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Synergistic effects of vehicular emissions (NO_2, SO_2 and SPM) on progression of *Crocus sativus* L.in Saffron bowl Kashmir

Shafat A. Mir[a,*], Javeed I.A. Bhat[a], Farooq Lone[a], Manzoor Ur Rehman[b], Nageena Nazir[c], Ajaz A. Lone[d], Tahir Ali[e], Arshid Jehangir[f]

[a] *Division of Environmental Sciences, SKUAST-K, Shalimar, Srinagar, Jammu and Kashmir 190025, India*
[b] *Division of Veterinary Biochemistry, SKUAST-K, Shalimar, Srinagar, Jammu and Kashmir 190025, India*
[c] *Division of Agri statistics, SKUAST-K, Shalimar, Srinagar, Jammu and Kashmir 190025, India*
[d] *Division of Plant Biotechnology, SKUAST-K, Shalimar, Srinagar, Jammu and Kashmir 190025, India*
[e] *Division of Soil Sciences, SKUAST-K, Shalimar, Srinagar, Jammu and Kashmir 190025, India*
[f] *Department of Environmental Sciences, University of Kashmir, Jammu and Kashmir 190025, India*

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ABSTRACT

Respirable dust sampler (Envirotech model APM-460DXNL) was used to estimate the spatial and seasonal variation of vehicular emissions (NO_2, SO_2, and SPM). Concentration of pollutants in ambient air have been extensively revealed. However, little information is provided on spatial and seasonal variation of vehicular emissions and their impacts on *Crocus sativus*. During the current study the concentration of vehicular emissions was SPM $>SO_2>NO_2$. Length, fresh & dry weight and biochemical parameters of *Crocus sativus* L. changed considerably along the pollution gradient. Shimadzu spectrophotometer model- UV1800ENG240V SOFT was used to estimate changes in biochemical parameters. The aforesaid findings manifest that vehicular emission is a vital factor impacting the progression of *Crocus sativus*. It is of utmost importance to mention here that on reducing vehicular emissions the progression of *Crocus sativus* can be increased substantially. Moreover, our results can also contribute in developing pollution models and hence reduce crop damage thereof.

"Capsule": *Vehicular emissions have a profound negative impact on the progression of Crocus sativus* L.

1. Introduction

For millions of years natural air pollution persisted, but prior to post industrialization anthropogenic pollution is continuously witnessing a steep rise. Throughout the world the proportion of motor vehicles towards air pollution is more than any other human activity (Chauhan, 2010; Iqbal et al., 2015). In India and other developing nations a progressive decline in air quality is experienced. The number of vehicles has shown a steep rise in India with each passing year (Agrawal et al., 2003). With the result agricultural land is exposed to air pollutants of vehicular origin. Multiple factors operate to influence air quality vehicular exhaust being one of them (Yi et al., 2018).The spatial circulation and phase configurations of each are different. The prime elements for the buildup and dispersal of atmospheric pollutants are local emissions, transport, and meteorological conditions (Baralis et al., 2016). Local transport is a direct factor since the pollutant sources and materialization result from local emissions and local transport. The indirect factors include weather, secondary productions, terrain and the time that is needed to pollute the environment. The interactions among these factors are further complex (Yi et al., 2018). Apart from the regular variation, certain particular elements can unexpectedly cause some alterations that can cause a severe decline in the air quality index (AQI) (Zheng et al., 2015).

There is no doubt that vehicular emissions give rise to several pollutants which are hazardous (Curtis et al., 2006: Pope and Dockery, 2006). These pollutants are gaseous phytotoxic such as SO_2, NO_2 and minute suspended particles called suspended particulate matter (SPM). These phytotoxic gases have an overall negative impact on crop production and crop safety (Marshall et al., 2001: Te Lintelo et al., 2002). Phytotoxic air pollutants usually exceed the threshold limits of toxicity to vegetation in numerous Indian cities (Pandey et al., 1992; Stone, 1992). Both nutritional and yield of crops get affected due to an alarming rise in air pollution (Ashmore and Marshall, 1999). Vehicular pollutants rarely exist singly in atmosphere rather they occur in combinations. Their chemical combinations have synergistic, additive or antagonistic effects on plants (Ilyas et al., 2010; Kapoor et al., 2013). Particulate matter cause hindrance in stomatal opening hence transpiration and photosynthesis get affected (Singh et al., 2017; Sharma et al., 2018).The Gaseous ex-

* Corresponding author.
E-mail address: shafatmir@skuastkashmir.ac.in (S.A. Mir).

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change between leaf and ambient air is also affected on deposition of SPM (Singh et al., 2020). Physiological behavior of leaves on exposure to vehicular emissions (SO_2, NO_2, SPM) get altered with the passage of time (Wittenberghe Et Al., 2012).

Our plant of interest for the study was *Crocus sativus* L. (Saffron). *Crocus sativus* is a spice used as food additive in Middle East and Asia and medicinal plant in China (Lertnimitphun et al., 2019). It is a monocot herbaceous species of Iridaceae family (Gismondi et al., 2012).*Crocus sativus* L. is a perennial, herbaceous, 10-25cm high plant, with numerous leaves, that develops from corms (Delgado et al., 2006; Mir et al., 2020). It is an important cash crop of the Kashmir valley and Kashmir being the second in terms of production after Iran (Husaini, 2014).

Enormous databases have been generated on vehicular emissions, but their possible impacts on agricultural crops has been scarcely presented. Moreover, the existing knowledge pertains mostly on experiments where plants were subjected to high doses of air pollutants for short durations (Li, 2003). The present study was therefore carried out in the saffron bowl of Jammu and Kashmir, under field conditions and along a gradient of pollution, in order to assess the changes in physical and biochemical parameters of *Crocus sativus* L. amid rising concentrations of Vehicular emissions (SO_2, NO_2& SPM).

