













Physiological impact of pollinator exposure on plants: A comparative study of cucumber and mint

Alicja Auriga^{*1)}  , Piotr Dąbrowski²⁾  , Piotr Ptak³⁾  ,
Kamila Bojko⁴⁾  , Ana L. Mamede Leite⁵⁾  , Hazem M. Kalaji⁶⁾  

¹⁾ West Pomeranian University of Technology in Szczecin, Department of Animal Anatomy and Zoology,
al. Piastów 17, 70-310 Szczecin, Poland

²⁾ Warsaw University of Life Science, Department of Environmental Management and Remote Sensing,
Nowoursynowska St, 159, 02-776 Warsaw, Poland

³⁾ West Pomeranian University of Technology in Szczecin, Department of Bioengineering, al. Piastów 17, 70-310 Szczecin, Poland

⁴⁾ West Pomeranian University of Technology in Szczecin, Department of Horticulture, al. Piastów 17, 70-310 Szczecin, Poland

⁵⁾ São Paulo State University (UNESP), International School of Pharmaceutical Sciences,
Rodovia Araraquara - Jaú Km 1, 14800-903 Araraquara, SP, Brazil

⁶⁾ Warsaw University of Life Science, Institute of Biology, Department of Plant Physiology,
Nowoursynowska St, 159, 02-776 Warsaw, Poland

* Corresponding author

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Abstract: It is well known that insects can influence yield and harvest quality of plants by activating plant response mechanisms to their activity. Scientists have been increasingly exploring these mechanisms in recent years, however analysis of plant responses to pollinators still requires further work. A research method based on chlorophyll fluorescence and gas exchange allows for quick and precise determination of a plant's condition, including whether it is experiencing physiological stress. The aim of this study was to determine the extent to which interactions with pollinators affect the photosynthetic efficiency of plants. The research involved two different plant species: perennial – pepper mint (*Mentha* sp.) and annual – pickling cucumber (*Cucumis sativus* L.). Plants were grown in two variants: exposed and unexposed to pollinators. Gas exchange and chlorophyll fluorescence parameters, as well as pigments content were analysed. The results showed no significant differences in pigment content, gas exchange, or chlorophyll fluorescence in mint. Cucumber exhibited significant changes in chlorophyll and carotenoid content, as well as in stomatal conductance and fluorescence parameters. Plants unexposed to pollinators showed increased photosynthetic pigment levels and improved PSII stability, although it simultaneously reduced CO₂ assimilation intensity. It may suggest that perennial mint tolerates pollinator effects more effectively, while annual cucumber responds with dynamic adjustments in photosynthetic metabolism.

Keywords: chlorophyll fluorescence, gas exchange, photosynthesis performance, pigments content, plant-insect interaction, pollinators, stress indicators

INTRODUCTION

Photosynthesis is a most fundamental physiological and biochemical process on Earth. Thanks to that process autotrophic organisms convert light energy into chemical energy stored in

organic molecules (Buchanan, Gruissem and Jones (eds.), 2015; Taiz *et al.*, 2015). Plant growth is related to photosynthesis however not solely. Growth itself is a complex, integrated outcome of multiple physiological processes. Biomass formation requires the uptake of essential minerals and water besides the

photosynthetic carbon, which are acquired through roots and physiological transport mechanisms that require significant metabolic energy (Lambers, Chapin and Pons, 2008). Respiration, another essential biochemical process, consumes a portion of photosynthetic to support cellular maintenance and active transport. Therefore, a comprehensive understanding of plant growth requires an assessment of total daily net photosynthetic carbon at the whole plant level (Poorter *et al.*, 2012).

All the above processes are exposed to external factors that affect their course. The literature reports various biotic stresses affecting plants, including pathogenic fungi (Mapuranga *et al.* 2023), aphid herbivory (Florencio-Ortiz, Sellés-Marchart and Casas, 2021), allelopathy between plants (Zhang *et al.*, 2020), and parasitic plants (Zagorchev *et al.*, 2022). However, the effects of insect activity on plant physiological processes remain insufficiently explored. In particular, the question of whether and how pollinators can influence photosynthesis in different plant species appears especially intriguing.

Warrington, Cottam and Whittaker (2013) examined the impact of leaf blade damage caused by the typhlocybine leafhopper (*Ossiannilssonola callosa*) on the physiology of plane tree seedlings. Measurements showed a 22% decrease in photosynthesis, a 25% decrease in the daily rate of water loss and a 34% increase in the nocturnal rate of water loss, likely due to impaired stomatal function. Similar results obtained Visakorpi *et al.* (2018) when investigated how the common winter moth (*Operophtera brumata*), a common herbivore in temperate forests, affects the photosynthesis and isoprene emission rates of its host plant, English oak (*Quercus robur*). Aldea *et al.* (2005) found that herbivory by Japanese beetle (*Popillia japonica*) and corn earworm (*Helicoverpa zea*) caused a 20–90% increase in transpiration from soybean leaflets while leaving carbon assimilation rates or photosynthetic efficiency (ΦPSII) unaffected. Experiment conducted with the large earth bumblebee (*Bombus terrestris*) indicated that the bumble bee workers affect tomatoes and black mustard (*Brassica nigra*) flowering time. In tomato (*Solanum lycopersicum*), the average flowering time of bee-affected plants was 30 days earlier than that of unexposed plants and for *B. nigra* it was 16 days earlier (Pashalidou *et al.*, 2020).

