease in chlorophyll *a* and *b* [21]. In a recent study with terrestrial plants, it has been shown that chlorophyll fluorescence decreases with an increase in carbon dioxide concentration [22]. In contrast, the total chlorophyll increased in chickpea grown at 700 ppmv CO₂ compared to the ambient conditions at the flowering stage. Such behavior could be explained by symbiotic nitrogen fixation, which can fulfill its N requirement through plant N uptake, accelerated under elevated atmospheric CO₂ [23]. An increase in chlorophyll content with 33% has been found for pak choi (*Brassica rapa* ssp. *chinensis*) plants grown at 800 ppmv carbon dioxide compared to the control [24]. It was also found that elevated carbon dioxide could increase the concentrations of photosynthetic pigments (chlorophylls and carotenoids) in different sorghum genotypes [25].

The synthesis of total flavonoids and monomers in the leaves of *Robinia pseudoacacia* L. seedlings is positively affected by elevated carbon dioxide, especially for older plants [26]. Elevated CO₂ concentration can cause higher chlorogenic acid accumulation in *Lactuca sativa* L. plants [27], while in barley and maize, high CO₂ decreases the total flavonoids and anthocyanins [28]. The summarized data on the effects of elevated carbon dioxide regarding different plant characteristics are presented in Table 1.

The plants from the *Brassicaceae* family have economic and agricultural importance as they represent an important dietary source of glucosinolates, vitamins, polyphenols, and minerals. The data regarding the influence of elevated carbon dioxide on *Brassicaceae* plants' characteristics are scarce. An early study showed that two *Brassica rapa* cultivars grows at 550 ppmv CO₂ showed increased crop productivity, even with the high-end soil nitrogen [34]. An increase in chlorophyll content of 33% has been found for pak choi (*Brassica rapa* ssp. *chinensis*) plants grown at 800 ppmv carbon dioxide compared to the control [24]. In *Brassica rapa* plants that grew at 700 ppmv carbon dioxide, the higher phenolic compounds increased the resistance to herbivore stress [35].

To evaluate the impact of two elevated carbon dioxide concentrations on the different plants, we grew plants at 800 and 1200 ppmv, respectively. Those concentrations could be achieved in the ambient atmosphere in the year 2100, according to the IPCC models [1]. To the authors' best knowledge, this is the first study that determines the plants' response regarding secondary and primary metabolites for plants grown at a higher than 1000 ppmv carbon dioxide concentration. This study also aims to determine the photosynthesis parameters, pigments, and secondary metabolites of 13 different *Brassicaceae* species and/or varieties to characterize plant signaling at elevated carbon dioxide.

Table 1. Examples of the effect of increased CO₂ concentrations on the photosynthetic characteristics, secondary metabolites, and pigment content of different plant species.

Plant Characteristic	Effect *	Species	Elevated Carbon Dioxide Concentration (ppmv)	Reference
Assimilation rate	↑	<i>Trifolium repens</i>	600	[9]
	↑	<i>Triticum aestivum</i>	583	[7]
	↑	<i>Oryza sativa</i>	475–600	[29]
	↑	<i>Glycine max</i>	475–600	[29]
	↑	<i>Carthamus tinctorius</i>	1000	[30]
	↑	<i>Phaseolus vulgaris</i> L.	700	[31]
	↑	<i>Populus × euroamericana</i>	550	[32]
	↑	<i>Camellia sinensis</i>	770	[17]
	↑	<i>Oryza sativa</i> L.	570	[20]
Isoprene emission	↓	<i>Phragmites australis</i>	800	[14]
	↓	<i>Platanus × acerfolia</i>	800	[14]
	↓	<i>Populus nigra × maximowiczii</i> NM6	800	[14]
	↓	<i>Populus × euroamericana</i>	550	[32]
	↔	<i>Populus tremula × Populus tremuloides</i>	780	[33]
Total phenols	↑	<i>Camellia sinensis</i>	770	[17]
	↑	<i>Cicer arietinum</i>	750	[18]
	↑	<i>Baccharis dracunculifolia</i>	750–800	[19]
	↑	<i>Baccharis platypoda</i>	750–800	[19]
Chlorophylls concentration	↓	<i>Oryza sativa</i> L.	570	[20]
	↓	<i>Solanum lycopersicum</i>	1000	[21]
	↑	<i>Cicer arietinum</i>	700	[23]
	↑	<i>Brassica rapa</i>	800	[24]
	↑	<i>Sorghum bicolor</i>	700	[25]
	↔	<i>Carthamus tinctorius</i>	1000	[30]
Total flavonoids	↑	<i>Robinia pseudoacacia</i> L.	750	[26]
	↓	<i>Hordeum vulgare</i>	620	[28]
	↓	<i>Zea maize</i>	620	[28]

* Effect: unchanged: ↔; increase: ↑; decrease: ↓.

2. Results

2.1. The Influence of Elevated Carbon Dioxide on Photosynthetic Parameters

The elevated carbon dioxide upregulated the assimilation rates for all *Brassicaceae* plants. The highest assimilation rates for plants grown at 1200 ppmv were found in Kale cabbage (*Brassica oleracea* var. *sabellica*) at a level of $36.66 \pm 0.46 \mu\text{mol m}^{-2} \text{s}^{-1}$, while the lowest assimilation rate was found for kohlrabi (*B. oleracea* var. *gongylodes*) (Figure 1a). Nonetheless, there are significant differences (one-way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p < 0.05$) between the assimilation rates for all plants species grown at 400, 800, and 1200 ppmv carbon dioxide, despite the differences in the trend. For example, the most negligible differences in assimilation rates for plants grown at 400 ppmv CO₂ compared with the one that was grown at 1200 ppmv CO₂ was for *B. oleracea* var. *cymose* ($14.01 \pm 0.19 \mu\text{mol m}^{-2} \text{s}^{-1}$ and $21.79 \pm 0.20 \mu\text{mol m}^{-2} \text{s}^{-1}$, respectively), while the greater values were found for *Lepidium sativum* ($6.69 \pm 0.21 \mu\text{mol m}^{-2} \text{s}^{-1}$ and $28.08 \pm 0.18 \mu\text{mol m}^{-2} \text{s}^{-1}$, respectively).

The stomatal conductance to water vapor values is not statistically different (one-way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p > 0.05$) for plants grown at 400 ppmv and 800 ppmv for all *Brassica oleracea* varieties (Figure 1b). At the same time, there are statistical differences for *Brassica napus*, *Sinapis alba*, and *Lepidium sativum*. In contrast, there are statistical differences (one-way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p < 0.05$) between the stomatal conductance to water vapor for plants grown at 1200 ppmv CO₂ and those grown at 400 and 800 ppmv CO₂. The highest stomatal conductance to water vapor was found for red cabbage ($116 \pm 5 \text{mmol m}^{-2} \text{s}^{-1}$),

which increased at 1200 ppmv carbon dioxide, while the smallest was registered for cress ($30 \pm 3 \text{ mmol m}^{-2} \text{ s}^{-1}$).