2. Material and methods

2.1. Study area

The study was conducted in saffron city Pampore, district Pulwamaof Union Territory Jammu and Kashmir, situated on the eastern side of river Jhelum on Srinagar-Jammu National Highway (Fig. 4). The National Highway 44 runs through the saffron fields. It is the only major highway that connects Srinagar with the Jammu and the southern part of the Kashmir with the Srinagar. Pampore has an elevation of 1573m with geographic coordinate's 34.02°N 74.93°E. The average weather conditions pertaining to Pampore area with respect to rainfall and temperature follow the trend with rainfall minima during the month of November (22mm) and maxima during March (105mm) whereas, the minimum temperature during the month of January(1.06°c) and maximum during July(24.6°c). The windier part of the year lasts for7.0 months from January 22 to August 21 with average wind speeds of more than 4.5 miles per hour. The windiest day of the year is March 24 with an average hourly wind speed of 5.4 miles per hour. The wind is most often from the west for 4.7 months from May 14 to October 4 with a peak percentage 74% on August 1. The wind is most often from the east for 7.3 Months from October 4 to May 14 with a peak percentage of 58% on January 1.

2.2. Study sites

Two sites *viz.* lethpora (S1) and Galandar (S2) were selected along the national highway that bisects the saffron fields at pampore. Each site comprises of four locations *viz.* L1, L2, L3 and L4 and there were three replications to each location. Locations: L1, L2, L3 and L4 being 10m, 20m, 30m and 1000m away from the national highway. Location 4 at each site was taken as control (Fig. 4). However, in case of vehicular emission sampling only sites *viz.* lethpora (S1) and Galandar (S2) along with their respective controls (L4) were taken.

2.3. Experimental layout

Saffron Corms of appropriate weight (>9.5g) were obtained from farmers. Forty Saffron corms were sown in a plot size of $1m^2$ at each location of corresponding sites and spacing was done according to the norms.Randomised completely block design was used for the layout.

2.4. Vehicular pollution monitoring and analysis

Ambient air samples were taken using respirable dust sampler (Envirotech model APM- 460DXNL). Ambient air quality monitoring was carried out by using impingers filled with absorbing solutions at ground level for 8 h from 9 to 17 h thrice in a month from March 2018 to February 2019. Sulphur dioxide (SO_2) and Nitrogen dioxide (NO_2) were scrubbed separately in 0.1M potassium tetrachloromercurate and 0.1N sodium hydroxide respectively. These absorbing solutions were later analysed colorimetrically. The method employed for SO_2 andNO_2 estimation were improved West and Geake (1956) and improved Jacobs and Hochheiser (1958) respectively. Suspended Particulate Matter (SPM) was determined gravimetrically by employing a method given by Rehme et al. (1984). SPM was collected on glass fiber filter papers by taking pre-sampling and post-sampling filter weights.

2.4.1. Calculation

2.4.1.1. Concentration of SO_2

$$C\left(SO_2\mu g/m^3\right) = \left(A_s - A_b\right) \times CF \times V_s/V_a \times V_t$$

Where,

C SO_2= Concentration of Sulphur dioxide
A_s =Absorbance of sample
A_b =Absorbance of reagent blank
CF= Calibration factor
V_s = Volume of sample ml
V_a = Volume of air sampled, m^3
V_{t} = Volume of aliquot taken for analysis, ml.

2.4.1.2. Concentration of NO_2

$$C\left(NO_2\mu g/m^3\right) = \left(A_s - A_b\right) \times CF \times V_s/V_a \times V_t \times 0.82$$

Where,

C NO_2= Concentration of Nitrogen dioxide
0.82 = Sampling efficiency.

2.4.1.3. Concentration of SPM

$$C\,SPM\left(\mu g/m^3\right) = \left(Wf - Wi\right) \times 10^6/V$$

Where,

C SPM ($\mu g/m^3$) = Concentration of Suspended Particulate matter
Wf= Final weight of filter in g
Wi= Initial weight of filter in g
10^6= conversion of g to μg
V = Volume of air sampled, m^3

2.5. Plant analysis

Physical parameters of plant*viz.* length and weight were measured by centrimetric scale and electronic weighing machine. The Length of leaf was taken after 45 days, 90 days and 135 days of flowering. The corm weight was taken during flowering stage and vegetative stage. The biochemical parameters *viz.* chl *a,* chl *b,* total chl, carotenoids were determined by Hiscox and Isralesham (1979). Total Nitrogen, total Phosphorus, total Potassium (Jackson, 1973), total Carbohydrate (Dubios et al., 1956) and total protein (Lowry et al., 1951) as a soluble fraction were determined. The absorbance of these solutions were measured by Shimadzu spectrophotometer model-UV1800ENG240V,SOFT.

The results were expressed by taking the mean of 15 samples of each replication. For determination of significant differences between sites for different variables Duncun's Multiple range test was performed. The analysis of data was performed through SAS software version 9.2.

