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Influence of No-Till System with or without Cover Crop on Glyphosate Tolerant Soybean Productivity and Adaptation to Drought

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ABSTRACT: Soybean are vulnerable to drought and temperature increases potentially induced by climate change. The purpose of this study was to determine if the use of cover crops (CC) can influence the gas exchange potential of glyphosate tolerant soybean when the vapor pressure deficit (Vpd) increases. This two years study was conducted in an open experimental field comprising Direct Seeding plots without CC (DS) or with CC (DSCC). Stomatal conductance (Gs) was measured five times on the same identified leaves following GBH application. These leaves were then collected in order to observe the stomata (size, density and stomatal index) with a scanning electron microscope and to perform measurements on foliar traits (venation density, width of midrib). The Vpd was calculated concomitantly to Gs measurements at the leaf surface. The results suggest that the use of CC promotes phenotypic change in soybean leaves (more elaborate venation and a higher stomatal density), which in turn may enhance their tolerance to drier conditions. In 2019, Gs could be up to 29% higher in DSCC plots compared to DS ones along with similar Vpd values. This study shows that the benefits of using CC can be observed through the morphological development strategies of the crop plants and their higher tolerance to drought.

Keywords: stomatal conductance; stomatal density; stomatal size; vapor pressure deficit; vein density; foliar traits; GBH: glyphosate-based herbicide

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1. INTRODUCTION

Over the recent years, alternative cropping systems have been proposed to challenge the negative environmental impacts of conventional field cropping caused by intensive mechanical soil disturbance and use of synthetic pesticides and fertilizers (Carlson et Stockweel, 2013; DeLonge *et al.*, 2016; FAO et ITPS, 2015; Magdoff, 2007; Oerke, 2006; Pimentel *et al.*, 1995; Triplett et Dick, 2008). Direct seeding (DS) systems have been put forward to reduce mechanical tillage and the incidence of soil erosion. DS systems allow reducing carbon and nitrate leaching from soils or emissions to the atmosphere, and maintaining soil organic carbon (SOC) and soil functions (Kassam *et al.*, 2019; Pimentel *et al.*, 1995; Triplett et Dick, 2008; Yu *et al.*, 2020). The use of glyphosate-based herbicides (GBH) in combination with glyphosate resistant (GR) seeds has promoted the adoption of DS systems on a larger scale (Derpsch, 1998; Derpsch *et al.*, 2010). During 1998 and 2008, DS area have increased by 71.6% in soybean production in the United States of America (Yu *et al.*, 2020). Although DS systems aim at maintaining SOC and soil functions, they tend to provoke surface soil compaction, in turn limiting water infiltration, seed germination and the development of crop plants (Triplett et Dick, 2008). Reduced water infiltration into the soil can result in water limitation for crops, which can influence their physiological activities and their gas exchange potential (Domec *et al.*, 2009; Driesen *et al.*, 2020). Stomata present on leaves constitute the main sites for CO_2 assimilation by the plants (Tanaka *et al.*, 2010; Zeiger *et al.*, 1987). Stomata also play an important role in plant transpiration, since for the uptake of CO_2 corresponds to water release, i.e., a significant trade-off for the metabolic management of the plant (Driesen *et al.*, 2020; Krober et Bruelheide, 2014). Along with climate change, the occurrence of drier periods will be more frequent and will have a considerable influence on the water content of soils during key periods of crop plants growth.