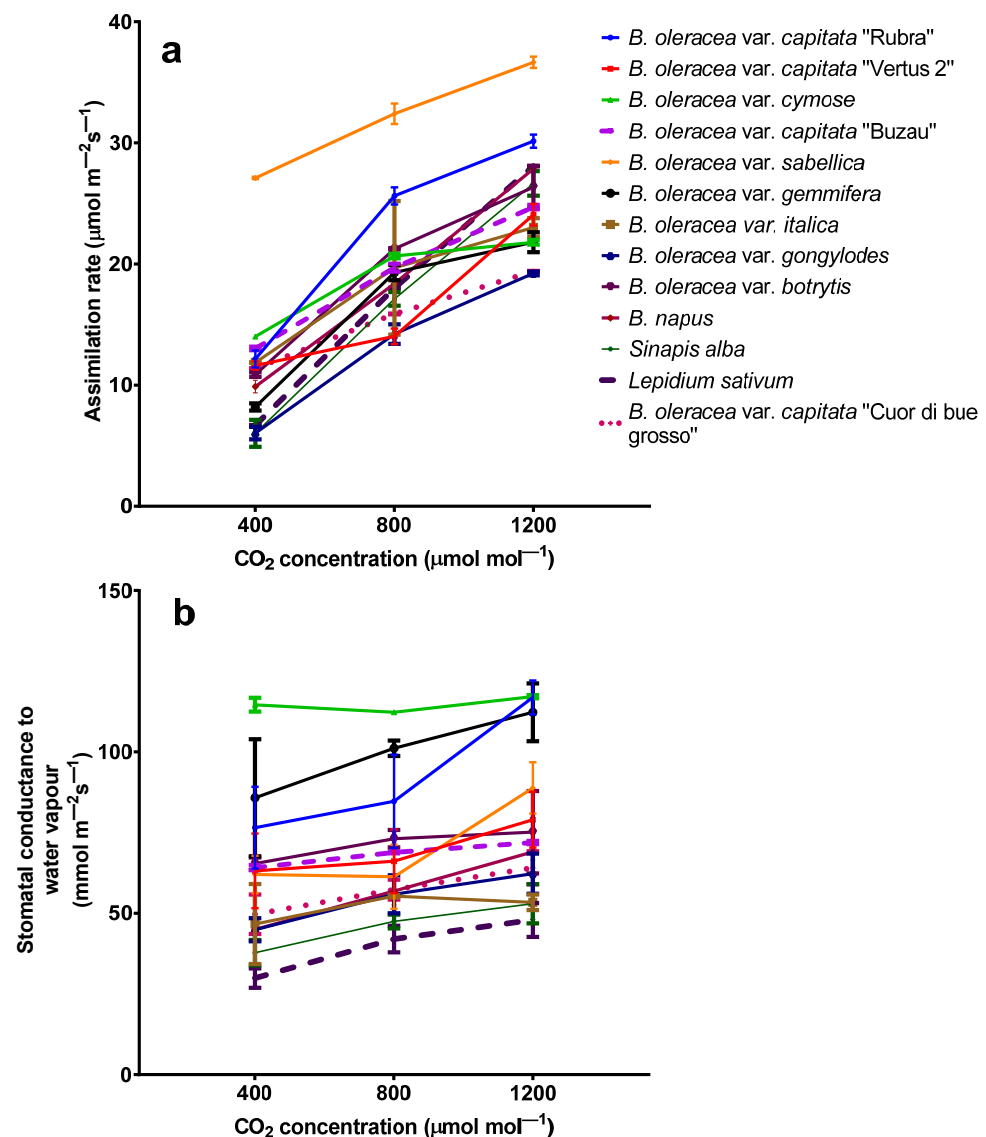


Figure 1. The assimilation rate (a) and stomatal conductance to water vapor (b) from *Brassicaceae* plants grown at three carbon dioxide concentrations. The values are averages of three independent measurements.

2.2. The Emission of Volatile Organic Compounds from Plants under Elevated Carbon Dioxide

Among the volatiles, six monoterpenes (camphene, β -pinene, 3-carene, D-limonene, *para*-cymene, and γ -terpinene) were found in the emission of all 13 *Brassicaceae* plants. The total emission from all different species are pretty low but is increasing for plants grown at a high carbon dioxide concentration (Figure 2). The emission of terpenes was most abundant in cress, rapes, and white mustard, whereas the lowest levels were found in red and white cabbages.

There is only one plant variety (*B. oleracea* var. capitata "Vertus 2") with no significant emission differences between all three growing conditions (one-way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p > 1$). In contrast, in all other 12 *Brassicaceae* varieties, there is an increase in monoterpene emission, at least for plants that grow at 1200 ppmv carbon dioxide. On the other hand, for kale, broccoli, and cabbage ("Cuor di bue grosso") the emission is not statistically different between plants that grow at 400 ppmv carbon dioxide and plants that grow at 800 ppmv carbon dioxide (one way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p < 0.05$).

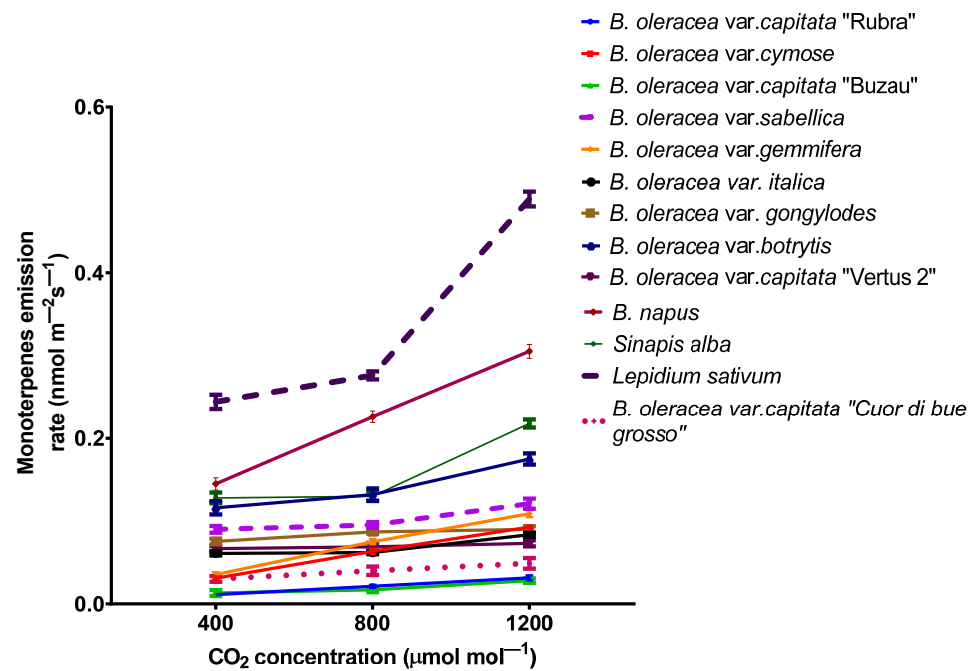


Figure 2. The emission rate of monoterpenes from *Brassicaceae* plants grown at three carbon dioxide concentrations. The values are averages of three independent measurements.

2.3. The Influence of Elevated Carbon Dioxide on Chlorophylls and β -Carotene

The chlorophyll *a* concentration in leaves of all 13 *Brassicaceae* varieties increase with the increase in carbon dioxide concentrations (Figure 3a). The medium chlorophyll *a* concentrations in the leaves of all 13 plants were 173 ± 63 mg m⁻² for plants grown at 400 ppmv CO₂, 231 ± 56 mg m⁻² for plants grown at 800 ppmv CO₂, and 283 ± 21 mg m⁻² for plants grown at 1200 ppmv CO₂, respectively. The chlorophyll *a* concentration for plants grown at 400 ppmv CO₂ varies from 100 ± 21 mg m⁻² in *B. oleracea* var. *gongylodes* to 273 ± 19 mg m⁻² in *Lepidium sativum*, while from plants grown at 1200 ppmv CO₂ vary from 200 ± 20 mg m⁻² in *Sinapis alba* to 371 ± 21 mg m⁻² in *B. oleracea* var. *capitata* "Rubra".

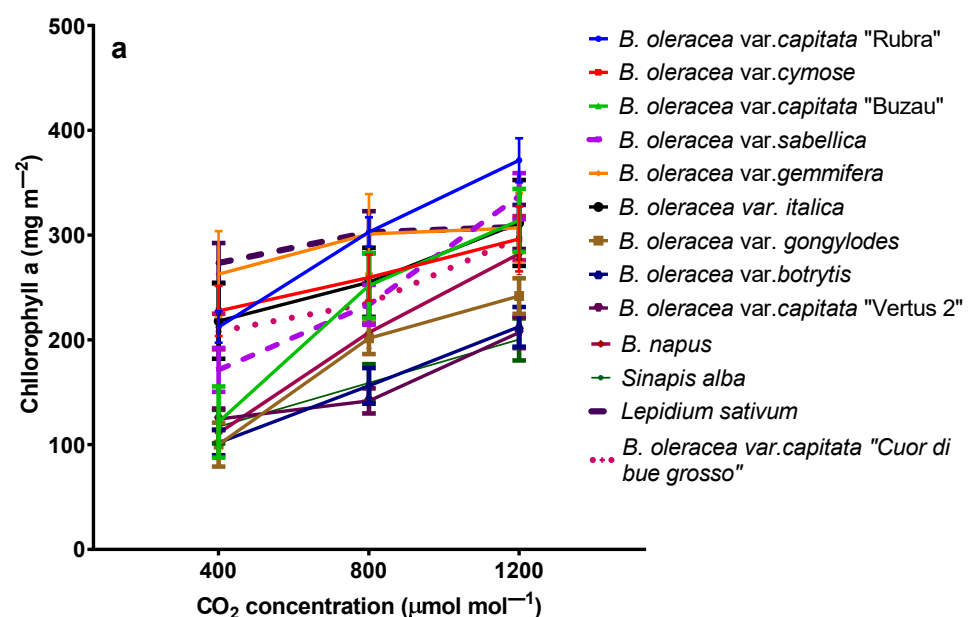


Figure 3. Cont.