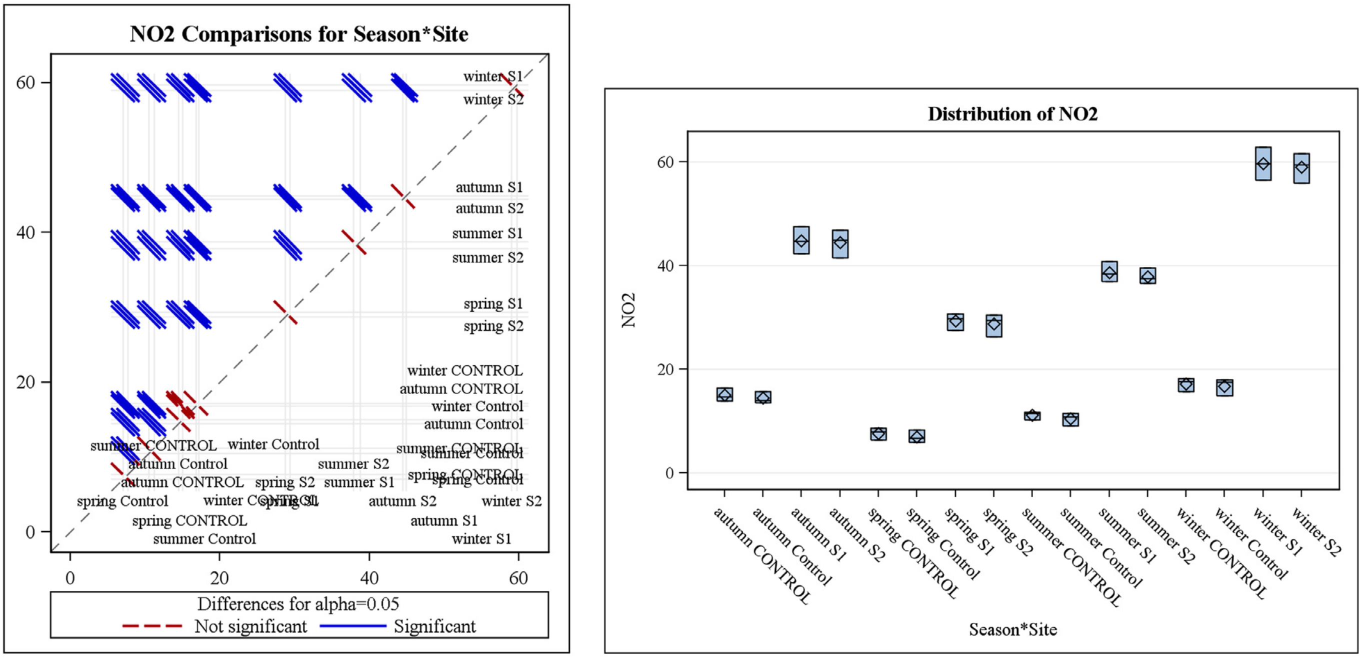

Fig. 1. Effect of vehicular pollution on levels (seasonal) of NO_2 at two different sites (S1 and S2) along with their respective controls.

Table 1
Correlation coefficient between various parameters of air quality at Lethpora and Galandhar during spring 2018-winter 2019.

	SO_2	NO_2	SPM
SO_2	1		
NO_2	0.875**	1	
SPM	0.893**	0.973**	1

** Correlation is significant at the 0.01 level.

3. Results

3.1. Ambient air quality

The study was carried out from March 2018 to February 2019. During the study period Nitrogen dioxide (NO_2) was lowest in spring (7.01 $\mu g/m^3$) at controlof each site while it was highest during winter (59.69 $\mu g/m^3$) at location 1 of each site. In case of Sulphur dioxide (SO_2) the lowest value was recorded in spring season (1.90 $\mu g/m^3$) at control ofboth the sites and the highest was recorded in winter season (24.43$\mu g/m^3$) at Location 1 of Site 1 and Site 2. The lowest value of Suspended Particulate Matter (SPM) was also found in spring (55.74 $\mu g/m^3$) at control of both the sites whereas, the highest value was found in winter (371 $\mu g/m^3$)at Location 1 of the two corresponding sites. There was significant seasonal variation in concentration of each vehicular pollutant -NO_2 $SO_{2,}$ SPM (Fig. 1a, b; Fig. 2a, b; Fig. 3a, b). However, on comparison, insignificant variation was observed between two sites.

Correlation coefficient among various parameters of air quality at two different sites were: r =+ 0.875 (NO_2 and SO_2), r = + 0.893 (SPM and SO_2), r = +0.973 (SPM and NO_2) respectively (Table 1).

3.2. Growth and biochemical parameters

During the course of study various parameters were taken into consideration which includes physical and biochemical parameters as affected by vehicular pollution. It was found that, "length of flower, stigma, stamen, leaf length after 45 days of flowering, leaf length 90 days of flowering and leaf length 135 days of flowering" was minimum i.e. 3.63, 2.16, 1.91, 6.45, 11.22, 17.34 at site 1 location 1 (S1L1) and maximum i.e. 5.76, 3.16, 2.63, 8.76, 14.38, 20.94 at Site 2 Location 4 i.e. control (S2L4) Table 2. Similarly,"fresh weight of flower, dry weight of flower, fresh weight of stigma, dry weight of stigma, fresh weight of stamen, dry weight of stamen, fresh weight of saffron plant, dry weight of saffron plant, fresh weight of leaf after 45 days of flowering, dry weight of leaf after 45 days of flowering, fresh weight of leaf after 90 days of flowering, dry weight of leaf after 90 days of flowering, fresh weight of leaf after 135 days of flowering, dry weight of leaf after 135 days of flowering, fresh weight of corm during flowering stage, dry weight of corm during flowering stage, fresh weight of corm during vegetative stage and dry weight of corm during vegetative stage" was minimum i.e. 0.28, 0.063, 0.033, 0.007, 0.024, 0.0043, 14.32, 3.29, 1.18, 0.253, 1.90, 0.472, 0.633, 10.16, 2.20, 10.19, 2.20and maximum i.e. 0.49, 0.087, 0.050, 0.008, 0.040, 0.0067, 16.57, 5.49, 1.44, 2.90, 2.18, 0.515, 0.670, 10.42, 2.47, 10.39, 2.40 at Site 2 Location 4 i.e. control (S2L4) Table 3. The biochemical parameters *viz.* "chlorophyll *a,* chlorophyll *b,* Total chlorophyll, Carotenoids, Total Nitrogen, Total phosphorus, Total Potassium, Total Carbohydrates, and Total Proteins" showed their minimum values i.e. 0.21, 0.07, 0.11, 1.47, 0.18, 0.96, 16.6, 8.98 at Site 1 Location 1(S1L1) and maximum values i.e. 0.52, 0.21, 0.32, 2.15, 0.49, 1.68, 22.5, 13.34 at Site 2 location 4 i.e. Control (S2L4) table 4. The results thus obtained were insignificant when sites were compared but along locations the results were comprehensively significant.

