



EFFECT OF ELEVATED TROPOSPHERIC OZONE ON PHOTOSYNTHETIC EFFICIENCY OF RICE CULTIVARS

C. Haritha Gowri¹, P. Vijaya Priya¹, A.Ramya² and P.Dhevagi^{3*}

¹Undergraduate Project students, ²Research Scholar, ³Associate Professor, Department of Environmental Science, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu- 641 003

*Corresponding author email: devagisivaraj@gmail.com

ABSTRACT

Tropospheric ozone (O₃) is an important secondary air pollutant and its concentration has mounted from approximately 10 ppb in the late 1800s to monthly average daytime concentrations exceeding 40–50 ppb in these days. In India, 8 h daily average concentrations of 100 ppb tropospheric ozone have been reported during the spring crop growing season. Rice an important food crop is known to be susceptible to tropospheric ozone, particularly in the Asian region. The current study focused on the effect of elevated ozone on photosynthetic efficiency of rice (*Oryza sativa* L.) cultivars namely CO 47, ADT 36, ADT 37, ADT 43, ADT(R) 45, ADT(R) 48, ASD 18, MDU 5 and Anna(R)4. The selected varieties were exposed to ozone level of 50 ppb in open top chamber and the same varieties incubated under ambient environmental condition were treated as control. The results showed that 50 ppb ozone caused significant reduction in photosynthesis rate and stomatal conductance of all rice cultivars. Lower reduction of both photosynthesis rate and stomatal conductance was observed in Anna(R) and higher reduction was in case of MDU5.

KEYWORDS: Tropospheric ozone, Rice cultivars, Photosynthetic rate, Stomatal conductance

INTRODUCTION

The tropospheric ozone, an important greenhouse gas, plays critical role in determining the oxidizing capacity of the troposphere. The O₃ distribution in the troposphere is strongly influenced by dynamical processes, as well as by the regional chemical sources and sinks of O₃ (Vogel *et al.*, 2014). The inhibitory actions of tropospheric ozone on crop photosynthesis, growth and yield have been documented in numerous studies (Feng *et al.*, 2019). Ozone enter into leaf intercellular space rapidly reacts with components of leaf apoplast to intimate a complex set of responses involving the formation of toxic metabolites and generation of plant defense responses that constitute effective counter measures (Daripa *et al.*, 2016). Ozone clearly impairs photosynthesis processes, which might include the effects on electron transport and guard cell homeostasis as well as carbon fixation via decreased Rubisco activity. Translocation of photosynthate could be inhibited by ozone exposure as well (Fiscus *et al.*, 2005). Rice accounts for a significant contribution to the total food grain production in India. It accounts nearly 43.86 million hectare area in India and it acts as the staple food nearly 75% of the people (Wanjari *et al.*, 2006). O₃ induced damage to rice in India was estimated to be 2.1±0.8 mt, which accounts for 6% yield loss (Ghude *et al.*, 2014). It is important to know about the changes that are produced in rice crop due to the effect of elevated tropospheric ozone to reduce the ozone induced yield loss. Hence the present study was carried out with a view to find out the effect of elevated tropospheric ozone on photosynthetic rate and stomatal conductance of rice plant.

MATERIALS AND METHODS

The experimental study on the effect of tropospheric ozone on photosynthesis of rice plant was carried out in the Department of Environmental science, AC & RI, Coimbatore. Short duration rice cultivars were chosen (CO47, ADT 36, ADT 37 ADT 43, ADT(R)45, ADT(R)48, ASD 18, MDU 5 and Anna(R)4) for the experimental study and seeds were obtained from various rice research stations of TNAU. The soil, compost and water used for irrigation were analyzed as per the standard methods (Walker and Black, 1934; Stanford and English, 1948; Olsen *et al.*, 1954; Subbiah and Asija, 1956; Piper, 1966; Jackson, 1973; APHA, 1980 and Gupta, 2002). Screening for elevated tropospheric ozone tolerance in rice genotypes was done in open top chamber (OTC) with the help of O₃ generator after 50 days of sowing to 80 days of sowing (30 days). Plants were receiving average of 50 ppb O₃, 7 h per day, during day time from 10.00 to 17.00 h of the local time. The concentration of O₃ was monitored with the help of ambient ozone monitor (G09-O₃-3121). The control plants with 3 replications were maintained without ozone exposure. The statistical design adopted as factorial completely randomized block design (FCRD).