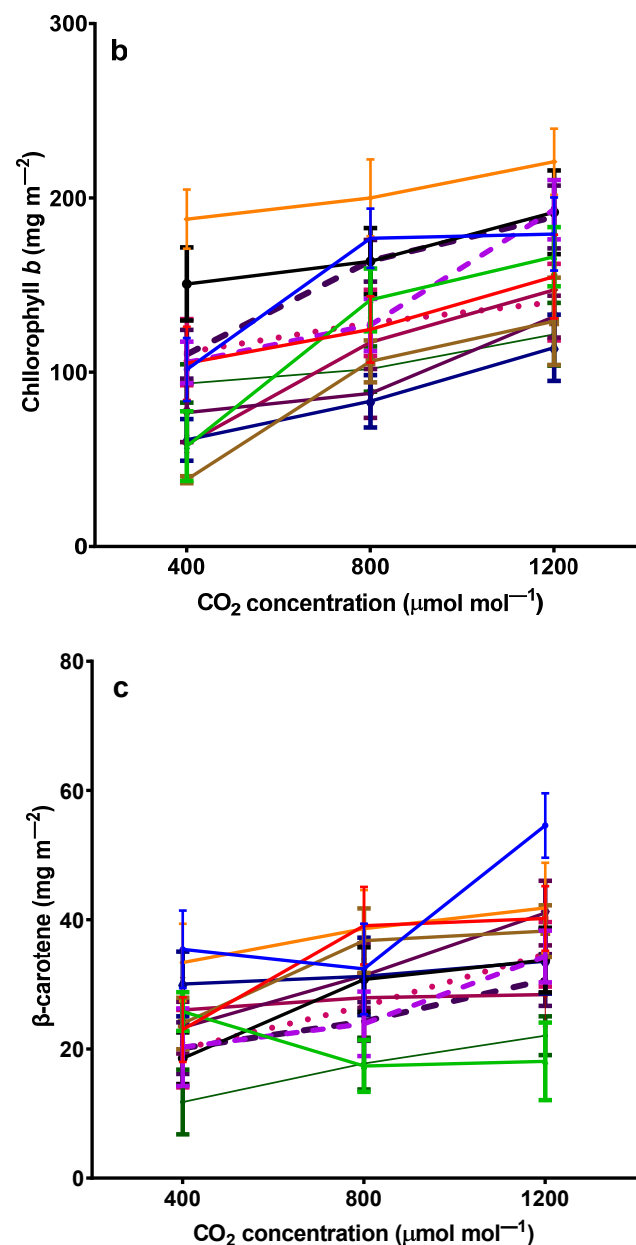


Figure 3. Chlorophyll *a* (a), Chlorophyll *b* (b) and β -carotene (c) concentrations from *Brassicaceae* plants grown at three carbon dioxide concentrations. The values are averages of three independent measurements.

The chlorophyll *b* concentrations generally increase for plants grown at high carbon dioxide concentrations, but it is not general for all varieties (Figure 3b). The medium chlorophyll *b* concentrations in the leaves of all 13 plants were $97 \pm 40 \text{ mg m}^{-2}$ for plants grown at 400 ppmv CO_2 , $132 \pm 35 \text{ mg m}^{-2}$ for plants grown at 800 ppmv CO_2 , and $160 \pm 33 \text{ mg m}^{-2}$ for plants grown at 1200 ppmv CO_2 , respectively.

The β -carotene concentration does not depend on the carbon dioxide growing conditions except for red cabbage (Figure 3c).

The medium β -carotene concentrations in the leaves of all 13 plants were $24 \pm 6 \text{ mg m}^{-2}$ for plants grown at 400 ppmv CO_2 , $29 \pm 7 \text{ mg m}^{-2}$ for plants grown at 800 ppmv CO_2 , and $35 \pm 9 \text{ mg m}^{-2}$ for plants grown at 1200 ppmv CO_2 , respectively. There are only statistical differences between plants grown at 1200 ppmv and 400 ppmv CO_2 (one-way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p < 0.01$).

2.4. The Change in Total Phenol Concentration for Plants Grown at Different Carbon Dioxide Concentration

Elevated carbon dioxide does not affect the total phenols concentration for some varieties but decreases for others (Figure 4). The most pronounced decrease in total phenols concentration was found in *Lepidium sativum* plants (227 ± 7 mg Eg gallic acid/L for plants grown at 400 ppmv compared with 145 ± 5 mg Eg ac gallic/L for plants grown at 1200 ppmv).

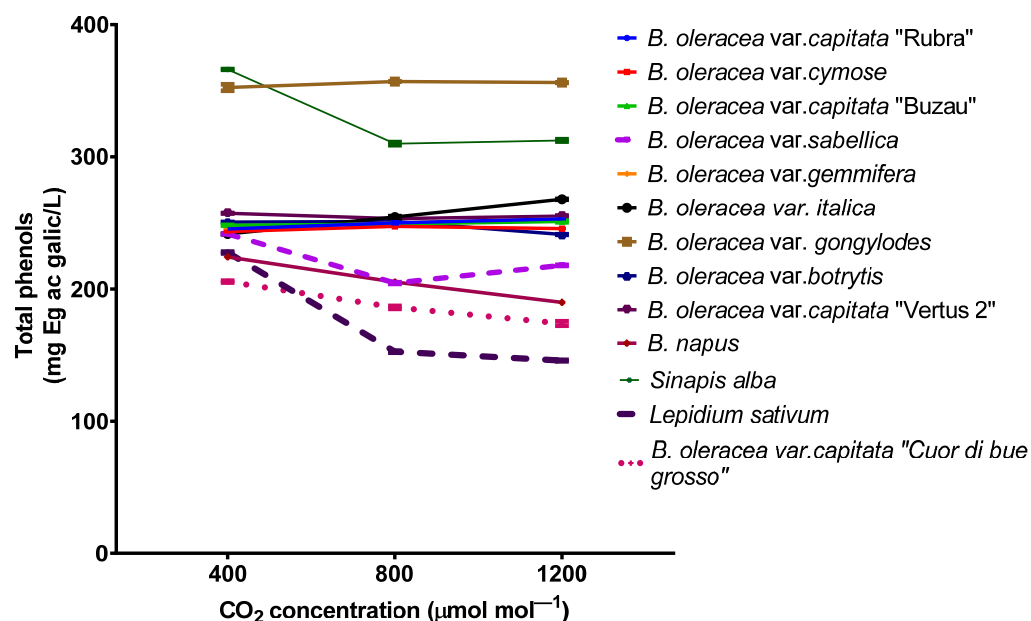


Figure 4. Total phenols concentration from *Brassicaceae* plants grown at three carbon dioxide concentrations. The values are averages of three independent measurements.

2.5. The Influence of Elevated Carbon Dioxide on Flavonoids Content in the Leaves of *Brassicaceae* Plants

The total flavonoid contents in leaves generally decrease for plants grown at elevated carbon dioxide, but there is no clear trend (Figure 5). There are some species (such as *Sinapis alba*) in which the concentration decrease significantly (one way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p < 0.001$) for plants grown at elevated carbon dioxide, while in others the concentration of flavonoids is not affected (as in *B. oleracea* var. *gemmifera*).