4. Discussion

4.1. Spatial and Seasonal variation of SO_2, NO_2 and SPM

The study was aimed at furnishing the details of pollutants such as SO_2, $NO_2$and SPM on the various physico and biochemical properties of *Crocus sativus* in the pampore area of Jammu & Kashmir. The vehicular pollution monitoring data at two different sites and Lethpora (S1) and Galandhar (S2) showed spatial and seasonal variations for SO_2, NO_2 and SPM. The spatial pattern of vehicular pollution of each site is marked by high SO_2, NO_2 and SPM levels due to its close proximity to the national

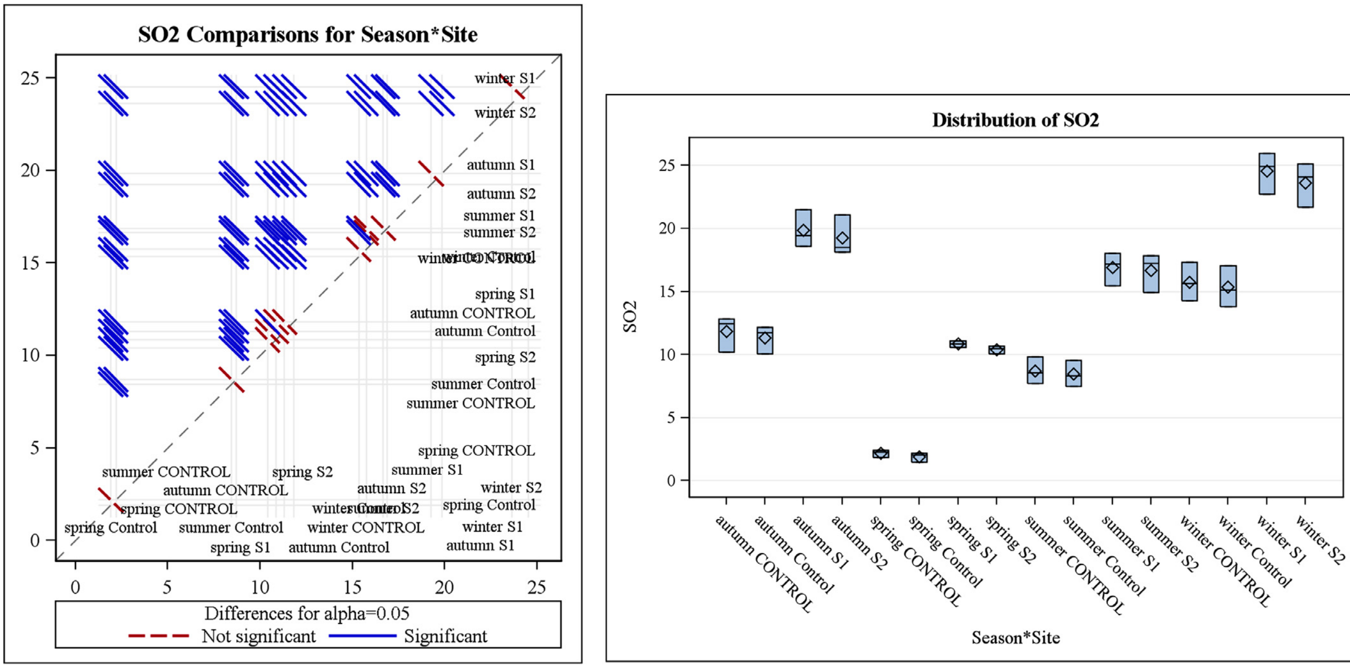

Fig. 2. Effect of vehicular pollution on levels (seasonal) of SO_2 at two different sites (S1 and S2) along with their respective controls.

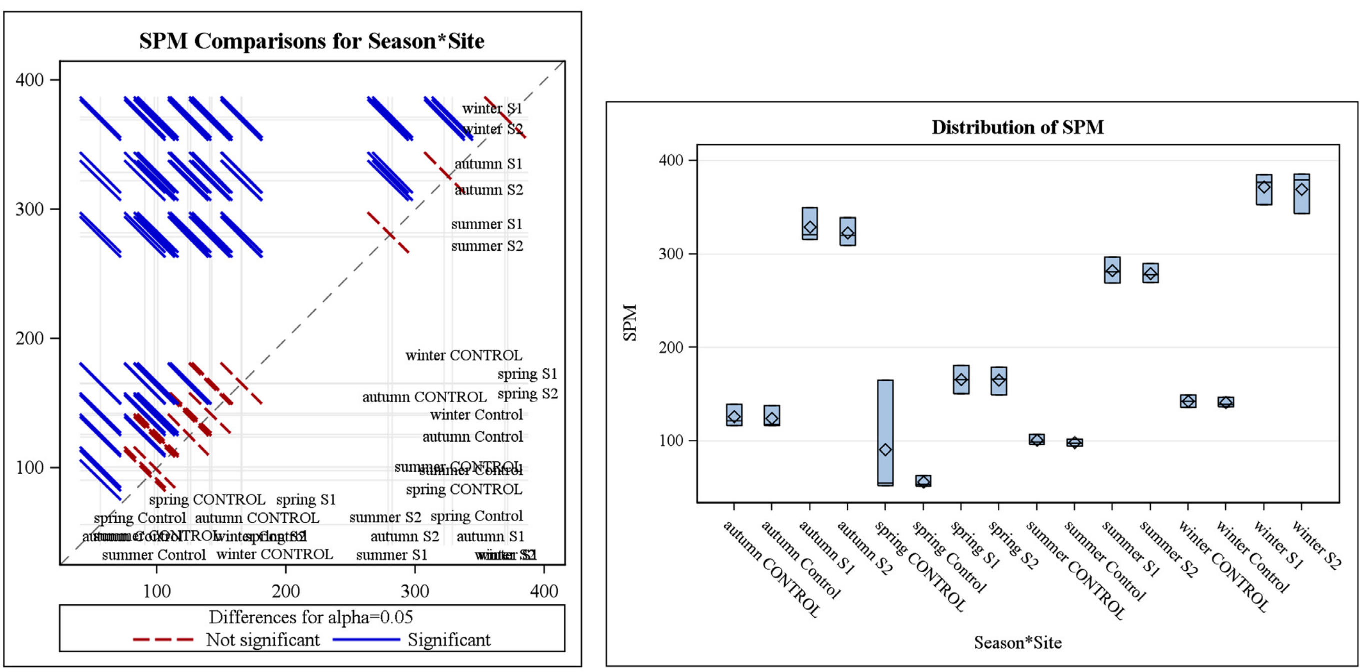

Fig. 3. Effect of vehicular pollution on levels (seasonal) of SPM at two different sites (S1 and S2) along with their respective controls.

highway with no plantation along the road whereas, L4 (control) corresponding to each site is marked by gradual decrease in SO_2, NO_2 and SPM concentrations. The reasons may be alley cropping and large populous plantation with wider canopies that may have absorbed a large proportion of pollutants and also helped in the dispersal of pollutants along the periphery. It is also due to mention here that at control (L4) of each site there was no visible source of vehicular pollution either. Among seasons, winter season recorded the highest pollutant concentrations as during winter weather conditions are calm leading to stability, slow dispersion of pollutants and inversion factor playing a pivotal role in stagnation of pollutants in the atmosphere (Agrawal et al., 2003; Kapoor et al., 2013; Li, 2003).