Measurement of photosynthetic rate and stomatal conductance

A non-destructive method of photosynthetic rate and stomatal conductance estimation was done using portable photosynthetic system (PPS, model ADC bioscientific LCpro+, UK). The PPS measures the uptake of CO₂ and expressed as micro mol CO₂ m⁻²s⁻¹ and stomatal conductance are expressed as mol H₂O m⁻²s⁻¹. The Photosynthetic rate and stomatal conductance of the nine rice cultivars has been estimated before O₃ exposure (50

DAS), 15th day after O₃ exposure (65 DAS) and 30th day after O₃ exposure (80 DAS).

RESULTS AND DISCUSSION

Plants were exposed with ozone (50 ppb) for duration of 30 days starting from 50 DAS. The exposure was given for about 7 h per day, during day time from 10.00 to 17.00 h of the local time. The changes in photosynthetic rate and stomatal conductance were analyzed 15 days interval after ozone exposure.

Tropospheric ozone is one of the major air pollutants and is a serious threat to crops, forest, and natural vegetation. Present experiment was designed to evaluate the impact of ambient and elevated concentrations of tropospheric ozone on highly popular rice cultivars grown in Tamil Nadu under near natural conditions using OTC. The soil, water and compost sample used for the study were analyzed and to find its suitability for growing rice cultivars.

Photosynthetic rate of rice cultivars

In general, tropospheric ozone severely affects the growth and development of plants. Tropospheric ozone and their generated ROS are known to alter membrane properties and membrane bound organelles like chloroplast, which

lead to destruction of photosynthetic pigments (Rai *et al.*, 2012 and Tiwari *et al.*, 2016). Destruction of photosynthetic pigments also reduces photosynthetic activity.

Photosynthetic rate of nine rice cultivars under ambient condition (control) 50 DAS, 65 DAS and 75 DAS ranged from 18.60 to 22.39 micro m CO₂ m⁻² S⁻¹, from 20.85 to 24.57 micro m CO₂ m⁻² S⁻¹ and 21.35 to 25.53 micro m CO₂ m⁻² S⁻¹ respectively. Among the nine cultivars, highest activity was observed in Anna(R) 4 during the study period.

In case of treated plants, during the exposure of 50 ppb ozone (65DAS), the photosynthetic activity ranged between 17.79 and 23.08 micro m CO₂ m⁻² S⁻¹. The highest activity was registered in the rice cultivar Anna(R) 4 (23.08 micro m CO₂ m⁻² S⁻¹) followed by ADT 37 (21.98 micro m CO₂ m⁻² S⁻¹) and the lowest activity was reported by ASD18 (17.79 micro m CO₂ m⁻² S⁻¹). After thirty days of exposure (80DAS), the highest photosynthetic rate was registered in the rice cultivar Anna(R) 4 (22.28 micro m CO₂ m⁻² S⁻¹) followed by CO 47 (20.37 micro m CO₂ m⁻² S⁻¹) and lowest activity was reported in ADT 43 (16.80 micro m CO₂ m⁻² S⁻¹) (Table 1 and Figure 1).

