

RESEARCH ARTICLE

Response of Blackgram Cultivars to Elevated Tropospheric Ozone

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ABSTRACT

As a secondary pollutant, Tropospheric ozone is inadvertently increasing every year, thereby causing severe loss to agricultural crops. The present study aimed at evaluating the response of tropospheric ozone against blackgram varieties. Eight ruling blackgram varieties (V.B.N. 1, V.B.N. 2, V.B.N. 3, V.B.N. 5, V.B.N. 6, V.B.N. 7, V.B.N. 8 and CO 6) were exposed to elevated tropospheric ozone concentration (50 ppb) in an open-top chamber. The exposure was given during 31 days after sowing (D.A.S.) to 40 D.A.S. for seven hours (10.00 h-17.00 h). The changes in physiological, biochemical, growth, and yield traits were observed by comparing them with control (ambient condition). Results indicate that physiological, biochemical, growth, and yield traits significantly differed under ozone stress. Amongst all varieties, the reduction of all observed traits was higher in V.B.N. 3 and least in V.B.N. 8. The reduction of photosynthetic rate, stomatal conductance, and chlorophyll content was higher in V.B.N. 3 (33.57, 29.17 and 35.67 %) and least in V.B.N. 8 (26.23, 22.92 and 31.78 %). In the case of biochemical traits, in V.B.N. 3, the malondialdehyde and proline content increased twice and ascorbic acid declined by 39.85 %. However, in V.B.N. 8, malondialdehyde and proline content increased by 78.26 and 89.01 %; while ascorbic acid decreased by 36.31 % only. Similarly, 100-grain weight reduced in V.B.N. 3 by 8.69 % while it was only 5.37 % in V.B.N. 8. The current investigation revealed that V.B.N. 3 is highly sensitive, while V.B.N. 8 is tolerant to ozone stress.

Keywords: Tropospheric ozone; Blackgram; Physiological; Biochemical; Growth; Yield attributes

INTRODUCTION

Tropospheric ozone (O₃) has become one of the world's most widely dispersed toxic pollutants in the last several decades (I.P.C.C. 2007; Booker *et al.*, 2009; Brauer *et al.*, 2016), exerting severe impact on humans, plants, and animals (Cho *et al.*, 2011; Ainsworth, 2017; Agathokleous *et al.*, 2018; Osborne *et al.*, 2019). This secondary pollutant, under bright sunshine, is produced by a series of photochemical reactions involving carbon monoxide (CO), methane (CH₄), nitrogen oxides (NO_x), as well as volatile organic compounds (V.O.C.s) (Collins *et al.*, 1997; Monks *et al.*, 2015). Besides anthropogenic activities and other industrial emissions, and transportation also contributes to ozone production in metropolitan areas. Ground-level ozone is an issue in rural places as well. Although being hundreds or thousand miles away from the original source, higher concentration of tropospheric ozone is experienced in rural areas wherein a majority of land is devoted to agricultural operations (Prather *et al.*, 2003; Agrawal *et al.*, 2006; Williams *et al.*, 2016). In recent decades, the background O₃ concentration has risen

by between 0.5 and 2 percent every year (Vingarzan, 2004). The projected trends in ozone precursors emission increases the global average tropospheric ozone concentration by 20–25% between 2015 and 2050, further raising by 40–60% by 2100 (Meehl *et al.*, 2007). Crop yield losses are predicted throughout the world by the year 2030, and India would be suffering the worst situation when it comes to relative yield (Van Dingenen *et al.*, 2009). It has been estimated that current levels of O₃ can result in economic losses of \$ 14–26 billion, with China and India alone accounting for 40% (Van Dingenen *et al.*, 2009; Danh *et al.*, 2016; Ashrafuzzaman *et al.*, 2017; Harmens *et al.*, 2018; Mills *et al.*, 2018).