4.2. Impacts of SO_2, $NO_2$and SPM on length of flower, stigma, stamen, leaf length (after 45,90, 135 days)

Vehicular pollution is one among the limiting factors related to physiological stressof a plant and inevitably determines the production and rates of survivorship. Reduction in length of flower, stigma, stamen, leaf length was recorded at location 1 of each site. Reports of NO_2 andSO_2 suggest that these are potential inhibitors of net photosynthesis. Inhibition of net photosynthesis restricts assimilated translocation which results in decline of leaf area (Kapoor et al., 2013). Dinova (2004) also reported reduction of leaf area and petiole length under stress pollution conditions. Nimantha and Indira (2005) revealed the drastic effects of

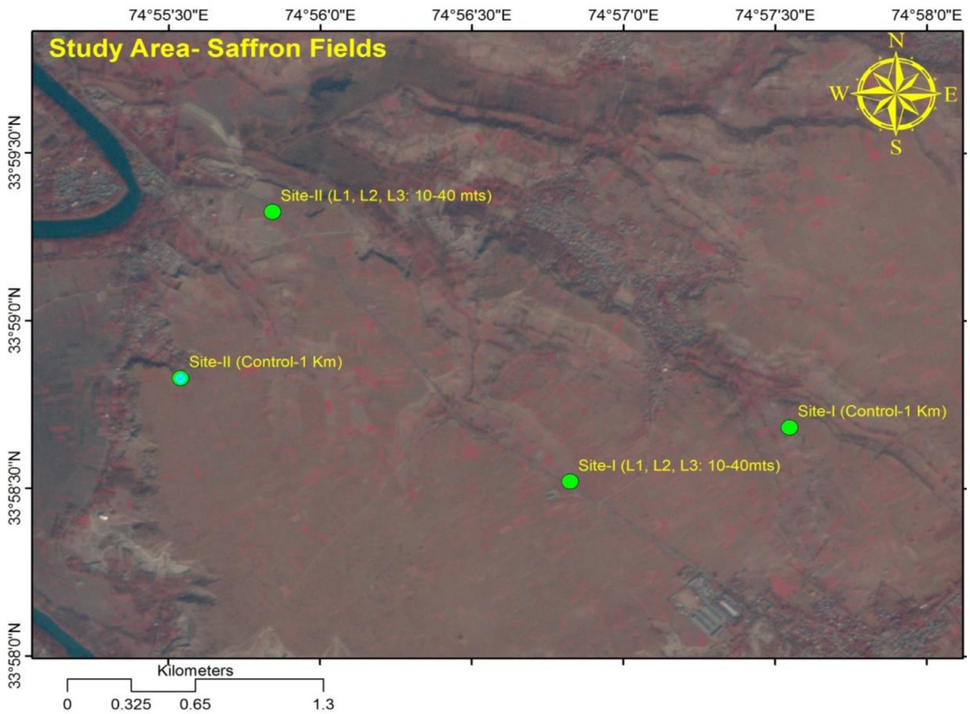

Fig. 4. Study area and study sites.

Table 2

Changes in length of flower, stigma, stamen and leaf at two different sites (Lethpora and Galandhar).

Parameters	Sites	L1	L2	L3	L4 (Control)	Overall mean
"Length of flower" (cm)	Lethpora	3.63 ± 0.05	3.80 ± 0.10	4.13 ± 0.05	5.70 ± 0.10	4.31[a]
	Galandhar	3.66 ± 0.05	3.86 ± 0.11	4.23 ± 0.11	5.76 ± 0.05	4.38[a]
"Length of stigma" (cm)	Lethpora	2.16 ± 0.05	2.36 ± 0.05	2.56 ± 0.05	3.06 ± 0.05	2.54[b]
	Galandhar	2.20 ± 0.10	2.40 ± 0.10	2.63 ± 0.11	3.16 ± 0.05	2.60[a]
"Length of stamen" (cm)	Lethpora	1.91 ± 0.10	2.06 ± 0.05	2.16 ± 0.05	2.53 ± 0.05	2.16[a]
	Galandhar	1.93 ± 0.05	2.13 ± 0.05	2.23 ± 0.05	2.63 ± 0.05	2.23[a]
"leaf length after 45 days of flowering" (cm)	Lethpora	6.45 ± 0.06	6.72 ± 0.11	7.10 ± 0.13	8.72 ± 0.08	7.25[a]
	Galandhar	6.46 ± 0.06	6.75 ± 0.12	7.12 ± 0.13	8.76 ± 0.07	7.27[a]
"Leaf length after 90 days of flowering" (cm)	Lethpora	11.22 ± 0.11	11.73 ± 0.18	12.11 ± 0.14	14.38 ± 0.36	12.36[a]
	Galandhar	11.23 ± 0.13	11.75 ± 0.17	12.14 ± 0.14	14.42 ± 0.37	12.38[a]
"Leaf length after 135 days of flowering" (cm)	Lethpora	17.34 ± 0.11	17.80 ± 0.10	18.28 ± 0.12	20.91 ± 0.27	18.58[a]
	Galandhar	17.34 ± 0.08	17.84 ± 0.14	18.31 ± 0.10	20.94 ± 0.25	18.61[a]

pollutants (SO_2, NO_2) on seed germination, length of pedicels, no. of flowers and inflorescence. A study conducted on growth along a gradient of ambient air pollution by Ashmore et al., (1988) putforths that air pollutants were a major factor influencing plant growth. Reports from Pandey and Agrawal (1994) also suggested reductions in height of perennials under varying levels of air pollution.

4.3. Impacts of SO_2, NO_2 and SPM on fresh and dry weight of flower, stigma, stamen, saffron plant, leaf (after 45, 90, 135 days) corm (during flowering and vegetative stage)

An inevitable reduction in fresh and dry weight of all the above parameters was observed along the line of pollution. Reduced chlorophyll, photosynthesis, carbohydrate and protiens at polluted locations may be the reasons to the pronounced reductions (Skinder et al., 2015: Kapoor et al.2013;Irshad et al 2011; Balkhi et al. 2009;; Ziegler, 1973). All these parameters showed negative correlation with the vehicular pollutants. Much pronounced healthy crop was observed on moving away from the distressed polluted locations. The fact that supports this statement may be attributed to the alley cropping at control locations and large foliage populous trees. They reduce the amount of pollutants through their leaf surfaces and also help in their dispersal. Thus reducing the amount of burden on the Saffron crop.

