Crop adaptation to air pollution I. Effect of particulate and SO$_2$ pollution on growth, yield attributes and sulphur nutrition of wheat, barley and chickpea

Poonam Yadav¹, Renu Dhupper¹, S.D Singh and Bhupinder Singh*

Centre for Environment Science and Climate Resilient Agriculture, ICAR-Indian Agricultural Research Institute, New Delhi-110 012, India.

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ABSTRACT

Sulphur dioxide (SO$_2$) and particulate matter (PM) are one of the major air pollutants emerging out of the industrial development and human activities. Plants exhibit differential sensitivity to SO$_2$ pollution and its affect on plant growth can be both direct and/or indirect. The present study was conducted in controlled tunnels to assess the effect of particulate matter (PM) and SO$_2$ on growth attributes of two cereals (bread and durum wheat and barley) and a legume (chickpea) species. Relative sensitivity of crops to elevated SO$_2$ followed the following order: durum wheat < bread wheat < barley < chickpea. This study clearly shows that the presence of particulate matter in the growing environment severely inhibits crop growth while the SO$_2$ enriched environment promotes plant growth and S uptake across crops and that the tolerant crop species are capable of utilizing SO$_2$ towards the plant S pool.

Key words: Growth attributes, Particulate matter, SO$_2$ stress, S-uptake.

INTRODUCTION

Air quality depends on presence of both particulate and noxious gas pollutants. Particulate pollutants may inhibit growth by causing physical impediment for the stomatal gas exchange and also as a site for heavy metal deposition, while sulfur dioxide and nitrous oxide are the primary noxious gases contributing towards air pollution. Crop yield reduction of 10-50% in response to SO$_2$ concentration in the range of 75-139 µg m$^{-3}$ has been reported (Burney and Ramanathan, 2014). Effect of SO$_2$ on a plant system may be direct or indirect. Phytotoxicity of SO$_2$ is determined by an interactive continuum between the soil, the plant and the environment. Wide variation in plant response to SO$_2$ may exist at the genetic and the species level (Prasad and Rao, 1982; Padhi et. al., 2013). Further, atmospheric SO$_2$ on absorption by the leave can enter the S assimilatory pathway directly or after oxidation to SO$_3^-$, be reduced to sulfide and incorporated into cysteine and subsequently into organic S compounds, and utilized as a S nutrient by the crop plants (Peter et. al., 2012). A schematic of the likely route of SO$_2$ stress response in crop plants is given in Fig 1. While these effects depend on the length and the concentration of subjection, the direct effect cause loss of chlorophyll or bleaching of the photosynthetically active surfaces. The direct effects may also be attributed to an inhibition of photosynthetic system, since the SO$_2$ stress causes an increased opening of the stomata leading to a rapid loss of water and/or an unregulated exchange of gases from the crop cover, to consequently cause a reduction in crop productivity and quality. However, here again the loss of yield and produce quality shall be governed by the duration and the concentration of not only the SO$_2$ subjection but also the presence of other pollutants such as NO$_2$. Variation in crop sensitivity to SO$_2$ may be attributed to difference in efficiency for absorption of gaseous pollutants and plant’s ability to detoxify the pollutant and dispose off the excess load. Most of the sulfur absorbed by the leaves is translocated to the other plant parts, thus diluting its concentration over time and with the vegetative growth. However, the exposure becomes threatening when the concentration of SO$_2$ and the sulphite derivatives reaches levels higher than the plants detoxification capacity. However, the extent of injury shall depend on the exposure dose with modest doses, showing a positive gain in terms of plant growth and development. Contribution of SO$_2$ towards plant S-nutrition may also improve plants capacity to tolerate abiotic stress by protecting protein structure and thus their activity against proteases and hydrolases, which need to be substantiated (Prasad and Rao, 1982; Hongfa et. al., 1999; Sha et. al., 2010). The present study attempts to assess the effect of particulate and SO$_2$ pollution on growth and development of two cereal (bread and durum wheat and barley) and a legume (chickpea) species.

MATERIALS AND METHODS

Experimental setup and plant material: Experiment was conducted in enclosed tunnels (size 10 x 2.5 x 2.5 m$^3$; PAR 800-1200 µmol m$^{-2}$·s$^{-1}$; temperature ~25±5°C, RH ~60-70%) to assess the effect of particulate and gaseous air pollutants on growth and sulfur nutrition of bread wheat i.e., *Triticum aestivum* var. HD-2967, durum wheat (*Triticum

*Corresponding author’s e-mail: bhupindersinghiari@yahoo.com

¹Amity Institute of Environmental Science, Amity University, Noida, Uttar Pradesh, India.
**RESULTS AND DISCUSSION**

Effect of gaseous and particulate pollutants on plant growth, yield and S-uptake of cereal and legume crops