TABLE 1: Photosynthetic rate of rice cultivars at different growth stages

Cultivars	Photosynthetic rate (micro m CO ₂ m ⁻² S ⁻¹)					
	Before ozone exposure (50 DAS)		During ozone exposure (65 DAS)		After ozone exposure (80 DAS)	
	Control	Treatment	Control	Treatment	Control	Treatment
CO 47	18.60	18.98	22.78	21.04	24.86	20.37
ADT 36	20.70	21.42	22.80	20.01	25.05	19.93
ADT 37	20.13	20.13	22.81	21.98	23.58	19.58
ADT 43	21.79	22.39	21.44	19.10	22.31	16.80
ADT(R) 45	20.98	21.55	20.21	19.12	22.08	17.95
ADT(R) 48	20.19	19.51	22.31	20.46	23.22	18.47
ASD 18	20.46	19.72	21.88	17.79	21.35	18.34
MDU 5	20.20	20.43	20.85	18.37	22.59	17.97
Anna(R) 4	21.56	20.94	24.57	23.08	25.53	22.28
Mean	20.51	20.56	22.18	20.11	23.40	19.08
SED	0.66	0.68	0.59	0.59	0.93	0.89
CD 5%	1.34	1.38	1.21	1.20	1.91	1.82

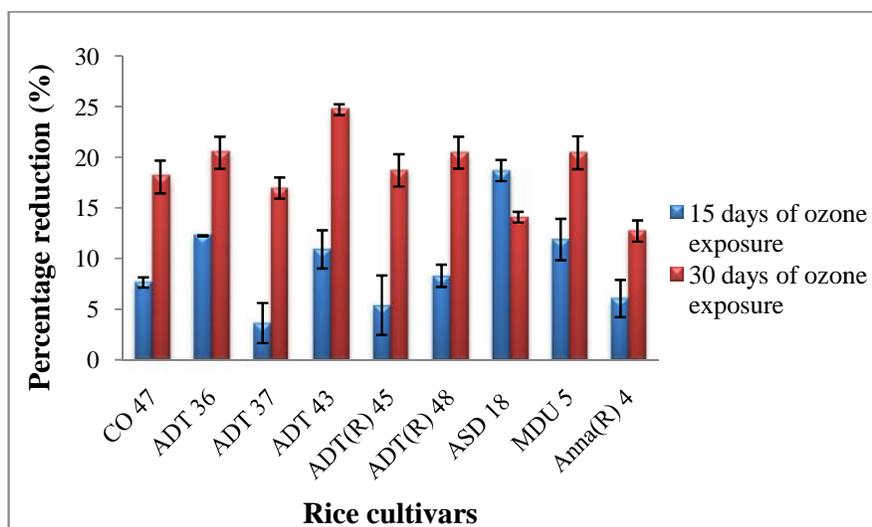


FIGURE 1. Percentage reduction in photosynthetic rate of rice cultivars

Photosynthesis, a core function in the physiology of plants is most susceptible to ozone. Reductions in photosynthesis have been widely reported under ambient field conditions at higher concentration of ozone (Rai *et al.*, 2007 and Tiwari *et al.*, 2006). In meta-analyses on several varieties of wheat (Feng *et al.*, 2008), soybean (Morgan *et al.*, 2003), wheat (Biswas *et al.*, 2008) and rice (Ainsworth, 2008) also found varying degrees of response of photosynthesis to ozone. It was observed that the reduction in mean photosynthetic rate was recorded with increasing days of ozone exposure when compared to control.

The results coincides with findings of Issii *et al.* (2004) who reported that Rice cv MR 84 reduced photosynthetic rate up to 53.3 per cent under elevated ozone level of 81.7 ppb for 8 hours. Rai and Agrawal (2008) revealed that Rice cv Saurabh 950 reduced photosynthetic rate up to 28.3 per cent under 35 ppb ozone for 12 hours. Ainsworth (2008) and Sarkar and Agrawal (2012) reported that Indian rice cultivars namely Shivani and Malviyadhan 36 reduced the photosynthetic rate under elevated ozone exposure up to 59.9 ppb for 6 hours.