4.4. Impacts of SO_2, NO_2 and SPM on chlorophyll a, chlorophyll b, total chlorophyll, carotenoids

The chlorophyll pigments are susceptible to be degraded by vehicular pollutants. Chlorophyll molecule has been described as a biomarker parameter for higher pollution levels (Agrawal et al., 2003). Chlorophyll is also regarded as productivity index hence any changes in chlorophyll changes the morphological, physiological and biochemical behavior of the plant (Skinder et al., 2015). The results revealed reductions in Chlorophyll *a,* Chlorophyll *b,* total chlorophyll and carotenoid concentrations at locations having close proximity to national highway i.e L1 of each site as compared to the control locations i.e L4 of each site. Reasons for decreased concentrations of these pigments are attributed to the action of constituents of vehicular emissions (SO_2, NO_2, SPM). Reductions may also be due to increase in chlorophyllase enzyme activities which affects chlorophyll concentration (Kapoor et al.2013; Balkhi et al. 2009; Irshad et al 2011). It has been observed that on exposure plants absorb and accumulate environmental pollutants (Agbaire and Esiefarienrhe, 2009). Higher levels of air pollutants increase the generation of electrons in chloroplasts hence induces oxidative stress (Sakaki et al. 1983; Woo et al. 2007). Research findings have shown that SOx, NOx and SPM cause damage to the thylakoid membrane system in the chloroplast

Table 3
Changes in fresh and dry weight of flower, stigma, stamen saffron plant and leaf at two different sites (Lethpora and Galandhar).

Parameters	Sites	L1	L2	L3	L4 (Control)	Overall mean
"Fresh wt. of flower" (g)	Lethpora	0.28 ± 0.01	0.33 ± 0.01	0.40 ± 0.01	0.48 ± 0.01	0.37[a]
	Galandhar	0.29 ± 0.01	0.35 ± 0.01	0.40 ± 0.01	0.49 ± 0.01	0.38[a]
"Dry wt. of flower" (g)	Lethpora	0.063 ± 0.001	0.070 ± 0.001	0.081 ± 0.001	0.086 ± 0.002	0.075[a]
	Galandhar	0.063 ± 0.002	0.071 ± 0.002	0.081 ± 0.001	0.087 ± 0.002	0.076[a]
"Fresh wt. of stigma" (g)	Lethpora	0.033 ± 0.001	0.035 ± 0.001	0.039 ± 0.001	0.048 ± 0.001	0.04[b]
	Galandhar	0.034 ± 0.001	0.037 ± 0.001	0.040 ± 0.001	0.050 ± 0.001	0.03[a]
"Dry wt. of stigma" (g)	Lethpora	0.0074 ± 0.001	0.0077 ± 0.001	0.0079 ± 0.001	0.0085 ± 0.001	0.007[a]
	Galandhar	0.0076 ± 0.001	0.0079 ± 0.000	0.0081 ± 0.001	0.0087 ± 0.001	0.008[a]
"Fresh wt. of stamen" (g)	Lethpora	0.024 ± 0.001	0.026 ± 0.001	0.028 ± 0.001	0.039 ± 0.001	0.029[b]
	Galandhar	0.025 ± 0.001	0.026 ± 0.001	0.030 ± 0.001	0.040 ± 0.001	0.030[a]
"Dry wt. of stamen" (g)	Lethpora	0.0043 ± 0.0001	0.0048 ± 0.0001	0.0053 ± 0.0001	0.0064 ± 0.0001	0.0052[b]
	Galandhar	0.0044 ± 0.0001	0.0050 ± 0.0001	0.0054 ± 0.0001	0.0067 ± 0.0001	0.0054[a]
"Fresh wt. of saffron plant" (g)	Lethpora	14.32 ± 0.13	14.52 ± 0.05	14.77 ± 0.03	16.52 ± 0.03	15.03[b]
	Galandhar	14.40 ± 0.11	14.56 ± 0.03	14.90 ± 0.01	16.57 ± 0.07	15.10[a]
"Dry wt. of saffron plant" (g)	Lethpora	3.29 ± 0.06	3.59 ± 0.04	3.73 ± 0.04	5.41 ± 0.05	4.00[b]
	Galandhar	3.37 ± 0.11	3.63 ± 0.05	3.85 ± 0.04	5.49 ± 0.03	4.09[a]
"Fresh wt. of leaf after 45 days	Lethpora	1.18 ± 0.02	1.25 ± 0.02	1.29 ± 0.02	1.42 ± 0.02	1.28[a]
of flowering" (g)	Galandhar	1.19 ± 0.01	1.26 ± 0.01	1.31 ± 0.01	1.44 ± 0.01	1.30[a]
"Dry wt. of leaf after 45 days	Lethpora	0.253 ± 0.002	0.258 ± 0.002	0.265 ± 0.002	0.288 ± 0.002	0.266[b]
of flowering" (g)	Galandhar	0.254 ± 0.001	0.259 ± 0.002	0.267 ± 0.002	0.290 ± 0.002	0.268[a]
"Fresh wt. of leaf after 90 days	Lethpora	1.90 ± 0.02	1.93 ± 0.01	1.97 ± 0.02	2.15 ± 0.03	1.99[a]
of flowering" (g)	Galandhar	1.92 ± 0.01	1.95 ± 0.01	2.00 ± 0.03	2.18 ± 0.03	2.01[a]
"Dry wt. of leaf after 90 days	Lethpora	0.472 ± 0.002	0.475 ± 0.001	0.481 ± 0.002	0.511 ± 0.002	0.485[a]
of flowering" (g)	Galandhar	0.474 ± 0.001	0.477 ± 0.001	0.483 ± 0.003	0.515 ± 0.003	0.487[a]
"Fresh wt. of leaf after 135	Lethpora	2.33 ± 0.02	2.37 ± 0.02	2.43 ± 0.02	2.59 ± 0.03	2.43[a]
days of flowering" (g)	Galandhar	2.35 ± 0.02	2.40 ± 0.01	2.47 ± 0.02	2.61 ± 0.02	2.46[a]
"Dry wt. of leaf after 135 days	Lethpora	0.633 ± 0.003	0.636 ± 0.001	0.642 ± 0.002	0.665 ± 0.003	0.644[a]
of flowering" (g)	Galandhar	0.635 ± 0.002	0.638 ± 0.002	0.645 ± 0.002	0.670 ± 0.002	0.647[a]
"Fresh wt. of corm during	Lethpora	10.16 ± 0.01	10.22 ± 0.01	10.27 ± 0.01	10.41 ± 0.01	10.26[a]
flowering stage" (g)	Galandhar	10.18 ± 0.01	10.21 ± 0.01	10.27 ± 0.01	10.42 ± 0.01	10.27[a]
"Dry wt. of corm during	Lethpora	2.20 ± 0.01	2.26 ± 0.01	2.31 ± 0.01	2.46 ± 0.01	2.31[b]
flowering stage" (g)	Galandhar	2.22 ± 0.01	2.26 ± 0.01	2.32 ± 0.01	2.47 ± 0.01	2.32[a]
"Fresh wt. of corm during	Lethpora	10.19 ± 0.01	10.23 ± 0.01	10.26 ± 0.01	10.37 ± 0.01	10.26[b]
vegetative stage" (g)	Galandhar	10.20 ± 0.01	10.24 ± 0.01	10.28 ± 0.01	10.39 ± 0.01	10.28[a]
"Dry wt. of corm during	Lethpora	2.20 ± 0.01	2.25 ± 0.01	2.28 ± 0.01	2.38 ± 0.01	2.27[b]
vegetative stage" (g)	Galandhar	2.22 ± 0.01	2.26 ± 0.01	2.29 ± 0.01	2.40 ± 0.01	2.29[a]