The medium total flavonoids concentrations in the leaves of all 13 plants were 0.054 ± 0.030 mg rutin equivalents/mL for plants grown at 400 ppmv CO₂, 0.043 ± 0.018 mg rutin equivalents/mL for plants grown at 800 ppmv CO₂, and 0.041 ± 0.0016 mg/mL for plants grown at 1200 ppmv CO₂, respectively. There are no statistical differences between plants grown at different CO₂ concentrations (one-way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p > 0.1$).

2.6. Microscopic Analyses

Stomatal characteristics did not differ between plants from different treatment groups (Figure 6). The stomata length among the three treatments are 26.37 ± 3.09 μm for 400 ppmv CO₂, 24.35 ± 1.84 μm for 800 ppmv CO₂, and 26.73 ± 1.71 μm for 1200 ppmv CO₂, which are not statistically different among treatments (one way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p = 0.7356$).

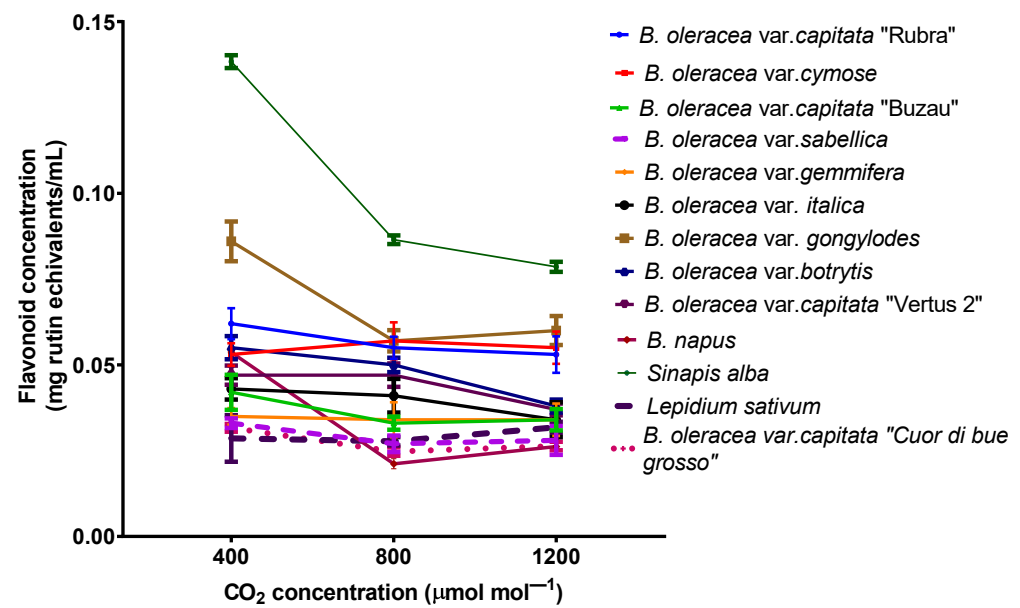


Figure 5. Total flavonoids content from *Brassicaceae* plants grown at three carbon dioxide concentrations. The values are averages of three independent measurements.

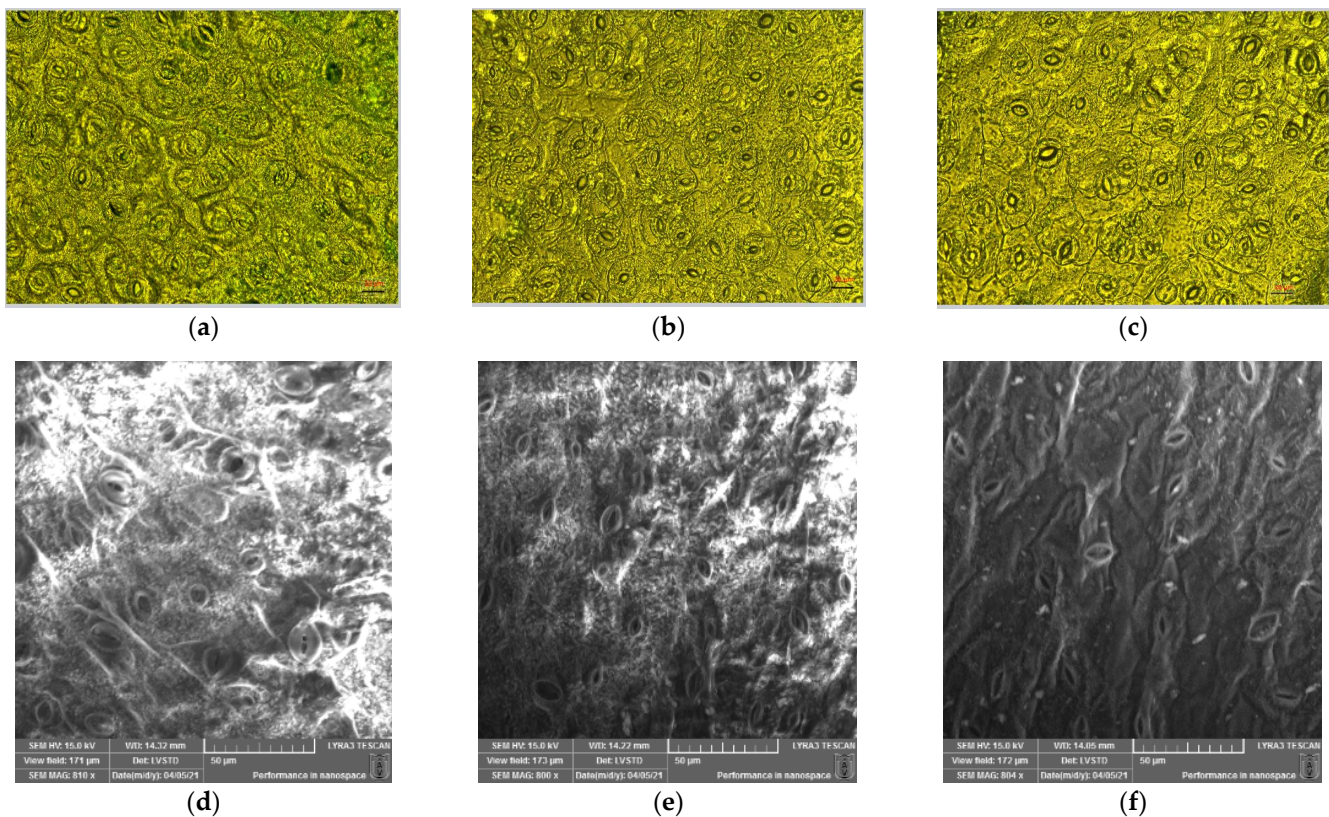


Figure 6. Microscopy images that present the stomatal pattern of leaf surfaces of *Brassica oleracea italic* recorded with an optical microscope ((a–c), with the scale bar 20 μm) and SEM micrographs ((d–f), magnification 800×) for leaf plants grown at 400 (a,d), 800 (b,e), and 1200 (c,f) ppmv CO₂.

The pore apertures of plants grown at lower concentrations of CO₂ remained larger than those of other plant groups. Stoma pore length slightly decreased from $14.65 \pm 2.88 \mu\text{m}$ at 400 ppmv CO₂ to $13.57 \pm 1.93 \mu\text{m}$ at 800 ppmv CO₂, but only became statistically differ-

ent at 1200 ppmv CO₂ ($8.01 \pm 0.88 \mu\text{m}$, one way ANOVA followed by Tukey's multiple comparisons *post hoc* test, $p < 0.05$).

3. Discussion

As expected, assimilation rates increased with the carbon dioxide concentrations for all species. The same results have been obtained from extensive experiments (FACE) in which legumes showed a 21% increase in saturated assimilation rates with growth at elevated CO₂ (see [29] for a review). Carbon dioxide is collected in the substomatal cavities, which could determine the carbon dioxide fixation due to the reaction with RuBP in the presence of the RuBisCo enzyme [36]. Moreover, it has been shown that increasing CO₂ in all species determines the decreasing percentage of leaf nitrogen allocated to RuBisCo, which suggests the acclimation of photosynthesis to elevated carbon dioxide [37]. Indeed, many studies have shown that elevated carbon dioxide increases the assimilation rates for C3 plants but decreases their nitrogen content, which is essential for vegetables [38,39].