**Shoot mass, leaf area and chlorophyll:** Between the treatments, highest shoot mass at 60 day growth stage, was measured under the ambient control in bread wheat, chickpea and barley, while in durum wheat, highest shoot biomass was measured under the elevated SO$_2$ condition (Fig 2). Least biomass across all crops, in general, was measured under the treatment where charcoal and particulate filters were used (T1). Durum wheat was the only crop which showed an increase in both shoot mass and leaf area under the SO$_2$ enrichment, while for all other crops leaf area declined under the elevated SO$_2$ condition. Use of activated charcoal filter provided a growth environment that facilitated the leaf area development in chickpea but not for the other experimental cereal species (Fig 2). No significant change in the leaf chlorophyll levels were measured (P>0.05) between different environmental conditions of crop growth. The SO$_2$ enrichment did not adversely affect the leaf chlorophyll measured at 60 days of crop growth. A significantly positive effect of particle removal from the growing environment on leaf chlorophyll was measured only for the durum wheat (Fig 2).

A look at plant biomass variation across the experimental crops at physiological harvest stage indicates a highest biomass yield in growth environment that is free from particulate pollutants (T2), followed by the SO$_2$ fumigated environment (T4) as compared to the ambient control (T3). Use of charcoal and particulate filter from the growing environment (T1) drastically reduced the plant biomass yield across all crops, in general, was measured under the ambient control in bread wheat, chickpea and barley, while in durum wheat, highest shoot biomass was measured under the ambient control (T3). Use of charcoal and particulate filters were used (T1). Durum wheat was the only crop which showed an increase in both shoot mass and leaf area under the SO$_2$ enrichment, while for all other crops leaf area declined under the elevated SO$_2$ condition. Use of activated charcoal filter provided a growth environment that facilitated the leaf area development in chickpea but not for the other experimental cereal species (Fig 2). No significant change in the leaf chlorophyll levels were measured (P>0.05) between different environmental conditions of crop growth. The SO$_2$ enrichment did not adversely affect the leaf chlorophyll measured at 60 days of crop growth. A significantly positive effect of particle removal from the growing environment on leaf chlorophyll was measured only for the durum wheat (Fig 2).

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**Leaf and grain sulphur:** Sulphate (SO$_2^{2-}$) level in the leaf and grain samples were estimated following turbidimetric method (Tabatabai and Bremner, 1970) at 60 days and at the harvest stage. The absorbance was read at 420 nm using spectrophotometer; S-concentration and S- content were determined from the S-standard curve prepared in a range of 0-10 mg S l$^{-1}$ and expressed as µg g$^{-1}$ dry weight and µg S plant$^{-1}$, respectively.

**Statistical analysis:** Data was statistically analyzed by using SPSS version 23 statistical program and significant difference between the treatments within the selected crop were determined using one-way ANOVA at n=3, as described by (Snedecor and Cochran, 1980) in combination with Duncan’s Multiple Range Test (DMRT) at $P < 0.05$. 

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**Shoot biomass, leaf area and leaf chlorophyll:** Shoot mass was recorded by subjecting the freshly harvested plants to drying in an open air oven maintained at 60°C for a few days until complete dryness and was expressed as g dw plant$^{-1}$. Leaf area was recorded using the leaf area meter (LiCOR 3000, Lincoln Nebraska, USA) and was expressed as cm$^{2}$ plant$^{-1}$. Leaf chlorophyll content was measured using dimethyl sulphoxide (DMSO) method of (Hiscox and Israelstam, 1979). The absorbance was measured at 645 and 663 nm and total chlorophyll was calculated using the formula given by (Arnon, 1949), and expressed as (mg g$^{-1}$ fw).

\[
\text{Total chlorophyll} = \frac{20.2 (A_{663}) + 8.02 (A_{645}) \times V}{W}
\]

Where, \(V\) = Final extract volume and, \(W\) = Weight of tissue extracted.

**Yield attributes:** Plant biomass, average spike or pod number and mass per plant and per unit cultivable area and grain/seed mass were recorded at the final harvest for ten plants per replicate in three replications. Terms pod and seed are used in case of chickpea.