Ma *et al.* (2022) in the experiment with spinach and pollinators proved that pollination has significant metabolic meaning in the plant. Analysis of RNA-seq showed that sepals after pollination indicate higher photosynthetic activity than before the interaction. On the other hand, Baek *et al.* (2021) investigated influence of western honeybee (*Apis mellifera*) presence on thale cress (*Arabidopsis thaliana*), findings indicated that bees cause increase level of stress-related metabolites, such as: glucosinolates, policosanols, tocopherols, amino acids, and sugars. The metabolic profile of plants exposed to bees differs significantly from that of unexposed plants.

Pollinators play crucial role for the environment. They maintain stability in biodiversity and ecosystems. The pollination itself enhances both the quality and quantity of agricultural crops, Klein *et al.* (2007) indicated that around 75% of crops rely on pollinators. According to EEA (2025), the economic value of pollination for agriculture in the EU is estimates as at least 5–15 bln EUR annually. Also, the decline in number of pollinators is considered as indicator of environmental degradation (Potts *et al.*, 2010). Taking into consideration the above the interaction plant – pollinator and plants physiological response to it, requires further

investigation. Methods based on chlorophyll fluorescence and gas exchange allows for quick and precise determination of a plant's condition, including physiological stress (Song and Zhu, 2024).

MATERIALS AND METHODS

EXPERIMENTAL DESIGN

The experiment was conducted in a municipal apiary located in Szczecin, in the North-Western Pomerania region. As part of the experiment, two variants of plant sites were prepared: exposed to contact with pollinating insects and protected from such interaction. The location of the sites within the apiary allowed free access of pollinators to the plants in the open variant. Plants isolated from pollinators were placed in net-house made of polyurethane net with a mesh size of 1 mm, sand-coloured, which prevented access of insects while maintaining light conditions similar to natural ones. Control plants were left without net. Light intensity was measured with a luxmeter – under the mesh it reached value of 98,000 lx, while in uncovered conditions it was approx. 100,000 lx. In order to increase the reliability of the obtained results, two plant species were used in the experiment: pepper mint (*Mentha* sp.) – perennial plant and pickling cucumber (*Cucumis sativus* L.) cultivar Delicius – annual plant. The pot experiment was conducted from April to August 2023 and included 15 pots per variant. In April, the tested plants were prepared; cucumbers were grown from seeds, and mint was grown from seedlings from the mother plant. In June all plants were replanted in containers of 5 dm³ capacity filled with a homogeneous, standard gardening substrate containing a starting dose of NPK fertiliser and placed at the experimental site. Pollination of exposed plants occurred through the transfer of pollen by pollinators onto the flower stigma. No herbivorous activity of insects was observed on tested plants. Measurement of tested parameters was conducted at the turn of July and August when mint was at 65 BBCH¹⁾ and cucumber was at 71 BBCH.

PIGMENTS CONTENT

The content of chlorophyll *a*, *b*, total (tot) and carotenoids (car) was analysed using fresh leaf material from tested plants. The determinations were carried out using the method by Arnon, Allen and Whatley (1956), modified by Lichtenthaler and Wellburn (1983). Leaf samples were homogenised in 10 cm³ of 80% acetone solution and then centrifuged to obtain the supernatant. Next, the optical density of the extracts obtained was determined spectrophotometrically at wavelengths of 440 nm, 645 nm, and 663 nm.

GAS EXCHANGE ANALYSIS

Gas exchange measurement was performed using the TPS-2 Portable Photosynthesis System (PP Systems) equipped with a PLC-4 camera. The following physiological parameters were

¹⁾ The BBCH-scale (Ger.: Biologische Bundesanstalt, Bundessortenamt und Chemische Industrie) is a system for a uniform coding of phenologically similar growth stages of all mono- and dicotyledonous plant species.

recorded: stomatal conductance (G), carbon dioxide concentration in the intercellular spaces (C_i), transpiration (E), and carbon dioxide assimilation rate (A).