Ozone enters the leaf through stomatal pores and generates reactive oxygen species (ROS OS) as they reach the intercellular spaces, and exerts oxidative stress by reacting with plant's leaves (Booker *et al.*, 2009). So, the antioxidant defense system is activated, which plays a vital role in keeping ROS levels under control, thereby preserving the equilibrium of the cell's redox potential. Therefore, it is clear that O₃ stress results in decreased carbon

assimilation and altered carbon partitioning in plants (McCrary and Andersen, 2000), reduced yield, and altered crop quality (Singh *et al.*, 2009; Tripathi and Agrawal, 2012; Sarkar *et al.*, 2015). Several studies have been reported that tropospheric ozone significantly reduced the crop yield (Feng and Kobayashi, 2009; Sawada and Kohno, 2009; Rai *et al.*, 2010; Sarkar and Agrawal, 2010; Mishra *et al.*, 2013; Singh *et al.*, 2014; Ziemke *et al.*, 2019). Tropospheric ozone exhibited significant toxicity on various crops like snap bean (Flowers *et al.*, 2007), mung bean (Chaudhary and Agrawal, 2015), potato (Suganthi and Udayasoorian, 2016), garlic (Gayathri *et al.*, 2019), cauliflower (Sethupathi *et al.*, 2018), forests (Elvira *et al.*, 2004; Murugaragavan and Udayasoorian, 2016) and rice (Ramya *et al.*, 2021 a, b).

India, being one of the fastest expanding countries in terms of economy in the Asian region, endures a significant increase in tropospheric ozone concentration (Berntsen *et al.*, 1996; Brasseur *et al.*, 1998; Oksanen *et al.*, 2013). Agriculture in India is the country's most populous economic sector, and it ranks second in the world (Ghude *et al.*, 2014; Brauer *et al.*, 2016). Increasing levels of O₃ pollution are a serious concern for India's agriculture, which is home to a fifth of the world's hungry people. As time goes on, reports indicate that the ambient levels of O₃ are growing in the northern region of India (Pandey *et al.*, 1992; Agrawal *et al.*, 2002; Tiwari *et al.*, 2008; Sarkar and Agrawal 2010), which might significantly result in crop yield loss (Agrawal *et al.*, 2005; Singh and Agrawal, 2010; Agathokleous *et al.*, 2015; Sugai *et al.*, 2018). In Tamil Nadu, various regions like Chennai (Pulikesi *et al.*, 2006; Padma *et al.*, 2014; Muthulakshmi *et al.*, 2017; Prabakaran *et al.*, 2017; Mohan and Saranya, 2019), Ooty (Udayasoorian *et al.*, 2013), Suchindrum (Sharma *et al.*, 2012), Kanyakumari (Sharma and Nagaveena, 2016), Tirunelveli (Usha *et al.*, 2018) were reported to experience higher concentration of ground-level ozone.

Pulses, one of the vital agricultural crops, occupies 252.29 lakh hectares of agricultural land with a production of 16.47 million tonnes (D.A.C. & F.W., 2018). They are susceptible to tropospheric ozone, and a critical level of 40 ppb is required for 5% yield reduction (Mills *et al.*, 2018). Blackgram is one of the vital pulse crop containing 26% protein and plays a major role in the Indian diet. Studies using Open-top chambers (OTC) have been extensively studied over the last 50 years to determine the yield losses owing to different air pollutants, including O₃. This study investigates the response of O₃ against eight ruling blackgram varieties since it is one of the vital pulses in India and provides a major protein requirement in the human's diet.

MATERIAL AND METHODS

Study Location and plant materials

The study was conducted at wetland (11.00° N, 76.92° E at an elevation of 426.72 m amsl) of Tamil Nadu Agricultural University, During the experimental period, the maximum temperature ranged from 29.9 to 30.8 °C; while the minimum temperature varied between 17.5 and 18.2 °C. The maximum relative humidity was from 71 to 85% and the minimum relative humidity varied from 48 to 54%. The study area is located in the under semi – arid tropics. The soil was deep clay loam with taxonomical classification as Typic Haplustalf. The pH was found to be 8.05 with an E.C. of 0.39 dS m⁻¹. The available nitrogen was found to be low (217 kg ha⁻¹) with medium available phosphorus (11.8 kg ha⁻¹) and high available potassium (295 kg ha⁻¹). The soil had low organic carbon content (0.48%).

The study was performed on eight ruling blackgram varieties (V.B.N. 1, V.B.N. 2, V.B.N. 3, V.B.N. 5, V.B.N. 6, V.B.N. 7, V.B.N. 8, and CO6) which was obtained from National Pulses Research Centre situated at Vamban. Soil test crop response (S.T.C.R.) based fertilizer application was given with regular irrigation to maintain uniform soil moisture.