Generally, under elevated carbon dioxide, stomata close due to higher depolarization of the guard cells [40,41]. The stomata closer to elevated carbon dioxide are induced by enhancing anion channel activity in guard cells [42]. Despite other experiments (in which stomatal conductance decreased on average by 20% in high carbon dioxide [43]), the stomatal conductance to water vapor is not affected by mildly increasing the carbon dioxide concentration. Such behavior has been found for *Cajanus cajan* L. during vegetative and reproductive growth phases [44] and could be due to altered guard cell signaling patterns. In *Arabidopsis* mutants with impaired Ca²⁺ priming sensors and HT1 protein kinase, the increase in carbon dioxide provoking stomatal conductance increased [45]. In our experiment, the stomatal conductance increases significantly only for plants grown at the highest carbon dioxide concentrations (1200 ppmv), which agrees with the results from previous papers [44,45].

The emission of terpenes from plants could be done from specialized secretory organs such as resin ducts, glandular trichomes, oil cavities, or *de novo* biosynthesis [46,47]. The monoterpenes emitted from plants in the atmosphere participates in secondary aerosol formations and could be implicated in different photochemical reactions. Generally, the emission of terpenes is decreasing for plants grown at elevated carbon dioxide concentrations [48]. Contradictory results have been found for the emission of volatiles from *Brassicaceae*. On the one hand, it has been shown that the emission of terpenes from *Brassica oleracea* ssp. *capitata* decrease for plants grown at high carbon dioxide [49] and, on the other hand, the emission of different volatile organic compounds from *Brassica napus* ssp. *oleifera* increase for plants grown at elevated carbon dioxide [50]. In our experiment, the monoterpene emission increased significantly (as in *Lepidium sativum*). In contrast, elevated carbon dioxide does not affect the emission for other species, as in the case of *B. oleracea* var. *sabellica*. Such data suggest that the monoterpene emission capacity of plants grown at elevated carbon dioxide is not affected by carbon accumulation in the leaf tissues [51]. As one of the most abundant terpenes in the emission blend for all species was limonene, the total enhancement in the emission at high carbon dioxide could be explained by increased activity of limonene synthase [52]. On the other hand, it has been demonstrated that isoprene emission decreases under high carbon dioxide due to either the stimulation of phosphoenolpyruvate carboxylase (PEPC), which competes for the pyruvate required for the MEP pathway, or the reduction in DMADP production [14]. However, in our experiment, the chlorophyll content at elevated carbon dioxide increases, suggesting that the N content in the leaves is not affected [53]. Indeed, the enhancement in soil-nitrogen supply affects the energy cycling between the reaction center and the chlorophyll pool, determining the chlorophyll increase. In contrast, the total phenolic compounds decrease for some species grown at elevated carbon dioxide due to the downregulation of the key enzyme PAL activity on the phenylpropanoid pathway [54]. Nonetheless, for most species, the total phenols are not significantly modified. The flavonoid contents decrease at elevated carbon dioxide for all species, probably due to a downregulation of leaf antioxidant

enzymes under elevated carbon dioxide. The same trend has been found in the seedling of *Oryza sativa* L., while for mature plants, the total phenols and flavonoids concentration increase [55].

The hierarchical model of ANOVA analysis between treatment (different carbon dioxide concentrations) and species/varieties confirms the statistical significance of the differences between species and varieties within treatment (Table 2).

Table 2. The hierarchical model of ANOVA analysis between treatment (different carbon dioxide concentrations) and species/varieties.

Source of Variation	Assimilation Rate	Stomata Conductance	Monoterpene Emission	Chlorophyll <i>a</i>	Chlorophyll <i>b</i>	β -carotene	Flavonoids	Polyphenols
	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value	df, MS, F, <i>p</i> Value
Carbon dioxide	2, 3182, 2849, <0.0001	2, 4907, 97.81, <0.0001	2, 0.0406, 1671, <0.0001	2, 119,198, 214.2, <0.0001	2, 39,098, 130.2, <0.0001	2, 1127, 44.26, <0.0001	2, 0.00333, 265.4, <0.0001	2, 2573, 5178, <0.0001
Species/varieties	12, 313.2, 280.4, <0.0001	12, 7206, 143.6, <0.0001	12, 0.0724, 2979, <0.0001	12, 25,178, 45.24, <0.0001	12, 10,211, 34.01, <0.0001	12, 389.9, 15.31, <0.0001	12, 0.00629, 501.0, <0.0001	12, 22,126, 44,530, <0.0001
Error	174.2	7827	0.001895	43,406	23,416	1986	0.001958	38.76

Some structural changes could be seen for plants growing at high carbon dioxide. Such a modification in mitochondria and chloroplast has been shown for plants from different families and could be due to increased cellular energy demands when plants are grown at elevated CO₂ [56].

4. Materials and Methods

4.1. Plant Material

The *Brassicaceae* (*Cruciferae* or mustard) family includes many species distributed worldwide (except Antarctica) and encloses approximately 338 genera and 3709 species.

Plants of 13 species from the *Brassicaceae* family were grown from the seeds as follows: Red cabbage (*Brassica oleracea* var. *capitata*, Langedijker Herfst (Sem-Luca, Timisoara, Romania)), Broccoli (*Brassica oleracea* var. *cymose*, Calabrese (Agrosel, Campia-Turzii, Romania)), Green Cabbage (*Brassica oleracea* var. *capitata*, Varza de buzau (Sem-Luca, Timisoara, Romania)), Vertus 2, (Legutko, Jutrosin, Poland), Cuor de Bue Grosso, (Legutko, Jutrosin, Poland)), Kale (*Brassica oleracea* var. *sabellica*, Black magic (Sem-Luca, Timisoara, Romania)), Broccoli (*Brassica oleracea* var. *italica*, Early Purple (Legutko, Jutrosin, Poland)), Brussels sprout (*Brassica oleracea* var. *gemmifera*, Groninger (Sem-Luca, Timisoara, Romania)), Kohlrabi (*Brassica oleracea* var. *gongyloides*, Gongylodes (Agrosel, Campia-Turzii, Romania)), Cauliflower (*Brassica oleracea* var. *botrytis*, Moldovita F1 (Agrosel, Campia-Turzii, Romania)), Cress (*Lepidium sativum*, Common (Legutko, Jutrosin, Poland)), Rapeseed (*Brassica napus* subsp. *napus*, Pioneer PT275, Pioneer Hi-Bred, România), and White Mustard (*Sinapis alba* L., Franchi Sementini, Bergamo, Italy).

The seeds were sown in 0.8 L plastic pots filled with a mixture of commercial garden soil and quartz sand. The plants have been fertilized with fertilizers for foliar (Bionat Plus, Panetone SRL, Timisoara, Romania) and radicular (Cropcare 11-11-21, YaraMila, Oslo, Norway). Day length was 12 h, and the light intensity at plant level of 800 $\mu\text{mol m}^{-2} \text{s}^{-1}$ was provided by led lamps (Hoff, Nürnberg, Germany). Day/night temperatures were maintained at 25/22 °C and a relative humidity of 65%. The plants were watered every day to soil field capacity. Seven-week-old non-bolted plants with at least three fully developed leaves were used in the experiments. We used the fully expanded leaves with the same development stage for all measurements. The plants were randomized to ensure that all plants grow in the same light. The leaves used for photosynthetic and volatile organic compounds measurements were used for the biochemical analysis.

4.2. Photosynthetic Measurements

A portable gas exchange system (GFS-3000, Waltz, Effeltrich, Germany) was used to determine the photosynthetic parameters, as reported earlier in [57,58]. The calculation of

the steady-state values of net assimilation (A) and stomatal conductance to water vapor (g_s) was performed as was depicted in [58].