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**Fig 1:** Schmatic of the likely route of SO$_2$ stress response in crop plants.
studied the toxic effect of sulphur dioxide in pomegranate and observed a decline in leaf number, leaf area and chlorophyll content with increasing SO$_2$ level. They, however, did not record any significant variation in leaf mass under elevated SO$_2$ condition when compared with that of control (ambient SO$_2$). Use of aqueous SO$_2$ (100-500 ppm) caused chlorophyll breakdown in lodge pole pine under the laboratory conditions with chlorophyll a showing greater sensitivity to SO$_2$ than the chlorophyll b (Malhotra, 1977). On the contrary, (Pandey and Agrawal, 1994) exposed 45 day old tomato plants with SO$_2$ (0.1 ppm) and NO$_2$ (0.2 ppm) either alone and in combination for 4 h daily for 50 days and found an increase in plant height, number of leaves, total leaf area, total leaf biomass, total shoot biomass and total plant biomass which they attributed to a greater allocation of photoassimilates for growth and development of the photosynthetic organs under enriched availability of SO$_2$ and NO$_2$. In the present study, with a 30 day exposure duration a reduction in shoot mass and leaf area was measured across crops under the SO$_2$ enrichment treatment. (Wilson and Murray, 1990) exposed two week-old seedlings of wheat to 0·004, 0·042, 0·121, 0·256 or 0·517 μl litre$^{-1}$ SO$_2$ for 79 days, 4 h per day and observed a significant negative effect on all the growth attributes at SO$_2$ concentrations above and including 0·042 μl litre$^{-1}$.

**Yield attributes:** Variation in different yield attributes as affected by air pollutants in the growing environment is depicted in Fig 3 and Fig 4. Highest ear/pod number per plant was measured under the combined use of SPM and charcoal filters (T1) across the cereal (bread and durum wheat and barley) and legume (chickpea) crops, however, the observed increase in ear/pod number at T1 was insignificantly different (P> 0.05) from the values measured at control (T3).
charcoal and particulate filter (Removes particle pollutants); T2 (Treatment tunnel 2): Particulates filter (Removes particle pollutants); T3 (Treatment tunnel 3): No filter (Ambient control); T4 (Treatment tunnel 4): SOx enrichment (25μg m⁻³ over ambient). Data bars for the treatments, within a crop, depicting different letters are significantly different as per DMRT at p<0.05 n=3.

particularly for bread wheat, durum wheat and chickpea. A significant reduction in the average number of ears on per plant basis was measured at T2 when compared with all other conditions of growth across cereal and legume species. SO₂ enrichment treatment (T4), when compared with the ambient control (T3) caused an insignificant change in the number of ear/pod per plant for the investigated legume and cereal species except barley which showed a decline in pod number under the SO₂ fumigation treatment. The pattern of variation for this attribute was altered when expressed on per unit area basis (Fig 3). Ear/pod number per square meter under SO₂ fumigation treatment was significantly improved for the investigated cereals except barley and in legume crop, when compared with their performance under the ambient control treatment (T3). (Chauhan and Joshi, 2010) measured a significant reduction in growth and yield attributes of wheat and mustard in response to gaseous and particulate pollutants in the growing environment. Average ear/pod mass per plant across the experiment crops, in general, was higher, except for chickpea, under the combined use of SPM and charcoal filter (T1 treatment), which yields a growth environment free of particulate and gaseous pollutants such as PM 2.5, PM 10, SOx, NOx, ozone etc (Fig 3). Positive effect of SO₂ enrichment (T4) was measured for the durum wheat and chickpea crops while the effect was negative for barley and insignificant for bread wheat when compared to ambient (T3) and other environmental treatments (T1, T2). Bread wheat observed the highest ear mass under no filter control (T3). The results when viewed with the effect of SO₂ on shoot growth indicate a higher SO₂ sensitivity of bread wheat and barley at the reproductive than the vegetative stage. A look into the grain productivity variation in different crops under different growth environments indicates, in general, a negative response under T1 (SPM and charcoal filter) except chickpea. Variation in grain productivity between T2, T3 and T4 environments was insignificant for bread wheat while a significant increase in grain mass was measured at T2 condition over control in barley (Fig 3, 4). SO₂ enrichment (T4) caused a significant decline in grain productivity over the ambient control (T3) in chickpea while for bread wheat and barley the difference in grain mass accumulation per unit area between T3 and T4 was insignificant. Durum wheat, A significant decline in grain mass was measured in T1 (both charcoal and particulate filter treatment) over T3 (ambient control). Affect of SO₂ enrichment on the grain mass was highly significant only in case of the durum wheat. A similar reduction in growth and yield attributes in response to SO₂ exposure was measured by (Wilson and Murray, 1990) in wheat. Banks. A reduction in soybean yield and mineral nutrient content, but not the protein content was measured in the seeds with SO₂ fumigation (Sprugel et al., 1980).

Leaf and grain sulphur: Variation in leaf sulphur accumulation in response to gaseous and particulate pollution, measured at 60 days of crop growth and at harvest and grain S at the harvest stage are presented in Fig 5 and Fig 6. Data on leaf sulphur indicates a positive effect of SO₂ enrichment more prominently for bread wheat then the other experimental crops however, when expressed one per plant basis, shoot sulphur content was significantly increased under the enrichment treatment over control for both bread and durum wheat. Barley and chickpea showed marginal decline in sulphur accumulation in response to SO₂ exposure which was less significant than the other crops. A reduction in soybean yield and mineral nutrient content, but not the protein content was measured in the seeds with SO₂ fumigation (Sprugel et al., 1980).