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We conducted a two years field study aiming at determining how the implementation of cover crops (CC) may influence the gas exchange potential of glyphosate-tolerant soybean in DS systems. Direct seeding on cover crops (DSCC) imply implementing catch crop between field crop production periods or intercropping during the field crop production periods (Hartwig et Ammon, 2002; Woolford et Jarvis, 2017). The addition of CC may bring agronomic benefits such as limiting surface soil compaction, increasing soil porosity and water infiltration and limiting soil water evaporation (Amsili et Kaye, 2021; Hartwig et Ammon, 2002; Liu *et al.*, 2005; Robertson *et al.*, 2014; Wagg *et al.*, 2021). C3 plants such as soybean are sensitive to abiotic factor such as temperature, air humidity, light intensity in turn influencing the gas exchange potential that contributes to water management and photosynthesis of the plant (Driesen *et al.*, 2020; Roche, 2015). In the coming years, climate change will have strong repercussion on the temperature and air humidity that will cause more frequent and longer drought period (Seager *et al.*, 2015; Zhao *et al.*, 2017). It has been reported that the increase of vapor pressure deficit (Vpd) has a negative impact on field crop production (Seager *et al.*, 2015; Zhao *et al.*, 2017). Vpd represents the difference between the amount of water vapor that air can contain at saturation and the actual vapor pressure in the air. Vpd can thus be considered as a direct measurement of the atmospheric desiccation power, an important factor influencing plants productivity and sensitivity to other abiotic stressors (Grossiord *et al.*, 2020; Ocheltree *et al.*, 2014). Because Vpd is highly dependent upon temperature, it will increase with global warming and thus raise questions regarding field crops water management. This study aims at determining if the use of CC in row crops may represent a clue for reducing soybean sensitivity to variation of Vpd and to drought periods. To our knowledge, few studies have yet reported the influence of CC on stomatal conductance while Vpd values are raising. Bernier Brillon et al. (2022) observed that higher Vpd values seemed to have a particular influence on soybean stomatal conductance. This 1-year study also pointed out the potential mitigation effect of CCs on crop plants on experiencing higher Vpd values (Bernier Brillon *et al.*, 2022). In addition, the present study also aims at integrating physiological activity data by complementing them with leaf drought tolerance traits data (ex. foliar size and vein architecture).

2. Materials and methods

2.1. Description of the experimental design

Experiments were conducted in 2019 and 2020 in an open field located at the Grain Research Center (CEROM) in St-Mathieu-de-Beloeil (Quebec, Canada, (45.5828 N, -73.2374 W). The experimental plots were established in 2018 on a humic Gleysol (AAFC, 1998) with a heavy clay texture (average and standard deviation percentage of clay: 72.625 ±0.916 %, loam: 27.375 ±0.916 % and sand: 0%). The 0-20 cm horizon has a soil mineral content of 12.87 ±2.51 mg kg^{-1} for P, 313.50 ±20.84 mg kg^{-1} for K, 2943.42 ±219.62 mg kg^{-1} for C, 803.17 ±48.27 mg kg^{-1} for Mg, 1056.71 ±19.32 mg kg^{-1} for Al, 11.00 ±0.47 mg kg^{-1} for Cu, 217.54 ±13.92 mg kg^{-1} for Fe, 24.92 ±5.72 mg kg^{-1} for Mn, 2.33 ±0.18 mg kg^{-1} for Zn and 47.60 ±3.06 mg kg^{-1} for Na. The size of each plot was 9 m x 20 m and each treatment was replicated four times. The experimental design relied on two Direct Seeding practices (DS and DSCC) with glyphosate tolerant soybean (Altitude R2®). In each plot, soybean was seeded on previous year corn residues at a rate of 90.81 kg ha^{-1}, May 18th 2019 and May 26th 2020. Autumn rye (200 kg ha^{-1}) was also sown before the corn harvest as catch crops in those plots. No cover crops were sown in the DS plots. Herbicide treatment (Roundup WheaterMax® with glyphosate a.i. at 540 g L^{-1}) has been applied twice at rates of 902 g ha^{-1} of glyphosate in DS and DSCC plots. The first application was done pre-sowing May 18th 2019 and June 2nd 2020. The second application was done post-emergence June 24th 2019 and July 3rd 2020 at V2 soybean growth stage. Soybean was harvested October 15th 2019 and October 31th 2020. The field meteorological data including total daily precipitation (mm) and minimum, maximum and average daily temperatures were recorded for the 2019 and 2020 production period with a weather station located on the CEROM main building (Figure 1a and 1b).

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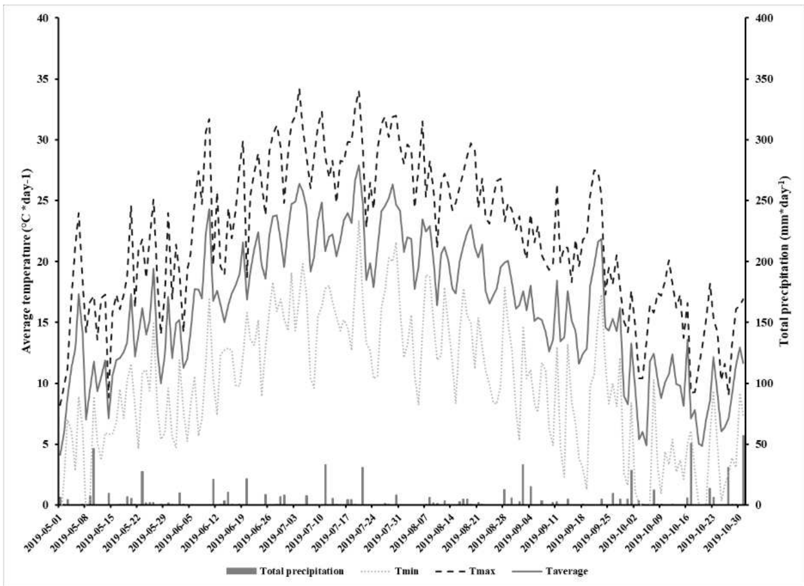
a)



b)