CHLOROPHYLL FLUORESCENCE

Chlorophyll *a* fluorescence was determined using a PEA Handy fluorimeter (Hansatech Instruments, Ltd., United Kingdom) on leaves completely darkened before measurement and then exposed to pulsed light. The analysed parameters included time to maximum fluorescence ($t(F_m)$), area under the fluorescence induction curve (Area), maximum fluorescence (F_m), variable fluorescence (F_v) and quantum yield for reduction of end electron acceptors at the PSI acceptor side ($\phi(RO)$).

STATISTICS

All the measured parameters were statistically analysed by the ANOVA model. Fischer's test was used as a post hoc at a 0.05 confidence level. The mathematical relationship between particular parameters were estimated based on Pearson's correlation coefficient at a 0.05 confidence level. Analyses were performed by using the Statistica 13.0 program (Statsoft, Inc. Tulsa, USA).

RESULTS

PIGMENTS CONTENT

The application of insect protection influenced the content of individual photosynthetic pigments (Fig. 1). However, the response to insect presence also depended on the plant species. The average chlorophyll *a* content (Chl *a*) in mint leaves under unprotected conditions was $1,084.0 \mu\text{g}\cdot\text{g}^{-1}$ and did not differ significantly from the value obtained under protected conditions ($1,133.4 \mu\text{g}\cdot\text{g}^{-1}$). In cucumber, the value of this parameter under unprotected conditions was $707.7 \mu\text{g}\cdot\text{g}^{-1}$, which was significantly lower than under protected conditions ($1,015.7 \mu\text{g}\cdot\text{g}^{-1}$). The average chlorophyll *b* content (Chl *b*) in mint leaves under control conditions was $392.3 \mu\text{g}\cdot\text{g}^{-1}$ and did not undergo significant changes after applying protection ($356.1 \mu\text{g}\cdot\text{g}^{-1}$). In cucumber, the absence of insects led to a significant increase in this parameter. Plants grown without insect protection had an average chlorophyll *b* content of $211.9 \mu\text{g}\cdot\text{g}^{-1}$, whereas in protected plants, this parameter increased by 39.1%, reaching $294.7 \mu\text{g}\cdot\text{g}^{-1}$. The total chlorophyll content (Chl tot) in mint leaves showed no significant differences between unprotected ($1,476.4 \mu\text{g}\cdot\text{g}^{-1}$) and protected conditions ($1,489.5 \mu\text{g}\cdot\text{g}^{-1}$). A different effect was observed in cucumber, where the average total chlorophyll content was $914.6 \mu\text{g}\cdot\text{g}^{-1}$ under unprotected conditions and significantly increased to $1,310.4 \mu\text{g}\cdot\text{g}^{-1}$ under protected conditions, indicating a different response of this species to the applied experimental conditions. The average carotenoid content (Car) in mint leaves under unprotected conditions was $534.9 \mu\text{g}\cdot\text{g}^{-1}$ and did not change significantly after the application of insect protection ($551.6 \mu\text{g}\cdot\text{g}^{-1}$). In cucumber, protection caused a significant increase in the content of this pigment. The average carotenoid content under unprotected conditions was $381.8 \mu\text{g}\cdot\text{g}^{-1}$, whereas after protection, it increased by 79.5%, reaching $480.3 \mu\text{g}\cdot\text{g}^{-1}$.