Experimental design

A pot experiment was conducted in two Open-Top Chamber (OTC) of standard size (3.5 × 3.5 m), which were installed in the experimental site and consisted of two treatments – Control – non-filtered chamber (N.F.C.) and N.F.C. + elevated O₃ (50 ppb). The elevated ozone was given using an ozone generator (A4G, Faraday, India). An ozone feed rate of 0.6 mg/min was maintained throughout the experimental period (Van Leeuwen, 2015) and the inlet oxygen flow was maintained at 12 L min⁻¹ to regulate the ozone flow. The plants were exposed to an elevated ozone concentration of 50 ppb in the open-top chamber during the flowering stage from 31 D.A.S. (days after sowing) to 40 D.A.S. for 7 hours (10.00 h – 17.00 h) with an AOT40 value of 0.714 ppm. h. The emission was given 30 cm above the canopy of the plant. In the control chamber, the ozone concentration was less than 7 ppb.

Plant sampling

Random plants were sampled in nine replications from each OTC for each blackgram variety (n = 8 variety × 9 replications) and analyzed. Sampling was done after 10 days of ozone exposure and was used to analyze all physiological and biochemical traits. Sampling was done during the harvest or crop maturity stage for growth and yield traits.

Plant analysis

Leaf injury percentage (L.I.P.) ranging from 0

to 100 was given to all the blackgram varieties (Chaudhary *et al.*, 2013). Physiological parameters like photosynthetic rate (A), stomatal conductance (gs), and chlorophyll content were measured at different points of the leaf during day light (09.30 AM to 12.00 PM) before and after 10 days of ozone exposure. The youngest leaf was chosen to measure the physiological traits. A portable photosynthetic system (A.D.C. Bio Scientific LCpro-SD System, U.K.) was used to measure A and gs; while chlorophyll content meter (CCM-200+, U.S.A.) was used to measure the chlorophyll content. Biochemical traits like malondialdehyde (MDA), proline, and ascorbic acid (AsA) were also analyzed. The standard protocol given by Heath and Packer (1968) was used to measure; while proline was measured using the procedure given by Bates *et al.* (1973) and for ascorbic acid (AsA) content the method given by Keller and Schwager (1977) was followed.

During the harvest stage, growth traits like root length (R.L.), shoot length (S.L.), and plant weight (P.W.), yield traits like number of branches per plant (N.B.P.), number of nodules per plant (N.N.P.), number of leaves per plant (N.L.P.), number of pods per plant (NPP), number of seeds per plant (N.S.P.), pod length (P.L.) and 100-grain weight (100 GW) were measured.

Statistical analysis

The significant difference amongst the cultivars and treatment were statistically evaluated using one-way ANOVA test, while their interactions were done by two-way ANOVA test. S.P.S.S. (Ver. 16.0.0), a statistical tool was used to perform the tests. The variation among the treatment means was studied using Tukey method and Pearson's correlation coefficient was used to determine the degree of correlation.

RESULTS AND DISCUSSION

Leaf injury percentage

Exposure to elevated ozone exhibited various leaf injury symptoms, and the leaf injury percentage (L.I.P.) varied among the cultivars. The symptoms were observed to intensify with an increasing exposure period. Younger leaves with early necrotic symptoms were identified in V.B.N. 1, V.B.N. 2, V.B.N. 3, and CO6, indicating its sensitivity towards ozone stress. The L.I.P. for the above-mentioned varieties was found to be 40.00, 40.00, 50.67 and 40.33%, respectively. Similarly, varieties like V.B.N. 5 (27.00%), V.B.N. 6 (34.00%), V.B.N. 7 (36.67%), and V.B.N. 8 (23.67%) exhibited lesser injury, validating their tolerance to ozone stress. Amongst all cultivars, the highest L.I.P. was observed in V.B.N. 3 (50.67%), while the least was observed in V.B.N. 8 (23.67%). Furthermore, cultivar-specific variation reported in

this study demonstrated that our test blackgram cultivars have varying resistance levels to high ozone exposure. This tendency might be explained by a more significant rise in reactive oxygen species (ROS) in comparison to an increase in ozone levels. According to Weadow *et al.* (2021), superoxide is produced under ozone exposure, causing leaf damage. Chaudhary *et al.* (2013), revealed that extended ozone exposure intensified the leaf injury in mung bean cultivars and that foliar L.I.P. can demonstrate different levels of ozone sensitivity of test cultivars. Similarly, Mishra and Agrawal (2015) found that ROS buildup inside the plant system caused foliar and cellular damage.