In the present study also, among the cultivars, ADT 43, MDU5 and ADT (R) 48 showed higher reduction in photosynthetic rate and Anna(R) 4, ASD 18 and ADT 37 showed lower reduction. The exposure of plants to ozone resulted in the accumulation of carbohydrate in the source leaves and reduced translocation to distant sink (Grantz and Yang, 2000). The pattern of photosynthate allocation directly affects the plant growth and reproduction. Ozone exposure reduced the available carbohydrates to roots in rice (Chen *et al.*, 2011), soybean (Morgan *et al.*, 2003), wheat (Biswas *et al.*, 2008), cotton (Grantz and yang,

2000) and a number of crops (Cooley and Manning, 1987; Andersen, 2003).

Stomatal conductance of rice cultivars

Stomatal uptake is the primary passage through which ozone enters the leaf intercellular spaces and generates physiological and oxidative damage in plant cells (Beauchamp *et al.*, 2005; Gerosa *et al.*, 2007; Fares *et al.*, 2008). Apart from alteration of basic metabolic processes such as photosynthesis and plant growth, ozone exposure leads to major changes in plant volatile emission rates and emission profiles during the initial stress impact and through recovery. Generally, the stomatal control of ozone uptake provides a coupling between environmental conditions and ozone uptake as well as a potential negative feedback to ozone uptake itself.

In control plants, the stomatal conductance of nine rice cultivars ranged from 0.33 (CO47) to 0.62 (ADT43) mol H₂O m⁻² S⁻¹. At 65 DAS, it ranged from 0.43 (MDU 5) to 0.55 (ADT36) mol H₂O m⁻² S⁻¹ and at 80 DAS 0.53 (MDU 5, ADT 37 and ADT(R) 48) to 0.64 (ADT36) mol H₂O m⁻² S⁻¹. However in treated plants, during 15 days of exposure with 50 ppb ozone, the photosynthetic activity ranged between 0.38 and 0.48 mol H₂O m⁻² S⁻¹. The highest activity was registered in the rice cultivar ADT 36 (0.48 mol H₂O m⁻² S⁻¹) and the lowest activity was reported in MDU 5 (0.38 mol H₂O m⁻² S⁻¹). After 30 of exposure, at 80 DAS the stomatal conductance recorded lowest of 0.39 mol H₂O m⁻² S⁻¹ (MDU 5 and ADT(R) 48) and highest was observed in Anna (R)4 (0.53 mol H₂O m⁻² S⁻¹).

TABLE 2. Stomatal conductance of rice cultivars at different growth stages

Cultivars	Stomatal conductance (mol H ₂ O m ⁻² S ⁻¹)					
	Before ozone exposure (50 DAS)		During ozone exposure (65 DAS)		After ozone exposure (80 DAS)	
	Control	Treatment	Control	Treatment	Control	Treatment
CO 47	0.33	0.36	0.52	0.46	0.59	0.47
ADT 36	0.50	0.49	0.55	0.48	0.64	0.49
ADT 37	0.50	0.45	0.48	0.41	0.53	0.40
ADT 43	0.62	0.62	0.49	0.43	0.59	0.44
ADT(R) 45	0.55	0.56	0.49	0.43	0.57	0.47
ADT(R) 48	0.38	0.41	0.47	0.41	0.53	0.39
ASD 18	0.48	0.51	0.53	0.46	0.55	0.42
MDU 5	0.37	0.43	0.43	0.38	0.53	0.39
Anna(R) 4	0.56	0.55	0.51	0.46	0.58	0.53
Mean	0.48	0.49	0.50	0.44	0.57	0.44
SED	0.03	0.03	0.03	0.03	0.03	0.03
CD 5%	0.07	0.07	0.06	0.06	0.07	0.06

The stomatal conductance is the most important regulator of ozone uptake under a given external concentration (Taylor and Hanson 1992; and Munger *et al.*, 1996). The result from the current study conducted to evaluate the effects of ozone on the physiological processes, have shown a decrease in the stomatal conductance. Ozone enters into the leaf through stomata and reacts with the appoplast mesophyll cells, generating reactive oxygen species and provoking signaling cascades, which cause

visible foliar injury, decrease stomatal conductance, and inhibit net photosynthetic rate (Morgan *et al.*, 2003). Reduction in the stomatal conductance, net photosynthetic CO₂ assimilation and carboxylation efficiency have all been associated with ozone exposure (Guidi *et al.*, 2001 and Morgan *et al.*, 2003). It was observed that the reduction in mean in the stomatal conductance was recorded with increasing days of ozone exposure when compared to control.