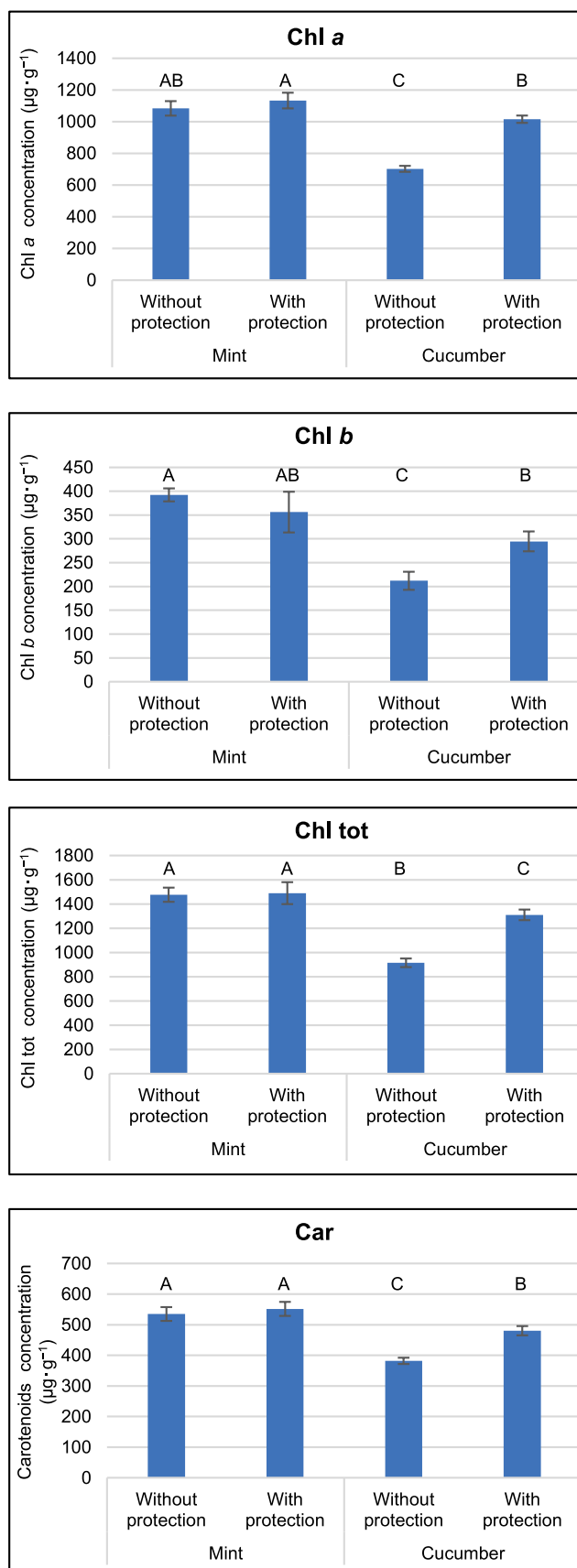


Fig. 1. The comparison of chlorophyll and carotenoids content in mint and cucumber cultivated exposed and unexposed to pollinators; the different letters represent the significance of the differences at the $p < 0.05$; Chl *a* = chlorophyll *a*, Chl *b* = chlorophyll *b*, Chl tot = total chlorophyll, Car = carotenoids; source: own study

GAS EXCHANGE PARAMETERS

Isolation plants from the insects influenced gas exchange parameters. Like photosynthetic pigments, the response to insect presence depended on the plant species (Fig. 2). The average CO_2 assimilation rate in mint under unprotected conditions was $5.0 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ and did not differ significantly from the value obtained under protected conditions ($5.2 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). In cucumber, the value of this parameter under unprotected conditions was $4.5 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ and was significantly higher than under protected conditions ($4.0 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). The average transpiration rate in mint leaves under control conditions was $0.77 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and did not undergo significant changes after applying the protection. In cucumber, no statistically significant differences were observed between variants. Stomatal conductance in mint leaves showed no significant differences between unprotected and protected conditions (50.8 and $56.0 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$, respectively). A different effect was observed in cucumber, where the average value of this parameter in unprotected plants was $58.0 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and significantly increased to $75.4 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ under protected conditions, confirming a different response of this species to the applied experimental conditions. The average intercellular CO_2 concentration in mint leaves under unprotected conditions was $499.2 \mu\text{mol mol}^{-1}$ and did not change significantly after insect protection ($457.8 \mu\text{mol mol}^{-1}$). In cucumber, protection caused a significant decrease in this parameter from 527.2 to $420.6 \mu\text{mol mol}^{-1}$.

CHLOROPHYLL FLUORESCENCE PARAMETERS

Radar charts show the average JIP test chlorophyll fluorescence parameters for mint and cucumber under unprotected and protected conditions (Fig. 3). These values were normalised, with the unprotected variant always set to 100%, and the protected variant values were compared accordingly. Statistical analysis showed no significant differences between variants for any analysed parameters in mint. Significant changes were observed in the parameters time to reach maximum fluorescence ($t(\text{Fm})$), area under the fluorescence induction curve (Area), maximum fluorescence (Fm), variable fluorescence (Fv), and quantum yield for reduction of end electron acceptors at the PSI acceptor side ($\phi(\text{Ro})$) in cucumber. The $t(\text{Fm})$ under unprotected conditions was 560.6 rel.u. , and significantly increased after protection to 703.3 rel.u. , indicating a prolonged PSII reaction time. The Area was significantly higher in plants under protection ($28,960 \text{ rel.u.}$) compared to control conditions ($22,290 \text{ rel.u.}$). The Fm under unprotected conditions was 888.6 rel. u. and significantly increased in protected plants to 1055.4 rel.u. , while Fv under control conditions was 654.7 rel. u. and it was significantly lower than in protected plants (810.4 rel.u.). Electron flux reducing end electron acceptors at the PSI acceptor (REo/RC) was significantly lower in protected plants (0.44 rel.u.) compared to control conditions (0.56 rel.u.). The quantum yield for reduction of end electron acceptors at the PSI acceptor side ($\phi(\text{Ro})$) was also significantly lower in protected plants, reaching 0.22 rel. u. compared to 0.26 rel.u. under control conditions.