4.3. Volatile Sampling and GC–MS Analyses

Volatile organic compounds (VOC) were sampled (by a flow air sample pump 210-1003 MTX (SKC Inc., Houston, TX, USA)) and analyzed (by a Shimadzu TD20 automated cartridge desorber coupled with a Shimadzu 2010 Plus GC–MS equipment (Shimadzu Corporation, Kyoto, Japan)), as earlier reported in [59].

4.4. Chromatographic Analysis of Photosynthetic Pigments

The pigments (chlorophyll a, chlorophyll b, and β -carotene) were extracted in acetone as described before [60], and the quantitative analyses were performed using the UHPLC-DAD apparatus (NEXERA 8030, Shimadzu, Kyoto, Japan) following the same method earlier published [57]. The concentration of chlorophyll a, chlorophyll b, and β -carotene was calculated using the pure chromatographic standards (Merck, Darmstadt, Germany).

4.5. Flavonoid Content Analysis

The total flavonoid content was determined using the spectrophotometric method [59]. A reaction mixture of aluminum chloride, sodium acetate, and sample was measured at 434 nm, and the results were expressed in mg rutin equivalents/mL.

4.6. Total Phenolic Content—Folin–Ciocalteu Method

Total phenolic content was determined according to the Folin–Ciocalteu, method as described in [59,61], and the results were expressed in mg gallic acid equivalents/mL.

4.7. Microscopy Analyses

The surfaces of leaves were examined by using a Zeiss Scope.A1 microscope equipped with the AxioCam MRc 5 camera and ZEN lite 2012 software (Carl Zeiss MicroImaging GmbH, Jena, Germany). The samples preparation was done by following the next steps: peel the epidermis from the backside of the leaf, mount the sample on a glass slide in distilled water, fix with a coverslip, and observe under the microscope at $40\times$ magnification. For scanning electron microscopy (SEM), leaves were mounted on stubs using carbon double-sided adhesive tape without any treatment. The samples were examined and photographed using LYRA3 scanning electron microscope (LYRA3 XMU, Tescan, Brno, Czech Republic) with Low Vacuum Secondary Electron Tescan Detector (LVSTD), at 15 kV and magnification $800\times$.

4.8. Statistical Analysis and Data Handling

One-way ANOVA, Tukey's multiple comparisons test, and two-way ANOVA were done using GraphPad Prism version 9.3.0 for Windows (GraphPad Software, San Diego, Ca, USA, www.graphpad.com (accessed on 30 January 2022)). Results were considered significantly different at p -values < 0.05 .

5. Conclusions

In this study, we have shown that elevated carbon dioxide increases the photosynthetic activities and emission of volatile organic compounds. On the other hand, plants that grow at a high concentration of CO_2 exhibit downregulation of polyphenols and flavonoids, which could become a significant problem in light of future climate change conditions. The results revealed that different species/varieties from the *Brassicaceae* family respond differently to increases in carbon dioxide concentration. Generally, all plants increase their assimilation rates, monoterpene emission, and chlorophylls and decrease their flavonoids and polyphenols content.

Author Contributions: Conceptualization, L.C.; methodology, A.L., C.M., S.G., M.D., D.C., V.C. and D.M.C.; software, A.L., C.M., M.D. and D.M.C.; validation, A.L., C.M., D.C., D.M.C. and L.C.; formal analysis, A.L., C.M., S.G., M.D., D.C., V.C. and D.M.C.; investigation, A.L., C.M., S.G., M.D., D.C., V.C. and D.M.C.; data curation, A.L., C.M., M.D. and D.M.C.; writing—original draft preparation, A.L., D.M.C. and L.C.; writing—review and editing, A.L., D.M.C. and L.C.; supervision, L.C.; project administration, L.C.; funding acquisition, L.C. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by CNFIS-UEFISCDI, project number PN-III-P4-ID-PCE-2020-0410.

Data Availability Statement: The data presented in this study are available in the article.

Acknowledgments: The authors thank Brenda Crystal Svinti for the professional editing of this paper.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. IPCC. *Climate Change 2014: Impacts, Adaptation, and Vulnerability. Part A: Global and Sectoral Aspects*; Cambridge University Press: Cambridge, UK; New York, NY, USA, 2014; p. 1132.
2. Ishigooka, Y.; Hasegawa, T.; Kuwagata, T.; Nishimori, M.; Wakatsuki, H. Revision of estimates of climate change impacts on rice yield and quality in Japan by considering the combined effects of temperature and CO₂ concentration. *J. Agric. Meteorol.* **2021**, *77*, 139–149. [\[CrossRef\]](#)
3. Reyer, C.P.O.; Leuzinger, S.; Rammig, A.; Wolf, A.; Bartholomeus, R.P.; Bonfante, A.; de Lorenzi, F.; Dury, M.; Gloning, P.; Abou Jaoudé, R.; et al. A plant's perspective of extremes: Terrestrial plant responses to changing climatic variability. *Glob. Change Biol.* **2013**, *19*, 75–89. [\[CrossRef\]](#)
4. Medlyn, B.E.; Barton, C.V.M.; Broadmeadow, M.S.J.; Ceulemans, R.; De Angelis, P.; Forstreuter, M.; Freeman, M.; Jackson, S.B.; Kellomäki, S.; Laitat, E.; et al. Stomatal conductance of forest species after long-term exposure to elevated CO₂ concentration: A synthesis. *New Phytol.* **2001**, *149*, 247–264. [\[CrossRef\]](#)
5. Drake, B.G.; González-Meler, M.A.; Long, S.P. More efficient plants: A Consequence of Rising Atmospheric CO₂? *Annu. Rev. Plant Biol.* **1997**, *48*, 609–639. [\[CrossRef\]](#) [\[PubMed\]](#)
6. Cernusak, L.A.; Haverd, V.; Brendel, O.; Le Thiec, D.; Guehl, J.M.; Cuntz, M. Robust response of terrestrial plants to rising CO₂. *Trends Plant Sci.* **2019**, *24*, 578–586. [\[CrossRef\]](#) [\[PubMed\]](#)
7. Ainsworth, E.A.; Leakey, A.D.B.; Ort, D.R.; Long, S.P. FACE-ing the facts: Inconsistencies and interdependence among field, chamber and modeling studies of elevated [CO₂] impacts on crop yield and food supply. *New Phytol.* **2008**, *179*, 5–9. [\[CrossRef\]](#) [\[PubMed\]](#)
8. Wang, D.; Heckathorn, S.A.; Wang, X.; Philpott, S.M. A meta-analysis of plant physiological and growth responses to temperature and elevated CO₂. *Oecologia* **2012**, *169*, 1–13. [\[CrossRef\]](#)
9. Ainsworth, E.A.; Rogers, A.; Blum, H.; Nösberger, J.; Long, S.P. Variation in acclimation of photosynthesis in *Trifolium repens* after eight years of exposure to Free Air CO₂ Enrichment (FACE). *J. Exp. Bot.* **2003**, *54*, 2769–2774. [\[CrossRef\]](#)
10. Rogers, A.; Humphries, S.W. A mechanistic evaluation of photosynthetic acclimation at elevated CO₂. *Glob. Change Biol.* **2000**, *6*, 1005–1011. [\[CrossRef\]](#)
11. Peña-Rojas, K.; Aranda, X.; Fleck, I. Stomatal limitation to CO₂ assimilation and down-regulation of photosynthesis in *Quercus ilex* resprouts in response to slowly imposed drought. *Tree Physiol.* **2004**, *24*, 813–822. [\[CrossRef\]](#)
12. Zheng, Y.; Li, F.; Hao, L.; Yu, J.; Guo, L.; Zhou, H.; Ma, C.; Zhang, X.; Xu, M. Elevated CO₂ concentration induces photosynthetic down-regulation with changes in leaf structure, non-structural carbohydrates and nitrogen content of soybean. *BMC Plant Biol.* **2019**, *19*, 255. [\[CrossRef\]](#)
13. Copolovici, L.; Popitanu, A.C.; Copolovici, D.-M. Volatile organic compound emission and residual substances from plants in light of the globally increasing CO₂ level. *Curr. Opin. Environ. Sci. Health* **2021**, *19*, 100216. [\[CrossRef\]](#)
14. Lantz, A.T.; Solomon, C.; Gog, L.; McClain, A.M.; Weraduwege, S.M.; Cruz, J.A.; Sharkey, T.D. Isoprene suppression by CO₂ is not due to triose phosphate utilization (TPU) limitation. *Front. For. Glob. Change* **2019**, *2*, 8. [\[CrossRef\]](#)
15. Chen, Y.; Martin, C.; Fingu Mabola, J.C.; Verheggen, F.; Wang, Z.; He, K.; Francis, F. Effects of host plants reared under elevated CO₂ concentrations on the foraging behavior of different stages of corn leaf aphids *Rhopalosiphum maidis*. *Insects* **2019**, *10*, 182. [\[CrossRef\]](#) [\[PubMed\]](#)
16. Daussy, J.; Staudt, M. Do future climate conditions change volatile organic compound emissions from *Artemisia annua*? Elevated CO₂ and temperature modulate actual VOC emission rate but not its emission capacity. *Atmos. Environ. X* **2020**, *7*, 100082. [\[CrossRef\]](#)
17. Li, L.; Wang, M.; Pokharel, S.S.; Li, C.; Parajulee, M.N.; Chen, F.; Fang, W. Effects of elevated CO₂ on foliar soluble nutrients and functional components of tea, and population dynamics of tea aphid, *Toxoptera aurantii*. *Plant Physiol. Biochem.* **2019**, *145*, 84–94. [\[CrossRef\]](#)
18. Sharma, H.C.; War, A.R.; Pathania, M.; Sharma, S.P.; Akbar, S.M.; Munghate, R.S. Elevated CO₂ influences host plant defense response in chickpea against *Helicoverpa armigera*. *Arthropod-Plant Interact.* **2016**, *10*, 171–181. [\[CrossRef\]](#)