Table 4
Changes in biochemical parameters at two different sites (Lethpora and Galandhar).

Parameters	Sites	L1	L2	L3	L4 (Control)	Overall mean
"Chlorophyll *a*"(mg/g)	Lethpora	0.21 ± 0.02	0.26 ± 0.01	0.34 ± 0.02	0.50 ± 0.01	0.30[a]
	Galandhar	0.23 ± 0.02	0.27 ± 0.01	0.36 ± 0.01	0.52 ± 0.01	0.34[a]
"Chlorophyll *b*"(mg/g)	Lethpora	0.07 ± 0.01	0.11 ± 0.01	0.14 ± 0.01	0.20 ± 0.01	0.13[a]
	Galandhar	0.08 ± 0.01	0.11 ± 0.01	0.15 ± 0.01	0.21 ± 0.01	0.14[a]
"Total chlorophyll"	Lethpora	0.29 ± 0.04	0.40 ± 0.03	0.52 ± 0.02	0.72 ± 0.02	0.48[a]
(mg/g)	Galandhar	0.32 ± 0.04	0.42 ± 0.03	0.55 ± 0.02	0.78 ± 0.03	0.52 [a]
"Carotenoids"(mg/g)	Lethpora	0.11 ± 0.01	0.17 ± 0.02	0.23 ± 0.01	0.30 ± 0.01	0.20[a]
	Galandhar	0.12 ± 0.01	0.18 ± 0.02	0.24 ± 0.01	0.32 ± 0.02	0.21[a]
"Total Nitrogen"(%)	Lethpora	1.47 ± 0.03	1.61 ± 0.03	1.76 ± 0.02	2.11 ± 0.02	1.74[a]
	Galandhar	1.52 ± 0.03	1.63 ± 0.01	1.79 ± 0.01	2.15 ± 0.02	1.77[a]
"Total phosphorus"(%)	Lethpora	0.18 ± 0.01	0.25 ± 0.01	0.33 ± 0.02	0.45 ± 0.01	0.30[a]
	Galandhar	0.21 ± 0.02	0.26 ± 0.01	0.35 ± 0.01	0.49 ± 0.01	0.33[a]
"Total Potassium"(%)	Lethpora	0.96 ± 0.02	1.05 ± 0.02	1.16 ± 0.01	1.61 ± 0.03	1.20[a]
	Galandhar	0.98 ± 0.02	1.07 ± 0.02	1.19 ± 0.01	1.68 ± 0.03	1.23[a]
"Total	Lethpora	16.6 ± 0.04	16.8 ± 0.06	17.3 ± 0.04	22.4 ± 0.15	18.31[a]
Carbohydrates"(%)	Galandhar	16.7 ± 0.03	16.9 ± 0.05	17.4 ± 0.02	22.5 ± 0.07	18.41[a]
"Total proteins"(%)	Lethpora	8.98 ± 0.10	9.22 ± 0.07	9.65 ± 0.04	13.26 ± 0.05	10.27[a]
	Galandhar	9.03 ± 0.08	9.25 ± 0.06	9.74 ± 0.01	13.34 ± 0.07	10.34[a]

(Wellburn 1998; Liu et al. 2007). Damages to the thylakoid membrane are responsible for the decline of pigment concentrations (Malhotra and Hocking 1976).It has been seen that heavy load of traffic converts chlorophyll *a* molecules to pheophytin through replacement of Mg^{2+} ions. Chlorophyll *b* is changed to chlorophyllide *b* after removal of phytol group by increased concentration of SO_2 (Roa and Leblanc, 1966; Saquib et al. 2010).

4.5. Impacts of SO_2, $NO_2$and SPM on total nitrogen (N), total potassium (K) and total phosphorus (P)

The concentration of Total Nitrogen, Total potassium and Total Phosphorus of *Crocus sativus* showed a marked decrease along the source of pollution. Leaching of minerals decrease the concentration of N and K at vehicular effected soils (Fenga et al., 2018; Singh et al., 2018). This

Table 5
Percent reduction over control.