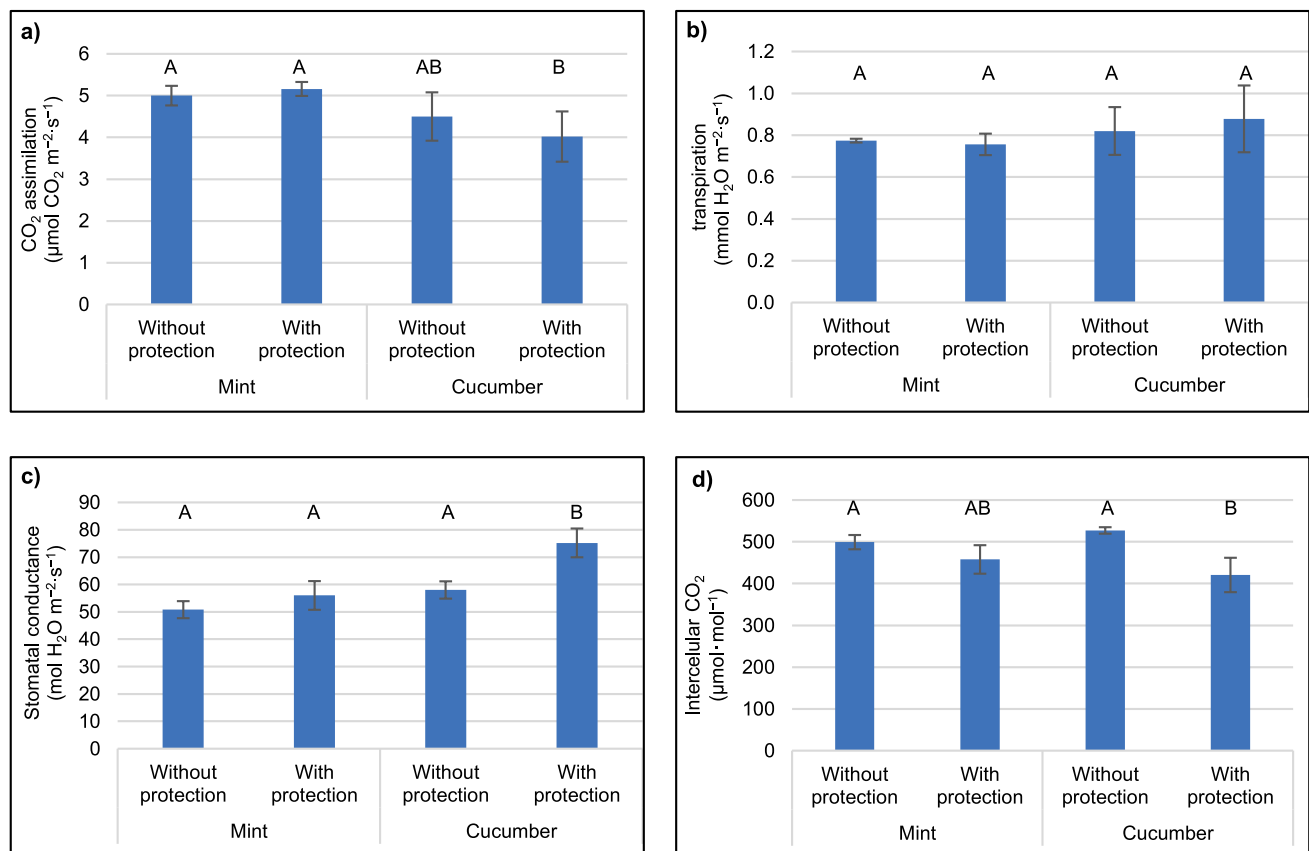


Fig. 2. The comparison of gas exchange parameters in mint and cucumber cultivated exposed and unexposed to pollinators: a) CO_2 assimilation, b) transpiration, c) stomatal conductance, d) intercellular CO_2 concentration; the different letters represent the significance of the differences at the $p < 0.05$; source: own study