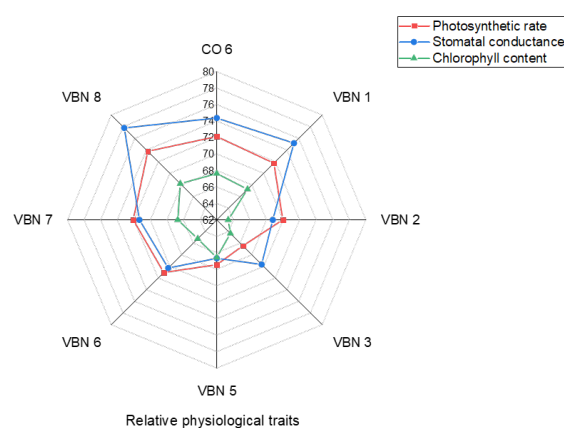


Figure 1 . Relative physiological traits of blackgram varieties under 50 ppb ozone stress

Physiological traits

Elevated ozone exposure significantly reduced the physiological traits like photosynthetic rate, stomatal conductance, and chlorophyll content in all blackgram varieties. The photosynthetic rate varied from 13.04 to 15.02 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$. Similarly, stomatal conductance varied between 0.33 to 0.39 $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and chlorophyll content between 18.34 and 20.67 (Table 1). V.B.N. 3 recorded the highest reduction in all physiological traits among the varieties, while V.B.N. 8 recorded the least reduction. This indicates that V.B.N. 3 is highly sensitive to ozone stress while V.B.N. 8 is tolerant. Like L.I.P., the varietal difference was also observed in physiological traits. Generally, the opening and closing of stomata regulate the entry of tropospheric ozone into the apoplast of the plant system (Tingey and Hogsett, 1985; Daszkowska-Golec and Szarejko, 2013; Rai, 2020). In this study, the reduction in photosynthetic rate and stomatal conductance is associated with the offset mechanism to prevent pollutant entry through stomatal closure (Fiscus *et al.*, 2005; Betzelberger *et al.*, 2010; Ghosh *et al.*, 2020).

Moreover, variation in the partial pressure of the guard cells and loss of osmotic potential might also lead to the closure of stomata under ozone

stress. With stomatal closure, the ability to uptake CO₂ declines, thereby decreasing the photosynthetic rate. Similar results were reported in mung bean (Mishra and Agrawal, 2015) and soybean (Sun *et al.*, 2014; Rai *et al.*, 2015; Ramya *et al.*, 2021 a,b). The decline in chlorophyll content due to ozone stress might be attributed to the destruction of the chloroplast structure, thereby suppressing chlorophyll synthesis (Castagna *et al.*, 2001; Biswas and Jiang, 2011; Jing *et al.*, 2016). Moreover, the decline in the carotenoid pigments might also reduce the chlorophyll content (Salvatori *et al.*, 2013). The results corroborate with studies given by Tetteh *et al.* (2016). The relative physiological characteristics of blackgram varieties varied among each other with a mean value of 70.80, 71.39, and 66.63% for stomatal conductance, photosynthetic rate, and chlorophyll content (Fig.1). The greater deviation was observed in stomatal conductance indicating

that stomatal conductance is highly responsive to ozone stress compared to the photosynthetic rate and chlorophyll content.

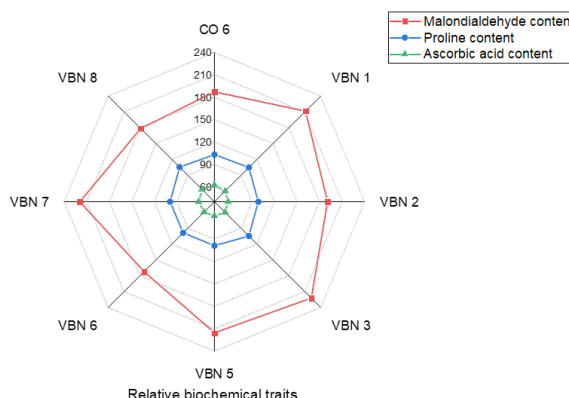


Figure 2. Relative biochemical traits of blackgram varieties under 50 ppb ozone stress