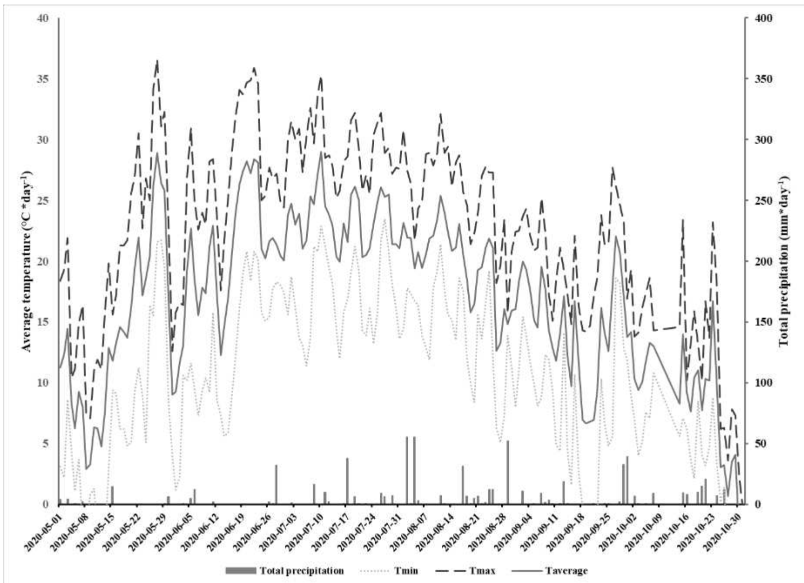


Figure 1. Total daily precipitation (mm) and minimum, maximum and average daily temperatures (°C) at the experimental field during the growth period (May to October) in 2019 (a) and 2020 (b).

2.2. Sampling and measurements

Stomatal conductance and stomatal traits

Stomatal conductance (Gs expressed as mmol m^{-2} s^{-1}) was measured with a steady-state diffusion porometer (SC-1 Leaf porometer, Decacon Devices®) using one leaflet from three different plants similarly arranged and with initially the same growth stage (V2). All plants and leaves were identified the V2 growth stage in order to follow the same plants and leaflets throughout the study period (from the V2 to the R2-R3 growth stages). The stomatal conductance was measured around midday on abaxial foliar surfaces during five fields sampling campaigns (48h and 7, 14, 21 and 28 days following the second GBH application). Leaves temperature and air relative humidity were also recorded using a portable psychrometer (REED instrument©, model#8706) at the leaf surface. The

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corresponding Vpd at the leaf surface was calculated according to the August-Roche Magnus formula, where Vpd = $6.1094^{17.625*T/T+243.04}$ (Alduchov et Eskridge, 1996; Murray, 1967).

After measuring the stomatal conductance 28 days after the second GBH application, each identified leaf (R2-R3 growth stage) was collected in order to calculate the stomatal size, density and index. On each leaf, three locations on one leaflet were observed for the stomatal density (StoDen) calculation with a scanning electron microscope (Hitachi S-3400N) at a magnification of 400x (Figure 2a). Pictures of those observations with the SEM were taken and the stomatal sizes (StoSize), width (StoWidth) and length (StoLength) were measured with ImageJ© software (NIH). The stomatal index (StoIndex) was calculated by multiplying the stomatal density by the stomatal size (Kim *et al.*, 2021).