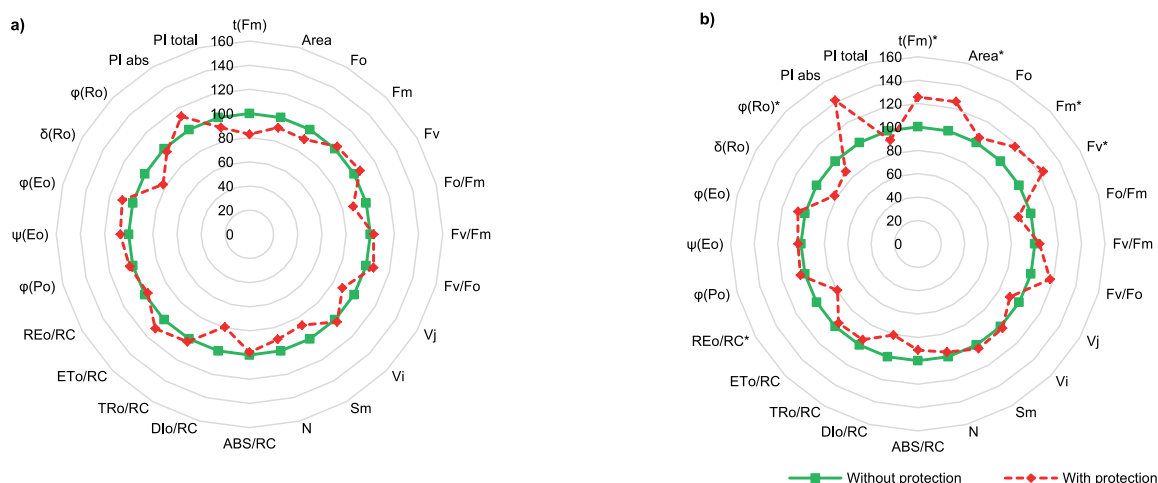


Fig. 3. Radar charts of the average JIP test chlorophyll fluorescence parameters for plants cultivated exposed and unexposed to pollinators: a) mint, b) cucumber; $t(F_m)$ = time to reach maximum fluorescence, Area = fluorescence area, F_m = maximum fluorescence, F_v = variable fluorescence, REo/RC = electron flux reducing end electron acceptors at the PSI acceptor, $\phi(Ro)$ = the quantum yield for reduction of end electron acceptors at the PSI acceptor side, * statistically significant difference; source: own study

CORRELATION MATRIX FOR PIGMENT CONTENT, GAS EXCHANGE PARAMETERS, AND JIP TEST CHLOROPHYLL FLUORESCENCE PARAMETERS

The correlation matrix presents the relationships between photosynthetic pigment content, gas exchange parameters, and JIP test chlorophyll fluorescence parameters (Fig. 4). The results for cucumber were analysed because, in this species only, the analysis of variance showed significant changes in the magnitude of individual parameters. Only those that showed sensitivity to insect influence were analysed among the chlorophyll fluorescence parameters. The chlorophyll *a*, *b*, and total chlorophyll contents were strongly correlated. Strongly correlated with

chlorophyll content were also Car (Car-Chl *a*: $r = 0.99$, Car-Chl *b*: $r = 0.98$, Car-Chl tot: $r = 1.00$). Strongly correlated negatively with chlorophyll and carotenoid content was A (e.g., A-Chl *a*: $r = -0.96$, A-Chl tot: $r = -0.94$). Stomatal conductance (*G*) positively correlated with chlorophyll content (e.g., *G*-Chl *a*: $r = 0.97$, *G*-Chl tot: $r = 0.96$). The REo/RC was negatively correlated with chlorophyll and carotenoid content (e.g., REo/RC -Chl *a*: $r = -0.91$, REo/RC -Chl tot: $r = -0.92$), suggesting that at higher pigment content, electron transport is less intense. The F_m showed a positive correlation with F_v ($r = 0.84$) and quantum yield for reduction of end electron acceptors at the PSI acceptor side ($\phi(Ro)$, $r = 0.84$), indicating that an increase in F_m is associated with an enhanced PSII electron transport capacity.

Parameter	Chl <i>a</i>	Chl <i>b</i>	Chl tot	Car	A	E	G	Ci	$t(F_m)$	Area	F_m	F_v	REo/RC	$\phi(Ro)$
Chl <i>a</i>		*	*	*	*	*	*						*	
Chl <i>b</i>	*		*	*	*	*	*						*	
Chl tot	*	*		*	*	*	*						*	
Car	*	*	*		*	*	*						*	
A	*	*	*	*		*	*	0.81					*	
E	*	*	*	*			0.90*	-0.63					*	
G	*	*	*	*		*		-0.80	*				*	
Ci	*	*	*	*		*			*				*	
$t(F_m)$	*	*	*	*		*	*	*					*	
Area					*	*	*						*	
F_m						*	*					*	*	
F_v						*	*				*		*	
REo/RC	*	*	*	*	*	*	*					*	*	
$\phi(Ro)$						*	*				*	*	*	
From -1.00 to -0.76 from -0.75 to -0.51 from -0.50 to -0.26 from -0.25 to 0.00 from -0.01 to 0.25 from 0.26 to 0.50 from 0.51 to 0.75 from 0.76 to 1.00														

Fig. 4. The correlation matrix of relationships between photosynthetic pigment content, gas exchange parameters, and JIP test chlorophyll fluorescence parameters; Chl *a* = chlorophyll *a*, Chl *b* = chlorophyll *b*, Chl tot = total chlorophyll, Car = carotenoids, A = CO_2 assimilation, E = transpiration, G = stomatal conductance, Ci = intercellular spaces, $t(F_m)$ = time to maximum fluorescence, Area = area under the fluorescence induction curve, F_m = maximum fluorescence, F_v = variable fluorescence, REo/RC = electron flux reducing end electron acceptors at the PSI acceptor, $\phi(Ro)$ = quantum yield for reduction of end electron acceptors at the PSI acceptor side; source: own study