Biochemical traits

In all blackgram varieties, malondialdehyde content (MDA) significantly increased under ozone stress compared to control and a varietal variation was also observed. The MDA content ranged from 1.07 to 1.49 $\mu\text{mol g}^{-1}$ F.W. in control and from 2.27 to 2.97 $\mu\text{mol g}^{-1}$ F.W. under 50 ppb ozone stress. The entry of tropospheric ozone into the plant system induces the generation of reactive oxygen species (ROS), thereby damaging the membrane components like lipids, chloroplast, nucleic acids, and proteins (Blokhina *et al.*, 2003; Hasanuzzaman *et al.*, 2012; Saxena *et al.*, 2019). The induction in

MDA content under ozone stress is related to its sensitivity suggesting greater lipid peroxidation of the membrane compared to ambient conditions. Furthermore, the destruction of membrane components due to ROS generation inhibits the scavenging ability of the plant cell (Sanmartin *et al.*, 2003). The results corroborate with the findings of Mishra and Agrawal (2015) observed a 30.8 and 21% increase in MDA content of mung bean cultivars under 68.9 ppb ozone stress. Significant reduction in biochemical traits was also observed in cauliflower (Sethupathi *et al.*, 2018). garlic (Gayathri *et al.*, 2019) and rice (Ramya *et al.*, 2021a).

Table 1. Physiological traits of blackgram varieties under 50 ppb ozone stress

Variety	Photosynthetic rate		Stomatal conductance		Chlorophyll content	
	Control	50 ppb	Control	50 ppb	Control	50 ppb
CO 6	18.48±0.37	13.33±0.22	0.52±0.04	0.39±0.01	28.57±0.70	19.32±0.70
V.B.N 1	20.52±0.28	14.72±0.44	0.51±0.04	0.38±0.01	29.37±0.60	19.75±0.87
V.B.N 2	18.79±0.67	13.15±0.85	0.48±0.01	0.33±0.02	28.93±0.85	18.34±0.35
V.B.N 3	19.63±0.55	13.04±0.47	0.48±0.01	0.34±0.02	29.83±0.90	19.19±0.55
V.B.N 5	20.16±0.35	13.60±0.13	0.49±0.01	0.33±0.01	29.00±1.00	19.30±0.66
V.B.N 6	21.15±0.33	15.02±0.27	0.49±0.01	0.35±0.02	29.83±0.96	19.47±0.84
V.B.N 7	19.02±0.35	13.71±0.33	0.50±0.01	0.36±0.03	29.23±0.33	19.50±0.70
V.B.N 8	18.53±0.68	13.67±0.71	0.48±0.01	0.37±0.01	30.30±0.50	20.67±0.55
	ANOVA (P values)		Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	Stomatal conductance ($\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$)	Chlorophyll content	
Varieties			<0.001	0.064	0.340	
Treatment			<0.001	<0.001	<0.001	
Varieties ×Treatment			0.520	0.821	0.964	
Treatment means						
Control			18.53	0.49	29.38	
Ozone			13.67	0.36	19.44	

n=9 (Sample size, N=144)

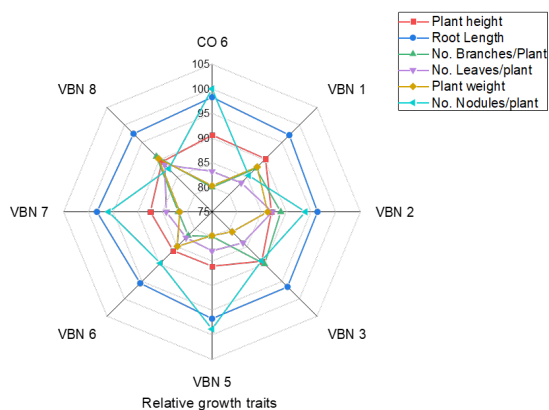


Figure 3. Relative growth traits of blackgram varieties under 50 ppb ozone stress