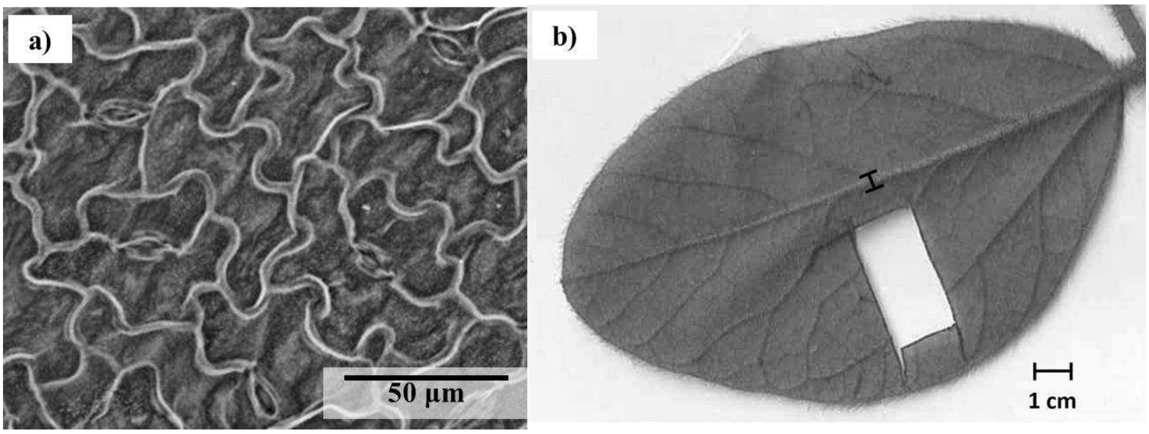

Figure 2. Observation of a) soybean stomata with a scanning electron microscope SEM at magnification of 400x and b) leaflet trait with an imagery software.

2.3. Foliar trait

One of the leaflets from each leaf collected in the field for stomatal trait measurements (28 days after second GBH application) were stored in a decolorizing solution (70% aqueous ethanol solution). Once decolored, these leaves were dipped in a safranin solution (4% v/v) until we obtained a sufficient staining of the foliar veins. This vein staining increased color contrast and allow better accuracy of leaf trait measurements. The colored leaflets were then scanned and the measurements were made using an imagery software (ImageJ© software).

In this study, distance between veins has been used to obtain a proxy of the venation density (Uhl et Mosbrugger, 1999). An average of 11 measurements has been taken between secondary vein for each leaflet. The midrib width and the leaves size of those leaflet have also been measured with the imagery software (ImageJ© software) (Figure 2b)

2.4. Statistical analyses

The Gs/Gsmax values from all plants and for both sampled years were used to obtain a generalized linear model (GLM) in function of the corresponding Vpd with beta distribution using a *logit* link function) (Figure 3) (Bernier Brillon *et al.*, 2022; Krober et Bruelheide, 2014). The inflexion point was calculated for each curve and considered as the optimal condition for gas exchange (Bernier Brillon *et al.*, 2022; Krober et Bruelheide, 2014). The Vpd values for these optimum points were calculated to determine if there is a difference in plant sensitivity to Vpd between DS systems with or without CC. Figure 3 shows an example of the fitted curves from the GLM as a function of Vpd centered value for each system and for the years 2019 and 2020. Since the logit function has only one rising point of inflection, the optimal gas exchange points were calculated from the second derivative for each curve. The confidence interval (95% CI) was calculated for those points to take into consideration the interval on the values for the stomatal conductance (the y-axis interval) and the Vpd (the x-axis interval).

An ANOVA analysis was carried out to assess whether there is a significant difference ($p \leq 0.05$) in Gs values between years for the stomatal traits, foliar traits, dry biomass production, different cultivation systems. Also, a Chi square test analysis was also carried out to evaluate the influence of both the year of production and the combination of year-agricultural management.

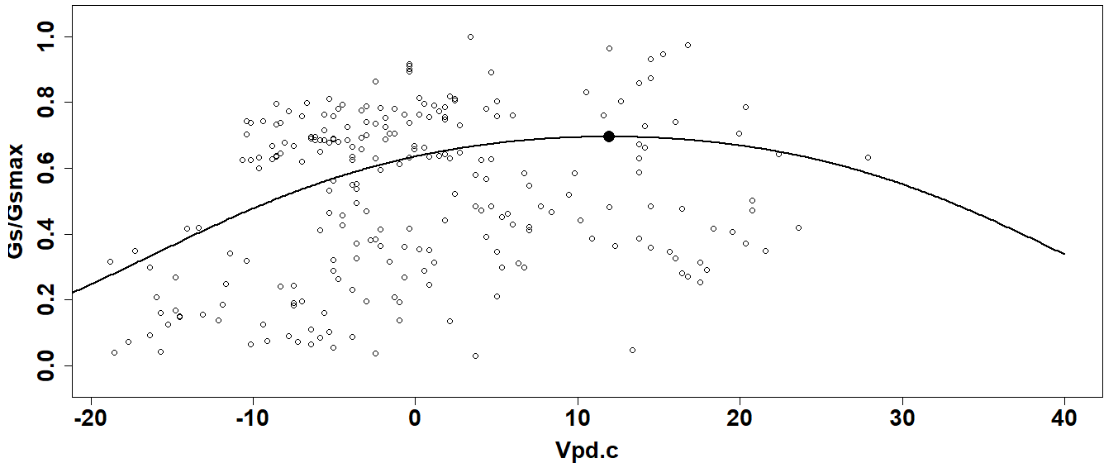

Figure 3. Example of Gs/Gsmax data (curve and point of inflexion) generated by the general linear model in function on centered values of vapor pressure deficit (Vpd.c).