The vector graph illustrates the relative *involvement* of each input variable in forming the principal components (Comp. 1 and Comp. 2). The vector magnitude reflects the influence on the corresponding chlorophyll fluorescence (ChFI) parameter. The vector direction indicates its impact on both components. The analysed parameters exhibited similar sensitivity to petroleum contamination and contributed comparably to the formation of the principal components (Dąbrowski *et al.*, 2024a). Modifications in the first principal component (Comp. 1) accounted for 68.9% of the total variance, while the second component (Comp. 2) explained 24.6%. The parameters can be categorised into three groups: the first group includes Ci, REo/RC, and A; the second group consists of Fm, Fv, and $\phi(Ro)$; and the last group comprises t(Fm), E, and G (Fig. 5).

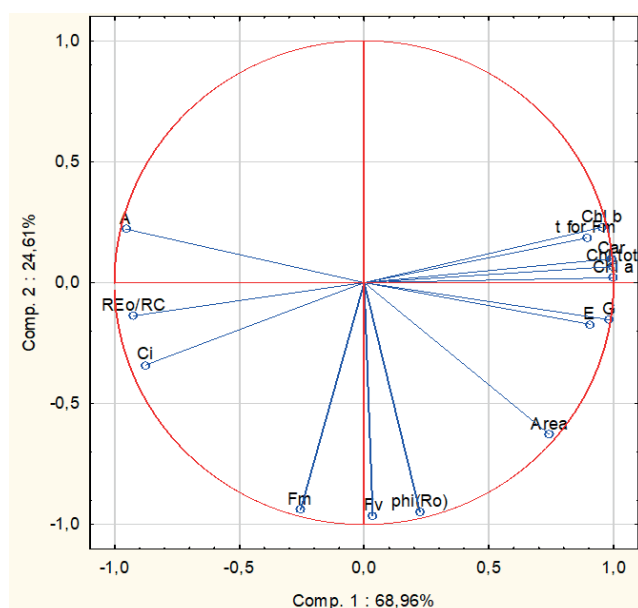


Fig. 5. The vector graph of the relative *involvement* of each input variable in forming the principal components (Comp. 1 and Comp. 2); variables as in Fig. 4; source: own study

DISCUSSION

The chlorophyll *a*, *b*, and carotenoid content (Chl *a*, Chl *b*, and Car, respectively) exhibited a varied response to protection depending on the plant species. In mint, no significant differences were observed between the experimental variants, suggesting that this species is less susceptible to the pollinators effect. The lack of significant changes in pigment content may indicate effective compensatory mechanisms that maintain a stable pigment level (Lichtenthaler, 1987). Cucumber's investigations indicate a significant increase in chlorophyll content under insect protection conditions, e.g., the total chlorophyll content (Chl tot) increased from 914.6 to 1,310.4 $\mu\text{g}\cdot\text{g}^{-1}$. A similar effect was observed for carotenoids, whose content increased by 79.5%. This may suggest that insect pressure limits the synthesis of photosynthetic pigments or accelerates their degradation, as confirmed by previous studies on plant responses.

Leaf photosynthesis is the primary energy source for the growth and development of all plants. It is now widely recognised that, in C3 plants, photosynthesis reacts to changes in their

environmental conditions mainly by stomatal conductance (G) and/or the biochemical capacity of the leaf (Flexas and Carriqui, 2020). After applying for protection, gas exchange analysis indicates that the mint did not exhibit significant changes in CO_2 assimilation (A) G, or transpiration (E). The stability of these parameters confirms the resistance of this species to insect presence, which may result from its ability to efficiently regulate photosynthetic mechanisms and limit photosynthetic losses (Farquhar and Sharkey, 1982). Conversely, cucumber showed a different trend – A was significantly higher under unprotected conditions ($4.5 \mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) than after applying protection ($4.0 \mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). This may result from a compensatory increase in photosynthesis intensity in response to mechanical damage or metabolic stimulation caused by feeding insects (Flexas *et al.*, 2009). The increase in stomatal conductance in cucumber after protection (from 58.0 to 75.4 $\text{mol H}_2\text{O m}^{-2}\cdot\text{s}^{-1}$) indicates more significant stomatal opening, which may be an effect of reduced stress and improved water availability (Lawson and Blatt, 2014). Also, the negative changes of intercellular CO_2 concentration in plants without protection suggest that this gas accumulates within intercellular spaces of the leaf when Rubisco²⁾ does not assimilate it (Dellero *et al.*, 2021).