19. Oki, Y.; Arantes-Garcia, L.; Costa, M.; Nunes, B.; Rúbia, B.; Gélvez-Zúñiga, I.; Franco, A.; Fernandes, G. CO₂ fertilizer effect on growth, polyphenols, and endophytes in two *Baccharis* species. *Braz. Arch. Biol. Technol.* **2020**, *63*, e20190302. [\[CrossRef\]](#)
20. Moynul Haque, M.; Hamid, A.; Khanam, M.; Biswas, D.K.; Karim, M.A.; Khaliq, Q.A.; Hossain, M.A.; Upreti, D.C. The effect of elevated CO₂ concentration on leaf chlorophyll and nitrogen contents in rice during post-flowering phases. *Biol. Plant.* **2006**, *50*, 69–73. [\[CrossRef\]](#)
21. Lanoue, J.; Leonardos, E.D.; Khosla, S.; Hao, X.; Grodzinski, B. Effect of elevated CO₂ and spectral quality on whole plant gas exchange patterns in tomatoes. *PLoS ONE* **2018**, *13*, e0205861. [\[CrossRef\]](#)
22. Huang, M.-Y.; Wong, S.-L.; Weng, J.-H. Rapid light-response curve of chlorophyll fluorescence in terrestrial plants: Relationship to CO₂ exchange among five woody and four fern species adapted to different light and water regimes. *Plants* **2021**, *10*, 445. [\[CrossRef\]](#) [\[PubMed\]](#)
23. Bhatia, A.; Mina, U.; Kumar, V.; Tomer, R.; Kumar, A.; Chakrabarti, B.; Singh, R.N.; Singh, B. Effect of elevated ozone and carbon dioxide interaction on growth, yield, nutrient content and wilt disease severity in chickpea grown in Northern India. *Heliyon* **2021**, *7*, e06049. [\[CrossRef\]](#) [\[PubMed\]](#)
24. Hou, L.; Shang, M.; Chen, Y.; Zhang, J.; Xu, X.; Song, H.; Zheng, S.; Li, M.; Xing, G. Physiological and molecular mechanisms of elevated CO₂ in promoting the growth of pak choy (*Brassica rapa* ssp. *chinensis*). *Sci. Hortic.* **2021**, *288*, 110318. [\[CrossRef\]](#)
25. Keramat, S.; Eshghizadeh, H.R.; Zahedi, M.; Nematpour, A. Growth and biochemical changes of sorghum genotypes in response to carbon dioxide and salinity interactions. *Cereal Res. Commun.* **2020**, *48*, 325–332. [\[CrossRef\]](#)
26. Zhang, C.; Jia, X.; Zhao, Y.; Wang, L.; Cao, K.; Zhang, N.; Gao, Y.; Wang, Z. The combined effects of elevated atmospheric CO₂ and cadmium exposure on flavonoids in the leaves of *Robinia pseudoacacia* L. seedlings. *Ecotoxicol. Environ. Saf.* **2021**, *210*, 111878. [\[CrossRef\]](#)
27. Shimomura, M.; Yoshida, H.; Fujiuchi, N.; Ariizumi, T.; Ezura, H.; Fukuda, N. Continuous blue lighting and elevated carbon dioxide concentration rapidly increase chlorogenic acid content in young lettuce plants. *Sci. Hortic.* **2020**, *272*, 109550. [\[CrossRef\]](#)
28. Selim, S.; Abuelsoud, W.; Al-Sanea, M.M.; AbdElgawad, H. Elevated CO₂ differently suppresses the arsenic oxide nanoparticles-induced stress in C3 (*Hordeum vulgare*) and C4 (*Zea mays*) plants via altered homeostasis in metabolites specifically proline and anthocyanin metabolism. *Plant Physiol. Biochem.* **2021**, *166*, 235–245. [\[CrossRef\]](#)
29. Ainsworth, E.A.; Long, S.P. What have we learned from 15 years of free-air CO₂ enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO₂. *New Phytol.* **2005**, *165*, 351–372. [\[CrossRef\]](#)
30. Jalal, S.; Jellings, A.; Fuller, M. Positive effects of elevated CO₂ and its interaction with nitrogen on safflower physiology and growth. *Agron. Sustain. Dev.* **2013**, *33*, 497–505. [\[CrossRef\]](#)
31. Radoglou, K.M.; Aphalo, P.; Jarvis, P.G. Response of photosynthesis, stomatal conductance and water use efficiency to elevated CO₂ and nutrient supply in acclimated seedlings of *Phaseolus vulgaris* L. *Ann. Bot.* **1992**, *70*, 257–264. [\[CrossRef\]](#)
32. Centritto, M.; Nascetti, P.; Petrilli, L.; Raschi, A.; Loreto, F. Profiles of isoprene emission and photosynthetic parameters in hybrid poplars exposed to free air CO₂ enrichment. *Plant Cell Environ.* **2004**, *27*, 403–412. [\[CrossRef\]](#)
33. Sun, Z.; Hüve, K.; Vislap, V.; Niinemets, Ü. Elevated CO₂ magnifies isoprene emissions under heat and improves thermal resistance in hybrid aspen. *J. Exp. Bot.* **2013**, *64*, 5509–5523. [\[CrossRef\]](#) [\[PubMed\]](#)
34. Thomson, G.; Mollah, M.R.; Partington, D.L.; Jones, R.; Argall, R.; Tregenza, J.; Fitzgerald, G.J. Effects of elevated carbon dioxide and soil nitrogen on growth of two leafy Brassica vegetables. *N. Z. J. Crop Hortic. Sci.* **2013**, *41*, 69–77. [\[CrossRef\]](#)
35. Karowe, D.N.; Grubb, C. Elevated CO₂ increases constitutive phenolics and trichomes, but decreases inducibility of phenolics in *Brassica rapa* (Brassicaceae). *J. Chem. Ecol.* **2011**, *37*, 1332–1340. [\[CrossRef\]](#)
36. Zhu, X.-G.; Long, S.P.; Ort, D.R. Improving photosynthetic efficiency for greater yield. *Annu. Rev. Plant Biol.* **2010**, *61*, 235–261. [\[CrossRef\]](#)
37. Hirose, T.; Ackerly, D.D.; Traw, M.B.; Bazzaz, F.A. Effects of CO₂ elevation on canopy development in the stands of two co-occurring annuals. *Oecologia* **1996**, *108*, 215–223. [\[CrossRef\]](#)
38. Baligar, V.C.; Elson, M.K.; Almeida, A.-A.F.; de Araujo, Q.R.; Ahnert, D.; He, Z. The Impact of carbon dioxide concentrations and low to adequate photosynthetic photon flux density on growth, physiology and nutrient use efficiency of juvenile cacao genotypes. *Agronomy* **2021**, *11*, 397. [\[CrossRef\]](#)
39. Dong, J.; Gruda, N.; Lam, S.K.; Li, X.; Duan, Z. Effects of elevated CO₂ on nutritional quality of vegetables: A review. *Front. Plant Sci.* **2018**, *9*, 924. [\[CrossRef\]](#)
40. Xu, Z.; Jiang, Y.; Jia, B.; Zhou, G. Elevated-CO₂ response of stomata and its dependence on environmental factors. *Front. Plant Sci.* **2016**, *7*, 657. [\[CrossRef\]](#)
41. Negi, J.; Hashimoto-Sugimoto, M.; Kusumi, K.; Iba, K. New approaches to the biology of stomatal guard cells. *Plant Cell Physiol.* **2013**, *55*, 241–250. [\[CrossRef\]](#)
42. Yamamoto, Y.; Negi, J.; Wang, C.; Isogai, Y.; Schroeder, J.I.; Iba, K. The transmembrane region of guard cell SLAC1 channels perceives CO₂ signals via an ABA-independent pathway in *Arabidopsis*. *Plant Cell* **2016**, *28*, 557–567. [\[CrossRef\]](#) [\[PubMed\]](#)
43. Wand, S.J.E.; Midgley, G.F.; Jones, M.H.; Curtis, P.S. Responses of wild C4 and C3 grass (Poaceae) species to elevated atmospheric CO₂ concentration: A meta-analytic test of current theories and perceptions. *Glob. Change Biol.* **1999**, *5*, 723–741. [\[CrossRef\]](#)
44. Sreeharsha, R.V.; Sekhar, K.M.; Reddy, A.R. Delayed flowering is associated with lack of photosynthetic acclimation in Pigeon pea (*Cajanus cajan* L.) grown under elevated CO₂. *Plant Sci.* **2015**, *231*, 82–93. [\[CrossRef\]](#)