Parameters	Sites	Location 1	Location 2	Location 3
"Length of flower"	Lethpora	36	33	27
	Galandhar	36.4	32.98	26.56
"Length of stigma"	Lethpora	29.41	22.8	14.05
	Galandhar	30.37	24.05	16.77
"Length of stamen"	Lethpora	24.5	18.57	14.62
	Galandhar	26.61	19.01	15.2
"Leaf length after 45 days of flowering"	Lethpora	26.03	22.93	18.57
	Galandhar	26.25	22.96	18.72
"Leaf length after 90 days of flowering"	Lethpora	21.97	18.42	15.78
	Galandhar	22.12	18.51	15.81
"Leaf length after 90 days of flowering"	Lethpora	17.07	14.87	12.57
	Galandhar	17.19	14.8	12.56
"Fresh wt. of flower" (g)	Lethpora	41	31.25	16.6
	Galandhar	40.81	28.57	18.36
"Dry wt. of flower"(g)	Lethpora	26.74	18.6	5.81
	Galandhar	27.58	18.39	6.89
"Fresh wt. of stigma" (g)	Lethpora	31.25	27.08	18.75
	Galandhar	32	26	20
"Dry wt. of stigma" (g)	Lethpora	12.94	9.41	7.05
	Galandhar	12.64	9.2	6.89
"Fresh wt. of stamen" (g)	Lethpora	38.46	33.3	28.2
	Galandhar	37.5	35	25
"Dry wt. of stamen" (g)	Lethpora	32.8	25	17.18
	Galandhar	34.32	25.37	19.4
"Fresh wt. of saffron plant" (g)	Lethpora	13.31	12.1	10.59
	Galandhar	13.09	12.13	10.07
"Dry wt. of saffron plant" (g)	Lethpora	39.18	33.64	31.05
	Galandhar	38.61	33.87	29.87
"Fresh wt. of leaf after 45 days of flowering" (g)	Lethpora	16.9	11.97	9.15
	Galandhar	17.36	12.5	9.02
"Dry wt. of leaf after 45 days of flowering" (g)	Lethpora	12.15	10.41	7.98
	Galandhar	12.41	10.68	7.93
"Fresh wt. of leaf after 90 days of flowering" (g)	Lethpora	11.62	10.23	8.37
	Galandhar	11.92	10.55	8.25
"Dry wt. of leaf after 90 days of flowering" (g)	Lethpora	7.63	7.04	5.47
	Galandhar	7.96	7.37	6.21
"Fresh wt. of leaf after 135 days of flowering" (g)	Lethpora	10.03	8.49	6.17
	Galandhar	9.96	8.04	5.36
"Dry wt. of leaf after 135 days of flowering" (g)	Lethpora	4.18	4.36	3.45
	Galandhar	5.22	4.77	3.73
"Fresh wt. of corm during flowering stage" (g)	Lethpora	2.4	1.82	1.34
	Galandhar	2.3	2.01	1.43
"Dry wt. of corm during flowering stage" (g)	Lethpora	10.56	8.13	6.09
	Galandhar	10.12	8.5	6.07
"Fresh wt. of corm during vegetative stage" (g)	Lethpora	1.73	1.35	1.06
	Galandhar	1.82	1.44	1.05
"Dry wt. of corm during vegetative stage" (g)	Lethpora	7.56	5.46	4.2
	Galandhar	7.5	5.83	4.58
"Chlorophyll *a*"(mg/g)	Lethpora	58	48	32
	Galandhar	55.76	48.07	30.76
"Chlorophyll *b*"(mg/g)	Lethpora	65	45	30
	Galandhar	61.9	47.61	28.57
"Total chlorophyll"(mg/g)	Lethpora	59.72	44.44	27.77
	Galandhar	58.97	46.15	29.48
"Carotenoids"(mg/g)	Lethpora	63.33	43.33	23.33
	Galandhar	62.5	43.75	25
"Total Nitrogen"(%)	Lethpora	30	23.69	16.58
	Galandhar	29.3	24.18	16.74
"Total phosphorus"(%)	Lethpora	60	44.4	26.66
	Galandhar	57.14	46.93	28.57
"Total Potassium"(%)	Lethpora	40.37	34.78	27.95
	Galandhar	41.66	36.3	29.16
"Total Carbohydrates"(%)	Lethpora	25.89	25	22.76
	Galandhar	25.77	24.88	22.66
"Total proteins"(%)	Lethpora	32.27	30.46	27.22
	Galandhar	32.3	30.65	26.98

argument lists the possibility of less availability of mineral nutrients to the plants. Less content of potassium in needles may be due to heavy metals accumulation which cause leakage of potassium from the cells of plant (Singh et al.,2018).Phosphorus availability may be attributed to pH the more the pH the more is availability (Ali and Tsega, 2018; Kimura et al., 2009).

4.6. *Impacts of SO_2, $NO_2$and SPM on total proteins and total carbohydrates*

Changes in carbohydrate and protein content along the pollution gradient were witnessed. As we moved away from location 1 to location 4(control) from each site a considerable increase in carbohydrate and protein content was observed. Davison and Barnes (1986) reiterated that degradatioin of carbohydrates in the leaves of plants at sites polluted by SO_2 NO_2 was prominent. Another possibility of carbohydrate concentration changes could be due to loss of chlorophyll molecules which hampers the rate of photosynthesis owing to affinity for the carboxylase enzyme between CO_2 and SO_2 (Ziegler, 1973; Skinder et al., 2015). Also stomatal pore is covered either fully or partially by Suspended Particulate Matter on deposition that regulates the transpiration and uptake of CO_2 for photosynthesis (Singh et al. 2017; Sharma et al 2018). Deposition also creates a substantial way for toxic gases to enter into the leaf hence causes acidic conditions (Singh et al. 2020).

5. Conclusion

To the best of our knowledge this is the first study on synergistic effects SO_2, NO_2 and SPM on *Crocus sativus* L. Throughout the study we have seen the changes in the crop because of heavy traffic load. Suspended Particulate Matter outdated SO_2 and NO_2 in terms of concentration at all locations and seasons. The results suggested that increased concentration of these pollutants not only reflected the levels of yield loss at different locations but stunted growth, reduced foliage and small flowers were also the outcomes of increased concentrations of these pollutants. To undone the gap on relations of pollutants with the crop, we suggest that further studies are needed to explore the responses of *Crocus sativus* to SO_2, NO_2 and SPM at molecular level.

Declaration of Competing Interest

All contributing authors declare no conflicts of interest

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