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Harmful effects of stone crushing dust pollution on pomegranate (*Punica granatum* L.) fruit plant

Muhammad Nasir Bazai¹, Saadullah Khan Leghari^{1*}, Saeed-Ur-Rahman Kakar¹, Saeed Ahmad Mughal¹, Manzoor Iqbal Khatak², Mahjabeen Tafuzal¹

¹Department of Botany University of Balochistan, Quetta, Pakistan

²Department of Chemistry University of Balochistan Quetta, Pakistan

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Abstract

This research was carried out on *Punicagranitum* L. grown near Abbaseen stone crushing plant at Kuchlak District Quetta of Balochistan, Pakistan. The investigation was aimed to understand the adverse effects of stone crushing dust pollution on morphological, physiological, growth, fruit quality and productivity of *Punica granatum* L.. The plants leaves and fruits were collected from three polluted sites with the distance of 200, 400 and 600 meters away from the stone crushing units respectively. Whereas a locality of 5000 meters away from stone crushing location comparatively clean air considered as a control sampling. Results showed that the dust load on the leaf surface was reported (1.16, 0.84, 0.62 and 0.13µg/cm²) at location 200, 400, 600 and 5000 m sites respectively. The site nearest (200 m) to the stone crushing units, contained highest dust amount showed significant destructive effects on the leaf physio-morphological characteristics, fruit yield and quality characteristics of *Punica granatum* L. The consequences of this study also revealed that a fine aerosol of stone dust is emitted from the crushing units has significantly effects on plant height (110.6-144 cm), Stem size (18.42-24.06cm) and tree canopy (108.6-142.0 cm²). Petiole length, leaf length, breadth and Leaf area near to the stone crushing units was found; 0.3-0.6 cm, 5.3-6.6 cm, 1.9-2.6cm and 6.2-7.2cm². Leaf relative water content (41.83-75.23%) and Fruit Moisture Content (58.21-77.43%) was also noted significantly less at polluted site. More number of closed stomata/ cm², decrease in photosynthetic rate and transpiration rate and stomatal conductance with an enhanced level of sub stomatal CO₂ concentration was also reported near the crushing unit with respect to the control site. Results also showed less chlorophyll contents at polluted site. Number of fruits/tree and Fruit Size was also noted significantly low as compared to control site (5000 m). From the result it was concluded that the distance is directly proportional to the growth, yield, physiological and morphological characteristics of this plant.

*Corresponding Author: Saadullah Khan Leghari ✉ drsaadullahleghari@gmail.com

Introduction

An important constituent related to groundwork expansion is the usage of crushed rock (e.g., for building and road construction). Through screening and crushing processes, the consequential dust is a key environmental air impurity that can have significant effects on nearby vegetation and wildlife (Leghari *et al.*, 2019a). Raina *et al.* (2008) reported that the Dust released from the crushing plants is hazardous to surrounding vegetation and by falling on the plant leaves cause injuries. The effects include a change in soil productivity and pH, decreased visibility in the neighboring areas, increased number of people with chronic respiratory illnesses and allergies, and degradation of natural habitats and resources, such as economic crops (Semban and Chandrasekhar, 2000; Das and Nandi, 2002; Mishra, 2004; Sivacoumar *et al.*, 2006). During the relatively simple stone crushing and screening process, large stones are broken into small pieces and sorted into different sizes which produce a large amount of dust that drifts over surrounding areas (Ravindran, 2013). This airborne dust falls onto the surrounding environment including plant leaves and people causing those adverse effects (Anon, 1999). Dust particulates accumulate on leaf surfaces causing a reduction in photosynthesis and stomatal activities and other foliar damages, all of which leads to a reduction in crop yield. A significant reduction on plant height, cover, number of leaves, and total chlorophyll content for *Vitis vinifera* L. was observed by Leghari *et al.*, (2014) due to roadside dust accumulation. Wallenborn *et al.*, (2009) found that stone crusher units add a significant amount of dust in the environment. Numerous investigations have revealed that the particulate matters settled down on the surface of the leaves of the plants due to gravitation and cause deterioration effects on the vegetation (Chatter, 1991). The dust is composed of homogenous mixtures as oxides of K, Ca, Al, N and S. (Raina *et al.*, 2008). It is important to monitor the effects of dust on vegetation. (Saini *et al.*, 2011). The accumulation of the dust on plants causes dangerous effects on the growth, yield, morphological and physiological features of the commercially valuable

plants (Mughal *et al.*, 2018). Significant reductions have been observed in the biomass of Apricots (*Prunus armeniaca* L.) fruits/tree, number of branches/tree and number of fruits/branch in stone dust pollution locations than those of the control sites. The results of this study were also revealed that the stones dust decreases the proximate and essential elements constituents at a considerable level (Leghari *et al.*, 2018). Yield of plants is decreased due to dust fall on leaves which leads to decrease photosynthesis, respiration and transpiration (Raina *et al.*, 2008; Saini *et al.*, 2011). Leghari *et al.*, (2019a) assess the effects of stone crushing dust pollution on three commonly cultivated fruit plant species (*Vitis vinifera* L., *Morus alba* L., and *Prunus armeniaca* L.) and on the health of workers working at crushing plants and found that the stone crushing units had high percentages of closed stomata both on the upper sides (Us) and lower sides (Ls) of leaves at 500-m distance from stone crushing installations. Results also showed that stone crushing workers suffered from symptoms of respiratory diseases (82.17%), allergies (72.13%), headaches (75.09%), coughing (78.36%), and tiredness (92.31%).