45. Hashimoto, M.; Negi, J.; Young, J.; Israelsson, M.; Schroeder, J.I.; Iba, K. *Arabidopsis* HT1 kinase controls stomatal movements in response to CO₂. *Nat. Cell Biol.* **2006**, *8*, 391–397. [\[CrossRef\]](#)
46. Yang, W.; Cao, J.; Wu, Y.; Kong, F.; Li, L. Review on plant terpenoid emissions worldwide and in China. *Sci. Total Environ.* **2021**, *787*, 147454. [\[CrossRef\]](#)
47. Popitanu, C.; Lupitu, A.; Copolovici, L.; Bungău, S.; Niinemets, Ü.; Copolovici, D.M. Induced volatile emissions, photosynthetic characteristics, and pigment content in *Juglans regia* leaves infected with the Erineum-forming mite *Aceria erineae*. *Forests* **2021**, *12*, 920. [\[CrossRef\]](#)
48. Peñuelas, J.; Staudt, M. BVOCs and global change. *Trends Plant Sci.* **2010**, *15*, 133–144. [\[CrossRef\]](#)
49. Vuorinen, T.; Reddy, G.V.P.; Nerg, A.-M.; Holopainen, J.K. Monoterpene and herbivore-induced emissions from cabbage plants grown at elevated atmospheric CO₂ concentration. *Atmos. Environ.* **2004**, *38*, 675–682. [\[CrossRef\]](#)
50. Himanen, S.J.; Nerg, A.-M.; Nissinen, A.I.; Pinto, D.M.L.; Stewart, C.N.; Poppy, G.M.; Holopainen, J.K. Effects of elevated carbon dioxide and ozone on volatile terpenoid emissions and multitrophic communication of transgenic insecticidal oilseed rape (*Brassica napus*). *New Phytol.* **2009**, *181*, 174–186. [\[CrossRef\]](#)
51. Staudt, M.; Joffre, R.; Rambal, S.; Kesselmeier, J. Effect of elevated CO₂ on monoterpene emission of young *Quercus ilex* trees and its relation to structural and ecophysiological parameters. *Tree Physiol.* **2001**, *21*, 437–445. [\[CrossRef\]](#)
52. Loreto, F.; Fischbach, R.J.; Schnitzler, J.-P.; Ciccioli, P.; Brancaleoni, E.; Calfapietra, C.; Seufert, G. Monoterpene emission and monoterpene synthase activities in the Mediterranean evergreen oak *Quercus ilex* L. grown at elevated CO₂ concentrations. *Glob. Change Biol.* **2001**, *7*, 709–717. [\[CrossRef\]](#)
53. Uprety, D.C.; Mahalaxmi, V. Effect of elevated CO₂ and nitrogen nutrition on photosynthesis, growth and carbon-nitrogen balance in *Brassica juncea*. *J. Agron. Crop Sci.* **2000**, *184*, 271–276. [\[CrossRef\]](#)
54. Mattson, W.J.; Julkunen-Tiitto, R.; Herms, D.A. CO₂ enrichment and carbon partitioning to phenolics: Do plant responses accord better with the protein competition or the growth differentiation balance models? *Oikos* **2005**, *111*, 337–347. [\[CrossRef\]](#)
55. Goufo, P.; Pereira, J.; Moutinho-Pereira, J.; Correia, C.M.; Figueiredo, N.; Carranca, C.; Rosa, E.A.S.; Trindade, H. Rice (*Oryza sativa* L.) phenolic compounds under elevated carbon dioxide (CO₂) concentration. *Environ. Exp. Bot.* **2014**, *99*, 28–37. [\[CrossRef\]](#)
56. Griffin, K.L.; Anderson, O.R.; Gastrich, M.D.; Lewis, J.D.; Lin, G.; Schuster, W.; Seemann, J.R.; Tissue, D.T.; Turnbull, M.H.; Whitehead, D. Plant growth in elevated CO₂ alters mitochondrial number and chloroplast fine structure. *Proc. Natl. Acad. Sci. USA* **2001**, *98*, 2473–2478. [\[CrossRef\]](#)
57. Copolovici, L.; Pag, A.; Kännaste, A.; Bodescu, A.; Tomescu, D.; Copolovici, D.; Soran, M.-L.; Niinemets, Ü. Disproportionate photosynthetic decline and inverse relationship between constitutive and induced volatile emissions upon feeding of *Quercus robur* leaves by large larvae of gypsy moth (*Lymantria dispar*). *Environ. Exp. Bot.* **2017**, *138*, 184–192. [\[CrossRef\]](#)
58. Niinemets, Ü.; Copolovici, L.; Hüve, K. High within-canopy variation in isoprene emission potentials in temperate trees: Implications for predicting canopy-scale isoprene fluxes. *J. Geophys. Res.-Biogeosci.* **2010**, *115*, G04029. [\[CrossRef\]](#)
59. Copolovici, L.; Lupitu, A.; Moisa, C.; Taschina, M.; Copolovici, D.M. The effect of antagonist abiotic stress on bioactive compounds from basil (*Ocimum basilicum*). *Appl. Sci.* **2021**, *11*, 9282. [\[CrossRef\]](#)
60. Opreș, O.; Copaciu, F.; Soran, M.L.; Ristoiu, D.; Niinemets, Ü.; Copolovici, L. Influence of nine antibiotics on key secondary metabolites and physiological characteristics in *Triticum aestivum*: Leaf volatiles as a promising new tool to assess toxicity. *Ecotoxicol. Environ. Saf.* **2013**, *87*, 70–79. [\[CrossRef\]](#)
61. Moisa, C.; Copolovici, L.; Pop, G.; Imbrea, I.; Lupitu, A.; Nemeth, S.; Copolovici, D. Wastes resulting from aromatic plants distillation-bio-sources of antioxidants and phenolic compounds with biological active principles. *Farmacia* **2018**, *66*, 289–295.