3. RESULTS

3.1. Stomatal conductance and vapor pressure deficit

Combining DS and DSCC, significant differences in Gs between years are observed ($p = 0.0006$) where the means ± SE values were 635.15 ± 23.80 mmol $m^{-2}s^{-1}$ for 2019 and 516.14 ± 24.51 mmol $m^{-2}s^{-1}$ in 2020. On the opposite, the mean ± SE values of Vpd are significantly higher ($p < 0.0001$) in 2020 (58.68 ± 0.84 hPa) than in 2019 (51.33 ±0.75 hPa).

By modelling the relation between Gs and the raising values of Vpd, we observe that the calculated inflexion points from Gs values in 2019 is similar between DS and DSCC plots (Figure 4 and Table 1). However, we observe significant difference in 2019 between DSCC and DS Vpd values based on the 95% confidence interval (Figure 4 and Table 1) where DSCC have similar Gs values at higher Vpd values. No difference is observed on the calculated inflexion points from Gs values and on Vpd values in 2020 between DSCC and DS. The large variation around the inflexion points values in 2020 do not allow to observe difference with 2019.

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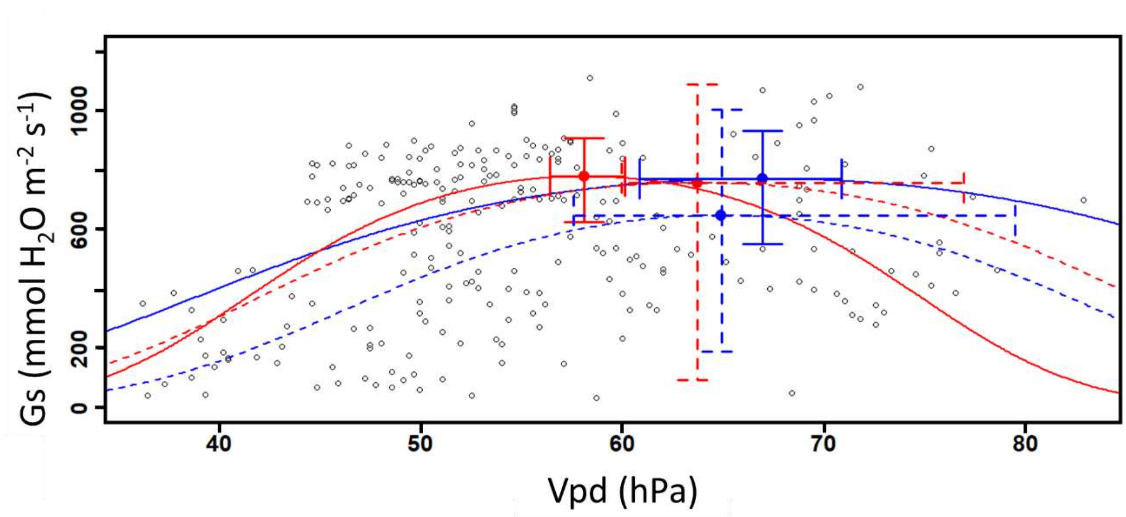

Figure 4. Stomatal conductance as a function of raising vapor pressure deficit in soybean (n=240). The curves represent the DS (Red) and DSCC (Blue) plots for 2019 (solid line) and 2020 (dashed line). The optimal points are defined here as the mean Gs and Vpd values with their associated confidence interval (95% CI).

Table 1. Calculated optimal gas exchange points following glyphosate-based herbicide application on soybean fields with DSCC and DS plots.

	2019		2020	
	DSCC	DS	DSCC	DS
Gs (mmol $m^{-2}*s^{-1}$)				
mean	772.45	780.00	649.15	759.07
95% CI [LL]	553.56	627.52	191.77	95.61
95% CI [UL]	932.89	909.13	1004.51	1088.46
Vpd (hPa)				
mean	66.94	58.10	64.92	63.73
95% CI [LL]	60.87	56.41	57.58	59.95
95% CI [UL]	70.86	60.14	79.49	76.95

Note: Data are presented as mean and their confidence interval (95% CI) with the lower limit (LL) and upper limit (UL) for the stomatal conductance (Gs) and the vapor pressure deficit (Vpd) values (r^2 = 0.2447).