Unlike MDA content, the ascorbic acid content was found to decline in all blackgram cultivars under ozone stress compared to control. The ascorbic acid varied between 1.33 to 1.72 mg g⁻¹ F.W. in control and between 0.78 and 1.03 mg g⁻¹ F.W. in elevated ozone condition. This decline might be due to the non-enzymatic defense mechanism of blackgram varieties under ozone stress. Antioxidants are produced to nullify the ROS toxicity (Caregnato *et al.*, 2013) and the redox condition of AsA becomes unstable, eventually resulting in insufficient detoxification by AsA (Tetteh *et al.*, 2016). The proline content increased under ozone stress varying from 4.81 to 6.08 μmol g⁻¹ F.W. under ambient conditions and from 10.15 to 13.46 μmol g⁻¹ F.W. under 50 ppb ozone stress. This increased proline content might

be attributed due to the scavenging ability of proline under ozone stress (Gill and Tuteja, 2010; Rejeb *et al.*, 2014). The relative biochemical traits showed variation with mean values of 201.01, 60.11 and 202.00% in MDA, AsA and proline content (Fig.2).

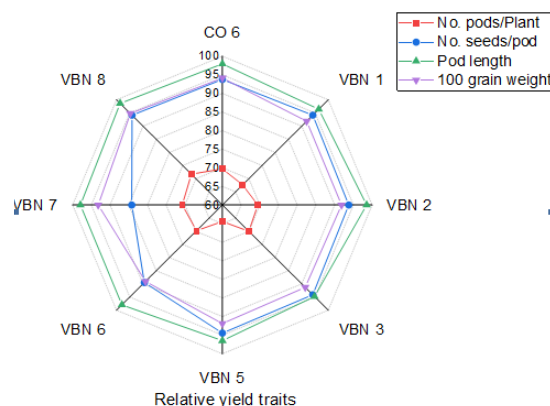


Figure 4. Relative yield traits of blackgram varieties under 50 ppb ozone stress

Growth and yield characteristics

Elevated ozone stress significantly decreased all the growth and yield traits in all blackgram cultivars. The shoot length varied from 20.67 to 37.27 cm (ambient) and from 17.80 to 33.30 cm (50 ppb ozone stress); while root length ranged between 13.47 and 16.20 cm under control and from 13.00 to 16.57 cm under ozone stress. In the case of number of branches per plant, under ozone stress, the highest value was observed in V.B.N. 8 (3.33) while the least was observed in V.B.N. 1 (2.33).

Table 2. Correlation between physiological, biochemical, growth and yield parameters

Parameters	LIP	100 GW	PL	NSP	NPP	PW	NLP	NBP	NNP	RL	SL	AsA	Proline	MDA	Chl	Gs	A
LIP	1																
100 GW	-0.57	1															
PL	-0.28	0.34	1														
NSP	-0.25	0.11	-0.29	1													
NPP	-0.61	0.31	0.42	0.35	1												
PW	-0.31	0.12	0.44	0.59	0.37	1											
NLP	-0.52	0.54	0.51	0.19	0.50	0.64	1										
NBP	-0.61	0.43	0.42	0.40	0.42	0.65	0.70	1									
NNP	0.47	0.47	0.09	-0.27	-0.01	0.02	-0.18	-0.36	1								
RL	-0.53	0.85	0.69	0.53	0.24	0.19	0.11	0.21	0.75	1							
PL	-0.52	0.57	0.56	0.36	0.33	0.69	0.11	0.33	0.53	0.57	1						
AsA	-0.65	0.77	0.29	0.56	0.58	0.25	0.35	0.14	0.36	0.67	0.65	1					
Proline	0.58	0.42	-0.34	-0.48	-0.30	-0.33	-0.19	-0.41	0.23	-0.14	-0.23	-0.52	1				
MDA	0.59	0	-0.73	-0.11	-0.40	-0.50	-0.31	-0.01	0.27	0.24	0.05	-0.28	0.49	1			
Chl	-0.57	0.65	0.21	0.34	0.52	0.34	0.36	0.50	0.66	0.67	0.48	0.76	0.45	-0.61	1		
Gs	-0.60	0.36	0.33	0.10	0.12	0.21	0.37	0.33	0.51	0.50	0.23	0.34	-0.73	-0.36	0.71	1	
A	-0.61	0.51	0.76	0.62	0.54	0.59	0.37	0.55	0.54	0.47	0.35	0.65	-0.19	-0.56	0.63	0.81	1