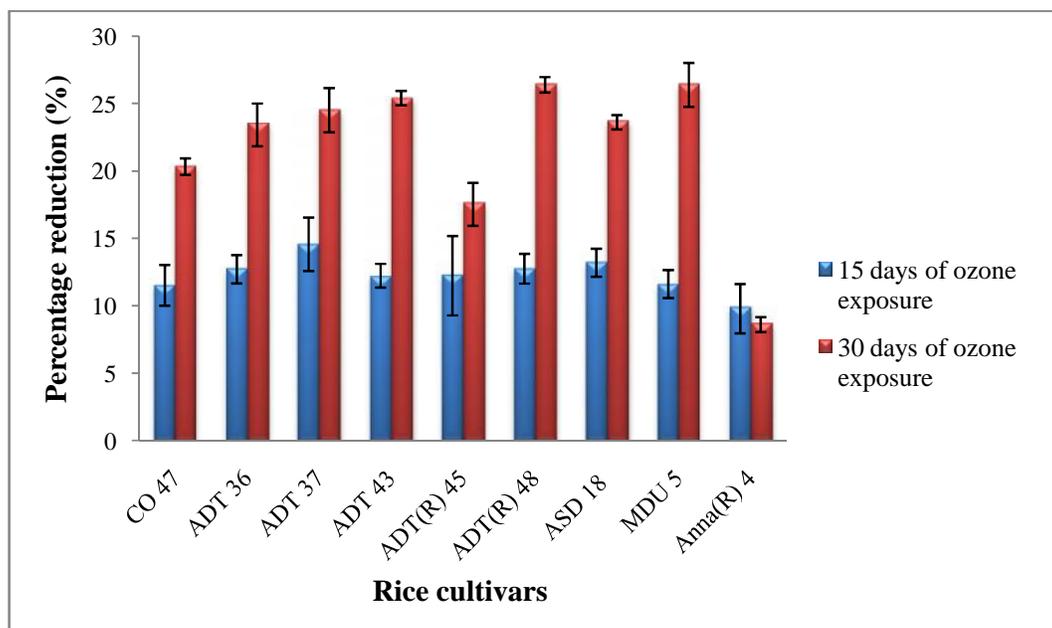


FIGURE 2. Percentage reduction in stomatal conductance of rice cultivars

The results are in line with the findings of Ainsworth (2008) and Sarkar and Agrawal (2012) who reported that Indian rice cultivars namely Shivani and Malviyadhan 36 reduced the stomatal conductance under elevated ozone exposure up to 59.9 ppb for 6 hours. Indian Rice cv Saurabh 950 reduced stomatal conductance up to 36.6 per cent under 35 ppb ozone for 12 hours (Rai and Agrawal, 2008).

Among nine rice cultivars, MDU 5, ADT(R) 48 and ADT 43 showed higher reduction in stomatal conductance and Anna(R) 4, ADT(R) 45 and CO 47 showed lower reduction. Current investigations clearly revealed a negative effect of ozone on plant photosynthetic efficiency. The decrease of photosynthesis mainly connected with stomatal closure. These relationships can vary among nine rice cultivars, although much of this variation is related to differences in photosynthetic rate and stomatal conductance.

CONCLUSION

Reductions in photosynthetic efficiency are one of the fundamental parameters towards ozone stress. The present study is also an evident for the effect of elevated levels of ozone on rice cultivars. The cultivar Anna(R)4 showed less reduction in photosynthetic rate and stomatal conductance, but few rice cultivars showed significant reduction under 50 ppb elevated ozone condition. This clearly pointed the differential cultivar response of rice against ozone and might be used for selecting suitable cultivars for an area experiencing higher concentrations of ozone.