3.2. Stomatal traits

Stomatal traits analysis (Table 2) shows that StoDen is significantly higher in DSCC plots compared to DS ones in both 2019 (p = 0.0581) and 2020 (p = 0.0247). StoLength, StoWidth and StoSize have higher values in DS plots in 2019 when compared to DSCC but no significant difference between the system is observed in 2020. In the case of StoIndex, no significant difference between DS and DSCC is observed for either year. However, different results on the stomatal traits are observed between both years where StoDen and StoIndex values for both systems are lower in 2020 compared to 2019. StoWidth and StoSize are significantly higher in DS plots in 2019 than in 2020 whereas no significant differences on those traits are observed in DSCC plots between 2019 and 2020.

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Table 2. Soybean stomatal traits measurements in direct seeding on cover crops (DSCC) and direct seeding (DS) for the years 2019 and 2020.

Stomatal traits	2019			2020		
	DSCC	DS	*p* value	DSCC	DS	*p* value
StoLength	15.91 ± 0.23 ^{bB}	16.78 ± 0.23 ^{aA}	**0.0068***	16.18 ± 0.23 ^{aAB}	16.12 ± 0.24 ^{aAB}	0.8379
StoWidth	6.75 ± 0.15 ^{bB}	7.60 ± 0.13 ^{aA}	**<0.0001***	6.93 ± 0.10 ^{aB}	6.97 ± 0.09 ^{aB}	0.7506
StoSize	109.34 ± 3.54 ^{bB}	128.72 ± 3.22 ^{aA}	**<0.0001***	112.60 ± 2.35 ^{aB}	113.43 ± 2.88 ^{aB}	0.8230
StoDen	307.0 ± 8.3 ^{aA}	282.6 ± 9.5 ^{bB}	**0.0481***	265.3 ± 9.2 ^{aBC}	247.0 ± 7.9 ^{bC}	**0.0247***
StoIndex	0.0343 ± 0.0023 ^{aAB}	0.0364 ± 0.0017 ^{aA}	0.4481	0.0297 ± 0.0011 ^{aBC}	0.0277 ± 0.0010 ^{aC}	0.2129

Note: Stomatal traits consists in stomatal length (StoLength) (µm), stomatal width (StoWidth) (µm), the stomatal size (StoSize) (µm^2), the stomatal density (StoDen) and stomatal index (StoIndex). Data are presented as mean ±SE from three leaves (with 3 observations by leaf) collected on all plots for each agricultural management replicates (four times) (n=72). The * and different lower-case letters indicate that mean values are significantly different between DS and DSCC plots ($p < 0.05$) according to a univariate test (ANOVA) and t-Student test respectively for each variable measured. Bold capital letters indicate significant difference between 2019 and 2020 according to a Chi square test ($p < 0.05$).

3.3. Foliar traits

No difference on the leaf size values between DSCC and DS is observed in 2019 and 2020 (Table 3). Also, the average leaf sizes are similar between years. Concerning the midrib width, significant differences exist between DSCC and DS in 2019 but not in 2020 (Table 3). In 2019, the midrib width values are higher in DSCC compared to DS plots. Also, significant smallest DistVein values are observed in DSCC for the year 2019 and 2020.

Table 3. Soybean foliar traits measurements in direct seeding on cover crops (DSCC) and direct seeding (DS) for the years 2019 and 2020.

	2019			**2020**		
	DSCC	DS	p value	DSCC	DS	p value
Leaf size (cm^2)	11.83 ± 0.48 ^{aA}	11.39 ± 0.43 ^{aA}	0.4912	11.77 ± 0.32 ^{aA}	12.47 ± 0.47 ^{aA}	0.2166
Midrib Width (mm)	9.13 ± 0.28 ^{aA}	7.88 ± 0.22 ^{bAB}	**0.001***	7.25 ± 0.35 ^{aB}	6.88 ± 0.29 ^{aC}	0.4201
DistVein (J.W. *et al.*)	0.63 ± 0.02 ^{bC}	0.80 ± 0.02 ^{aA}	**0.001***	0.69 ± 0.01 ^{bB}	0.76 ± 0.02 ^{aAB}	**0.0442***

Note: Foliar traits consists in leaf size (cm^2), midrib width (mm) and distances between secondary vein (DistVein) (J.W. *et al.*). Data are presented as mean ±SE from three leaves (with 3 observations by leaf for Leaf size and Midrib Width (n=72) and 11 measures by leaf for DistVein (n=264) collected on all plots for each agricultural management replicates (four times). The * and different lower-case letters indicate that mean values are significantly different between DS and DSCC plots ($p < 0.05$) according to a univariate test (ANOVA) and t-Student test respectively for each variable measured. Bold capital letters indicate significant difference between 2019 and 2020 according to a Chi square test ($p < 0.05$).