In general, the reduction in the photosynthetic efficiency of photosystem II (ΦPSII) in plants exposed to stress may serve as a mechanism to protect PSII reaction centres (Rapacz *et al.*, 2015; Dąbrowski *et al.*, 2024b; Dąbrowski *et al.*, 2024c). In this work, we analysed the ΦPSII by chlorophyll fluorescence measurements. Chlorophyll fluorescence analysed using the JIP test reveals interesting relationships regarding ΦPSII . In mint, no significant differences were observed between the variants, suggesting that PSII function remains stable regardless of insect presence. In cucumber, insect protection significantly affected several key fluorescence parameters. The time to reach maximum fluorescence (t(Fm)) increased from 560.6 to 703.3 rel.u., indicating an extended PSII response time. This may suggest that the PSII antenna system has adapted to altered light conditions or a reduced need for oxidative stress protection (Baker, 2008). The Fm significantly increased after protection was applied (from 888.6 to 1055.4 rel.u.), which can be attributed to higher pigment content and, consequently, an enhanced capacity for light energy absorption (Müller, Li and Niyogi, 2001). Electron transport flux (REo/RC) and the quantum yield for reduction of end electron acceptors at the PSI acceptor side ($\phi(Ro)$) were significantly lower in protected plants, meaning that electron transport to reductive acceptors was slowed. This may be due to lower oxidative stress and reduced photoprotection needs (Demmig-Adams and Adams, 1996).

The literature shows strong positive correlations between Chl *a*, Chl *b*, and Chl tot (Lichtenthaler and Wellburn, 1987). This is expected, as both chlorophyll types coexist in photosystems I and II antenna complexes. The strong correlation between Car and Chl ($r \approx 1.00$) is also reflected in studies suggesting that Car play a protective role and are proportionally associated with chlorophylls in chloroplasts (Demmig-Adams and Adams, 1996).

The strong negative correlation between CO_2 assimilation (A) and Chl and Car content (e.g., $r = -0.96$ for Chl *a*) may

²⁾ Ribulose-1,5-bisphosphate carboxylase/oxygenase – the enzyme that catalyses the first step of carbon fixation in photosynthesis.

suggest that higher pigment content does not necessarily correlate with higher net photosynthesis efficiency. This may result from chlorophyll accumulation under stress conditions (e.g., caused by insect activity) and the simultaneous limitation of photosynthetic efficiency by other factors, such as G (Flexas *et al.*, 2009).

Conversely, the strong positive correlation between G and Chl (e.g., $r = 0.97$ for Chl *a*) is consistent with findings suggesting that plants with higher pigment content may exhibit more open stomata, increasing CO₂ uptake and transpiration (Farquhar and Sharkey, 1982).

The strong negative correlation between electron transport flux (REo/RC) and pigment content (e.g., $r = -0.92$ for Chl tot) suggests that increased pigment levels may limit electron transport in PSII. This may result from photoprotective mechanisms that enhance energy integration into Car at the expense of electron transport (Müller, Li and Niyogi, 2001).

In contrast, the positive correlation ($r = 0.84$) between maximum fluorescence (Fm) and variable fluorescence (Fv) as well as the quantum yield for reduction of end electron acceptors at the PSI acceptor side ($\phi(Ro)$, $r = 0.84$) suggests that plants with higher Fm may have more efficient PSII. It is worth noting that an increase in Fm may also be associated with increased chlorophyll accumulation and reorganisation of photosynthetic complexes, optimising light capture and energy transfer (Baker, 2008).

The presented results align with previous studies on the relationships between photosynthetic pigments, gas exchange, and chlorophyll fluorescence. The interpretation of the strong correlation between chlorophyll content and photosynthetic parameters suggests that insect-induced stress may affect the functioning of the photosynthetic apparatus through changes in energy distribution and electron transport efficiency.

CONCLUSIONS

The research findings indicate that plant responses to insect protection were highly species-dependent. Mint showed no significant differences in pigment content, gas exchange, or chlorophyll fluorescence, suggesting that it is less sensitive to pollinators' effect. Cucumber exhibited significant changes in chlorophyll and carotenoid content, as well as in stomatal conductance and fluorescence parameters. Insect protection increased photosynthetic pigment levels and improved PSII stability, although it simultaneously reduced CO₂ assimilation intensity. These differences may result from the distinct adaptive strategies of plants – mint, as a perennial species, may better tolerate pollinator exclusion. In contrast, cucumber, an annual plant, exhibits more dynamic responses in photosynthetic metabolism. Future studies should aim to disentangle the effects of pollinator activity from environmental modifications caused by protective measures, for example by simulating similar light conditions in the non-protected variant or applying an open mesh net. Integrating physiological measurements with yield assessments may help clarify the mechanisms behind these species-specific reactions and their agronomic significance.

CONFLICT OF INTERESTS

All authors declare that they have no conflict of interests.

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