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REFERENCES

- Ainsworth, E.A., Yendrek, C.R., Stith, S., Collins, W.J. and Emberson, L.D. (2008) The Effects of Tropospheric Ozone on Net Primary Productivity and Implications for Climate Change. *Annu Rev Plant Biol.* 63(1), 637-661.
- Anderson, C.P. (2003) Source –sink balance and carbon allocation below ground in plants exposed to ozone. *New Phytol.* 157(2), 213-228.
- APHA-American Public Health Association. (1999) Standard book for analysis of water and waste water 20th edition.
- Beauchamp, J., Withaler, A. Hansel, E. Kleist and Miebach, M. (2005) Ozone induced emissions of biogenic VOC from tobacco: relationships between ozone uptake and emissions of LOX products. *Plant, Cell Environ.* 28(10), 1334-1343.
- Biswas, D.K., Xu, H. LiSun, Y.G. (2008) Genotypic differences in leaf biochemical, physiological and growth responses to ozone in 20 wheat cultivars related over the past 60 years. *Glob. Change. Boil.* 46-59.
- Chen, C., Arjomandi, M., Balmes, J., Tager, I.B. and Holland, N. (2011) Effect of chronic and acute ozone exposure on lipid peroxidase and antioxidant capacity in healthy young adults. *Environ Health Perspect.* 115, 1732-7.
- Cooley, D.R. and Manning, W.J. (1987). The impact of ozone on assimilate partitioning in plants: A review. *Environ. Pollut.* 47(2), 95-113.
- Daripa, A., Bhatia, A., Ojha, S., Tomer, R., Chattaraj, S., Singh, K. P. and Singh, S. D. (2016). Chemical and natural plant extract in ameliorating negative impact of tropospheric ozone on wheat crop: a case study in a part of semiarid north west india. *Aerosol and Air Quality Research.* 16(7), 1742-1756.
- Fiscus, E.L., Booker, F.L. and Burkey, K.O. (2005) Crop responses to ozone: uptake, modes of action, carbon