4. DISCUSSION

The observed correlation between Vpd and Gs confirms that Vpd influences the physiological activity of plants, which is consistent with other publications (Bernier Brillon *et al.*, 2022; Driesen *et al.*, 2020; Ocheltree *et al.*, 2014). Here, a positive relation between Vpd and Gs is observed until the gases exchange potential reaches an inflexion point defined as an optimal point (Figure 4). Once this value is reached, Gs values decrease along with higher Vpd values. Higher Vpd values promote the ascension of water in the xylem, enhancing water accumulation in the sub-stomatal cavities and its exit through plant transpiration (Driesen *et al.*, 2020; Sinclair *et al.*, 2017). The decreasing Gs values observed in this study can be explained by the fact that plants close their stomata in order to limit excessive water loss when their ambient environment becomes drier (higher Vpd values) (Bernier Brillon *et al.*, 2022; Driesen *et al.*, 2020; Krober et Bruelheide, 2014). Here, DSCC plots in 2019 appear less sensitive to Vpd while plants maintain the physiological activities and stomata opening during a less favorable periods (i.e., drought and hydric stress episodes). Gs values can be up to 29% higher in DSCC plots compared to DS ones for the same Vpd values in 2019 (Figure 4 and Table 1).

Interestingly, we are able to observe differences in morphological traits of plants grown in DS and DSCC plots (Table 2 and Table 3). This observation corroborates other research, which proposed that a difference in physiological activity and gas exchange can be explained by different leaf morphological traits. Those differences in leaf can be explained by morphological plasticity to optimize plant performance according to growth conditions (Carins Murphy *et al.*, 2014; Franks *et al.*, 2009; Puglielli *et al.*, 2017; Xiong *et al.*, 2017). These morphological differences can occur in stomata and other foliar traits such as the main vein structure (Carins Murphy *et al.*, 2014; Scoffoni *et al.*, 2011). Stomatal activity could be explained by the stomata density considering that it can be positively correlated with stomatal conductance (Chen *et al.*, 2016; Driesen *et al.*, 2020; Gaskell et Pearce, 1983; Qi et Torii, 2018; Roche, 2015; Tanaka *et al.*, 2010; Tanaka et Shiraiwa, 2009; Tanaka *et al.*, 2008). StoDen is a good drought proxy and is an indicator of the strategy that plants adopt to develop their stomata while reducing stressful conditions (Bernier Brillon *et al.*, 2023; Bernier Brillon *et al.*, 2022; Driesen *et al.*, 2020; Tanaka et Shiraiwa, 2009). These strategies closely influence the number and size of stomata, which in combination represent the potential for foliar epidermal cells surface allocation for gas exchange and optimal stomatal conductance potential (Franks *et al.*, 2009). In this study, the epidermal cells surface allocation for gas exchange is represented by the StoIndex value, which shows no significant differences between agricultural managements and years (Tableau 2). We observe that in 2019, StoDen of soybean growing in DSCC plots is higher than that of plants growing in DS plots (Table 2). This could explain the differences in stomatal behaviour and gas exchange where DSCC StoDen values are significantly different from those for plants growing in DS plots in 2020 (p value = 0.0247) (Table 2). Moreover, the stomata of plants growing in DSCC plots were significantly smaller than those in DS plots (Table 2). This was expected considering that it has been largely demonstrated that a negative relationship generally exists between stomatal size and number of stomata (Franks *et al.*, 2009). However, smaller and more numerous stomata allows soybean to quickly adapt their stomatal aperture for optimal conductance or closing them in order to avoid excessive transpiration (Aasama *et al.*, 2001; Bernier Brillon *et al.*, 2022; Franks *et al.*, 2009). This can be a significant short-term advantage, especially for crops that have to react quickly without allocated epidermal cell for the development of new stomata. For short-lived crops like soybean, plants tends to optimise resource acquisition by minimizing construction cost (Correia et Ascensão, 2017; Puglielli *et al.*, 2017). In the case of DSCC plants in 2019, a higher StoDen allowed physiological plasticity, which allows to maintain gas exchange in a context where Vpd values were higher.