Fig 4: Effect of gaseous and particulate pollutants on mean pod/spike mass and ear and pod number per plant at the harvest stage in cereals and legume crops under variable growing environments i.e., T1 (Treatment tunnel 1): Charcoal and particulate filter (Removes noxious gases and particle pollutants); T2 (Treatment tunnel 2): Particulates filter (Removes particle pollutants); T3 (Treatment tunnel 3): No filter (Ambient control); T4 (Treatment tunnel 4): SOx enrichment (25μg m⁻³ over ambient). Data bars for the treatments, within a crop, depicting different letters are significantly different as per DMRT at p<0.05 n=3.
environments in chickpea and barley at the harvest stage (Fig 5). Removal of gaseous and particulate pollutants, in general, did not affect the sulphur accumulation at harvest across crops. Grain S accumulation was higher in the absence of particulate and gaseous pollutant (T1) in bread wheat and barley when compared with their respective grain S levels under ambient (T4) condition. Use of particle filter (T2) surprisingly resulted in a lower grain sulphur accumulation in almost all crops, however, the treatment differences were significant for durum wheat and chickpea only. SO\textsubscript{2} enrichment caused a significantly higher accumulation of grain sulphur in bread wheat and chickpea while the effect was insignificant for barley and was significantly negative for durum wheat when compared with the ambient control (Fig 6). Low level of SO\textsubscript{2} exposure has been shown to have positive impact on plant growth and development (Lee et al., 2017). A highly significant positive correlation between shoot sulphur concentration and ambient SO\textsubscript{2} concentration was reported by (Wilson and Murray, 1990) in wheat. An increase in leaf SO\textsubscript{2}\textsuperscript{2-4}-S and a decrease in leaf P content in response to SO\textsubscript{2} exposure were reported in tomato by (Pandey and Agrawal, 1994). (Padhi et al, 2013) also measured an increase in leaf sulphur content with an increase in the levels of SO\textsubscript{2} fumigation in tomato. The observed increase in sulphur content of tissues with increasing level of SO\textsubscript{2} in the present and earlier reported studies, may be attributed to the ability of plants to metabolize and assimilate the excess SO\textsubscript{2} at cellular level, thus, leading to its accumulation in the leaf. Further a higher leaf S, in general, may increase the available

**Fig 5:** Effect of gaseous and particulate pollutants on S accumulation (A) leaf S concentration (µg S g\textsuperscript{-1} dw) and leaf S content (µg S plant\textsuperscript{-1}) in cereals and legume crops at 60 DAS under variable growing environments i.e., T1 (Treatment tunnel 1): Charcoal and particulate filter (Removes noxious gases and particle pollutants); T2 (Treatment tunnel 2): Particulates filter (Removes particle pollutants); T3 (Treatment tunnel 3): No filter (Ambient control); T4 (Treatment tunnel 4): SOx enrichment (25 µg m\textsuperscript{-3} over ambient). Data bars for the treatments, within a crop, depicting different letters are significantly different as per DMRT at p<0.05 n=3.

**Fig 6:** Effect of gaseous and particulate pollutants on tissue sulphur accumulation (A) Shoot S (µg S g\textsuperscript{-1} dw) and (B) Grain S (µg S g\textsuperscript{-1} grain/saad) at the harvest stage in cereals and legume crops under variable growing environments i.e., T1 (Treatment tunnel 1): Charcoal and particulate filter (Removes noxious gases and particle pollutants); T2 (Treatment tunnel 2): Particulates filter (Removes particle pollutants); T3 (Treatment tunnel 3): No filter (Ambient control); T4 (Treatment tunnel 4): SOx enrichment (25 µg m\textsuperscript{-3} over ambient). Data bars for the treatments, within a crop, depicting different letters are significantly different as per DMRT at p<0.05 n=3.
S pool for an efficient retranslocation of S from the senescing leaves to the developing sink, leading to a higher accumulation of S in the grains.

CONCLUSION

The present study assessed the effect of variably polluted growth environments and SO$_2$ fumigation on performance of three cereals and a legume species. SO$_2$ exposure, in general, benefitted the experimental crops in terms of dry matter and S accumulation and thus suggest that SO$_2$ fumigation at low levels can contribute to the plants’ S nutrition, and result in enhanced crop productivity, especially in plants growing in sulfur-deficient soils. Wheat particularly the durum wheat, is more likely to be benefitted under the elevated SO$_2$ condition of the environment. However, threshold values of these pollutants across crops need to be determined.

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