- assimilation and partitioning. *Plant Cell Environ.* 28, 997-1011.
- Feng, Y., Komatsu, S., Furukawa, T., Koshiba, T. and Kohno, Y. (2008) Proteome analysis of proteins responsive to ambient and elevated ozone in rice seedlings. *Agric. Ecosyst. Environ.* 125, 255-265.
- Feng, Z., De Marco, A., Anav, A., Gualtieri, M., Sicard, P., Tian, H., Fornasier, F., Tao, F., Guo, A. and Paoletti, E. (2019) Economic losses due to ozone impacts on human health, forest productivity and crop yield across China. *Environ. Int.* 131(1), 104966.
- Fares, S., Loreto, F., Kleist, E. and Wildt, J. (2008). Stomatal uptake and stomatal deposition of ozone in isoprene and monoterpene emitting plants. *Plant Biol.*, 9(1), 69-78.
- Guidi, L., Nali, C. Lorenzini, G., Filippi, F. and Soldatini, G.F. (2001) Effect of chronic ozone fumigation on the photosynthetic process of poplar clones showing different sensitivity. *Environ. Pollut.* 113(3), 245-254.
- Ghude, S. D., Jena, C., Chate, D., Beig, G., Pfister, G., Kumar, R. and Ramanathan, V. (2014). Reductions in India's crop yield due to ozone. *Geo. Res. Let.*, 41(15), 5685-5691.
- Gupta, P. K. (2002) *Methods in Environmental Analysis: Water, Soil and Air.* Updesh Puro hit for Agrobios, Jodhpur, India.
- Grantz, D.A and Yang, S. (2000) Ozone impacts on allometry and root hydraulic conductance are not mediated by source limitation nor developmental age. *J. Exp. Bot.* 51(346), 919-927.
- Gerosa, G., Ferretti, M., Bussotti, F. and Rocchini, D. (2007) Estimates of ozone AOT40 from passive sampling in forest sites in south-western Europe. *Environ. Pollut.* 145(3), 629-635.
- Issii, S., Marshal, F.M. and Bell, J.N.B. (2004) Physiological and morphological responses of locally grown Malaysian rice cultivars to different ozone concentrations. *Water, air and soil pollut.* 205-221.
- Jackson, M. L. (1973) *Soil and Chemical Analysis.* Prentice Hall of India (Pvt.) Ltd., New Delhi.
- Morgan, P.B., and Bolleroo, T.A. (2003) Season long elevation of ozone concentration to projected 2050 levels under fully opened air conditions substantially decreases the growth and production of soybean. *New Phytol.* 170(2), 333-343.
- Munger, W.J., Michael, L. Song-Miao, F., Bruce, C. D. and Steven, C. W. (1996) Measurement of carbon sequestration by long-term eddy covariance: methods and a critical evaluation of accuracy. *Glob. Chan. Boil.* 2, 33.
- Olsen, S. R., Cole, L. L., Watanabe, F. S. and Dean, D. A. (1954) Estimation of available phosphorus in soils by extraction with sodium bicarbonate. *U. S. D. A.* 939.
- Piper, C. S. (1966) *Soil and Plant Analysis,* Inter Science Publications, New York, pp. 368.
- Rai, R., Agrawal, M., Agrawal, S B. (2007) Assessment of yield losses in tropical wheat using open top chamber. *Atmos Environ.* 41, 9543-54.
- Rai, R. and Agrawal, M. (2008) Evaluation of physiological and biochemical responses of two rice (*Oryza sativa L.*) cultivars to ambient air pollution using open top chambers at a rural site in India. *Sci. Total Environ.* 407, 679-691.
- Rai, R., Agrawal, M., Agrawal, S.B. (2012) Threat to food security under current levels of ground level ozone: a case study for Indian cultivars of rice. *Atmos. Environ.* 44, 4272-4282.
- Sarkar, A, Agrawal, S.B. (2012) Evaluating the response of two high yielding Indian rice cultivars against ambient and elevated levels of ozone by using open top chambers. *J Environ Manage.* 95, 19-24.
- Subbiah, B. V. and Asija, G. L. (1956) A rapid procedure for estimation of available nitrogen in soils. *Curr. Sci.* 25(8), 259-260.
- Stanford, S. and English, L. (1948) Use of flame photometer in rapid soil tests of K and Ca. *Agron. J.*, 41(9), 446-447.
- Tiwari, S., Agarwal, M. and Marshall, F.M. (2006) Evaluation of ambient air pollution impact on carrot plants at a suburban site using OTC. *Environ. Monit. Assess.* 119(1), 15-30.
- Tiwari, S., Grote, R., Churkina, G. and Butler, T. (2016) Ozone damage, detoxification and the role of isoprenoids - new impetus for integrated models. *Funct. Plant Biol.* 43(4), 324-336.
- Taylor, G.E. and Hanson, P.J. (1992) Forest trees and tropospheric ozone: role of canopy deposition and leaf uptake in developing exposure - response relationships. *Agricul. Ecosys. Environ.* 42(3-4), 255-273
- Vogel, E. M., Lee, B., Mordi, G., Kim, M. J., Chabal, Y. J., Wallace, R. M., Cho, K. J., Colombo, L. and Kim, J. (2014) Characteristics of high-k Al₂O₃/Al₂O₃ dielectric using ozone-based atomic layer deposition for dual-gated graphene devices. *Plant Biol.* 3, 75-79.
- Wanjari, R. H., Mandal, K. G., Ghosh, P. K., Tapan Adhikari and Rao, N. H. (2006) *Rice in India: Present Status and Strategies to Boost Its Production Through Hybrids.* *Envt. Pollut.* 5, 89-91.
- Walkley, A. and Black, I. A. (1934) An examination for the outline of method for determining soil organic matter and a proposed modification of the chromic acid titration method. *Soil Sci.* 37(1), 29-39.