In addition, the different stomatal traits between agricultural managements can also be explained by the morphological differences of the foliar veins, i.e., another indicator of drought tolerance of the plants (Scoffoni *et al.*, 2011). In our case, it was observed that soybean growing in DSCC plots in 2019 had a significantly wider midrib and a significantly lower DistVein, which represents a higher venation density. Higher venation density can be an indicator of willingness and resilience of the plants growing in plots with CC during high Vpd or drought episode (Carins Murphy *et al.*, 2014). A more elaborated venation may be linked to a better water management (Uhl et Mosbrugger, 1999). Scoffoni et al. (2011) have proposed that large midrib and small distance between secondary veins allows a more important number of stomata which is also consistent with our observations. Higher major vein density would thus have lower hydraulic vulnerability allowing a larger number of stomata (Scoffoni *et al.*, 2011). Also, the presence of CC can have a positive influence on soil functions, which can explain the willingness that facilitate phenologic plasticity of plants in DSCC plots. It has been shown that CC can increase the number and diversity of root systems in the field which can improve soil porosity, aggregation and fertility (Amsili et Kaye, 2021; Liu *et al.*, 2005). These soil functions can facilitate the accessibility and the uptake of water by crop plants, in turn favoring gas exchange and transpiration with less restriction.

Other factors resulting from the choice of a given agronomic management can influence the activity of stomata such as the use of herbicides in field crop (Albrecht *et al.*, 2014; Gomes *et al.*, 2014; Kim *et al.*, 2021; Krenchinski *et al.*, 2017; Smedbol *et al.*, 2019; Zobiole *et al.*, 2010). With the increase in the number of weeds resistant to GBH, producers are inclined to use heavier doses of the same herbicide or to combine different types of herbicides in order to efficiently control weeds (Gerhards et Schappert, 2020; Lemessa et Wakjira, 2015; Osipitan *et al.*, 2019). Moreover, this phenomenon of herbicide resistance is likely to be exacerbated with climate change, which is not without impact on cash crops (Fernando *et al.*, 2016). It has been shown that GBH can have a negative impact on the activity and development of stomata in glyphosate tolerant soybean (Albrecht *et al.*, 2014; Gomes *et al.*, 2014; Krenchinski *et al.*, 2017; Smedbol *et al.*, 2019; Zobiole *et al.*, 2010). Glyphosate tolerant soybeans are not resistant to aminomethylphosphonic acid (Kanissery *et al.*), the main degradation metabolite of glyphosate in soils (Gomes *et al.*, 2014; Reddy *et al.*, 2004; Zobiole *et al.*, 2010). Among the impacts considered, some authors have observed that stomatal conductance was lower following exposure of glyphosate tolerant soybeans to GBH (Bernier Brillon *et al.*, 2023; Bernier Brillon *et al.*, 2022; Smedbol *et al.*, 2019; Zobiole *et al.*, 2010). Other studies have also shown that the use of herbicides other than GBH could also affect stomatal activity and development (Anastasov, 2010a, 2010b; Chen *et al.*, 2016; Semerdjieva *et al.*, 2015). The response of certain crops such as soybean to herbicide applications can therefore induce stresses and decrease the crop tolerance to other disturbances such as water stress induced during a period of water limitation (Petter *et al.*, 2016). In this context, the use of CC can be beneficial considering that on one hand the presence of CC seems to positively influence plants to develop morphological traits making them more tolerant to drought and that on the other hand the use of CC seems to be effective in limiting the presence of weeds in the fields (Gerhards et Schappert, 2020; Osipitan *et al.*, 2019).

5. CONCLUSION

This study suggests that CC contribute to maintain gas exchange potential in a context of soybean exposed to higher Vpd values. The implementation of CC would thus favour a higher resilience to potential combined stress of drought and GBH application by increasing crop plasticity in GR soybean field crops. At similar Vpd values, the stomatal conductance of plants growing in DSCC plots was significantly higher than that of plants growing in DS plots. This can be explained by a higher tolerance under conditions that can cause water limitation to plants. This tolerance is expressed by a more elaborate venation and higher StoDen in plants growing in DSCC plots. Through the response of the plants and their development strategy, the benefits of CC on crops could be observed in the short term in this study, Finally, CC seem to represent, in part, a sustainable solution to fight against drought and future climate changes. CC also seem to be a promising alternative to minimize the reduction of gas exchange of soybean triggered by herbicides spraying during a drought period.

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

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