The number of nodules per plant ranged between 92.50 and 100. Similarly, V.B.N. 8 recorded the highest number of pods per plant (11.00), number of seeds per pod (5.67), pod length (4.83 cm), number of leaves per plant (59.00), 100-grain weight (4.23 g), and plant weight (18.60 g). The reduction was relatively higher in V.B.N. 3 in most growth and yield traits, signifying its sensitivity to ozone stress. Reduced photosynthetic capacity under elevated ozone conditions might, in turn, decrease the plant biomass by altering the allocation of photosynthates to several parts of the plant (Sarkar and Agrawal, 2010; Feng *et al.*, 2011; Chaudhary *et al.*, 2013; Ruiz-Vera *et al.*, 2017; Ghosh *et al.*, 2020). Similar results were observed in mung bean where the plant height and number of leaves per plant decreased by 25.7 and 24% under 70.9 ppb ozone stress (Chaudhary and Agarwal, 2015). The reduction in yield traits might be due to extended closure of stomata and decline in carbon fixation thereby reducing the availability of assimilates to its reproductive parts. Alterations in physiological and biochemical characteristics under ozone stress might in turn might have altered the growth and yield traits in blackgram varieties. V.B.N. 3 and V.B.N. 1 exhibited senescence due to ozone stress which might be ascribed due to the induction of genes associated with senescence (Miller *et al.*, 1999). Similar results were also observed by Chaudhary and Agarwal (2015) in mung bean and by Ghosh *et al.* (2020) in maize. The relative root, plant length, number of nodules per plant, number of branches per plant, number of leaves per plant, and plant weight had a mean value of 88.32, 96.89, 92.50, 84.66, 83.60, and 83.23% (Fig.3). The relative number of pods per plant, number of seeds per pod, pod length, and 100-grain weight had a mean value of 69.73, 93.93, 97.93, and 91.85%, respectively (Fig.4). The Pearson's correlation depicted a negative correlation between leaf injury percentage and other characteristics (Table 2). Similarly, a positive correlation was observed between stomatal conductance and photosynthetic rate, between chlorophyll content and photosynthetic rate, and between chlorophyll content and stomatal conductance. Likewise, linear relationship was observed between photosynthetic rate and growth traits (number of seeds per plant and pod length).

CONCLUSION

Understanding the influence of elevated tropospheric ozone on blackgram is highly imperative for global food security. The current study exhibited varietal variation among blackgram cultivars to ozone stress. Results indicate that elevated ozone concentration increased the leaf injury percentage, while all physiological traits like photosynthetic rate, stomatal conductance, and chlorophyll content

declined significantly. Similarly, the malondialdehyde and proline content increased, while ascorbic acid content decreased under ozone stress. Significant reduction in the growth and yield traits were also observed in all blackgram varieties under ozone stress. This indicates that, among the blackgram varieties under study, V.B.N. 3 is sensitive and V.B.N. 8 is tolerant to ozone stress. Hence, this finding serves as a preliminary base to evaluate and assess the choice of cultivars to regions experiencing high tropospheric ozone concentration and for future breeding programmes. Varietal-specific alterations in plant attributes under ozone stress might pave the way for developing ozone-tolerant blackgram cultivars using genome mapping and quantitative trait loci (QTL)-based approaches, thereby boosting global production. Significant genetic heterogeneity in ozone sensitivity and tolerance across test cultivars might be helpful in creating models to anticipate ozone-induced yield loss and adaptation techniques for long-term cultivation.

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Ethics statement

No specific permits were required for the described field studies because no human or animal subjects were involved in this research.

Originality and plagiarism

Authors ensure the originality of the manuscript and the work and/or words of others, has been appropriately cited. We acknowledge that plagiarism in all its forms constitutes unethical publishing behavior and is unacceptable.

Consent for publication

All the authors agreed to publish the content.

Competing interests

There were no conflict of interest in the publication of this content

Data availability

All the data of this manuscript are included in the M.S. No separate external data source is required.

Author contributions

Idea conceptualization - Periyasamy Dhevagi, Experiments- Periyasamy Dhevagi, Ramya Ambikapathi ,Guidance -Periyasamy Dhevagi, Writing original draft – Poornima Ramesh, Ramya Ambikapathi, Writing- reviewing & editing - Periyasamy

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