Plants are an integral part of life in many indigenous communities. A great attention has been given in developed countries about the effects of dust and metal toxicities on germination, growth of plant and their productivities. Stone dust and metal toxicity is an important factor governing germination of seeds, growth and productivity of plants. Therefore, the main objective of the study was to determine the effect of stone dust on Pomegranate (*Punica granatum* L.) Fruit plants locally grown nearby the stone crushing components in Kuchlakh district Quetta Balochistan, Pakistan.

Materials and methods

Study site and plant selection

The study was conducted in Kuchlakh area district Quetta, Balochistan during 2019. Kuchlakh area is situated at latitude 30°19'31"N and longitude 66°56'33"E (Fig.1). It receives 212mm rain fall per annum with annual average temperature remain 15.8

°C The altitude of the study area is about 1680 meters above sea level. Three sites (200 m, 400 m 600 m) near stone crushing units were selected for plant material and considered as polluted sites and comparatively less polluted site 5000 meter away from crushing unit was designated control site where the dust load was found zero. An orchard of Pomegranate (*Punica granatum* L.) fruit plants were selected to investigate the effects stone dust which were locally grown nearby the stone crushing components in Kuchlakh district Quetta Balochistan, Pakistan and these fruit plants (Pomegranate) were available at all four sites equally. In addition to that all the study sites had same physio-ecological conditions.

Collection of plant material

Mature areal parts (Leaf and fruits) of *Punica granatum* L. were collected from randomly selected 15 plants. All the plant samples were taken at 2-3m height from all four sides of the sampled trees at each study site as the methods used by Mughal *et al.*, (2018); Leghari *et al.*, (2018). Then these samples were brought to the laboratory in an ice can carefully for further analysis.

Leaf morphological characteristics

Leaf morphological characteristics as leaf length (cm) breadth (cm) and petiole length (cm) was determined by using ruler and leaf area (cm²) were determined by leaf Area Meter as described by Saha and Padhy (2012); Leghari and Zaidi, (2013).

Measurements of dust fall on leaf surface

For the measurement of dust fall on the leaf surface of selected plant was measured from particular leaves marked at the lower branches. The leaf surface was cleaned nicely by wet cotton. After 15 days marked leaves were taken and each of them was kept in pre weighed (b) poly packs. The weight of the leaf along with pack were noted (a). Then leaves were nicely washed with tap water in the laboratory of Botany University of Balochistan Quetta and were dried at room temperature. Clean foliage were then weighted (c). Deposited dust on leaf surface were calculated

with the help of formula $[a - (b + c)]$. After the interval of 15 days dust fall on leaves were measured during the whole study period then average were calculated and expressed as $\mu\text{g}/\text{cm}^2$ and samples were taken three time during while study (Gamiand Parel, 2015); Prusty *et al.*, 2005).

Yield and quality determination

Fruit plant yield was determined by fruit diameter and through counting the number of mature fruits at time of harvesting and sizes of fruits were measured by Vernier caliper as the method used by Olszyk, (1989); Mughal *et al.*, (2018). Fruit quality was determined by measuring fruit size, fruit moisture content % (FMC), fruit grain size, fruit grain taste and fruit grain colours as method described by Leghari *et al.*, (2019b); Prajapati and Tripathi, (2008) after some modifications.

Growth parameters

For the determination of growth parameters plant height (cm), stem diameter (cm) and Canopy (cm²) were measured. The plant height was noted by using Clinometer, stem diameter was measured by measuring tab and the canopy measured by using following formula:

$$\text{Tree canopy} = A \times B \quad (\text{Eq. 1})$$

Where A is the east to west cover and B is the north to south cover and the cover was noted by the use of measuring tape as used by Mughal *et al.*, (2018).

Leaf and fruit moisture content

Leaf and fruit moisture content was calculated by using the following formula as described by A.O.A.C (2005).

$$\text{Moisture content (\%)} = \frac{\text{Fresh weight} - \text{Oven dry weight}}{\text{fresh weight}} \times 100 \quad (\text{Eq. 2})$$

Leaf stomatal study

Leaves stomata was studied by method recommended by Salisbury, (1927) and reviewed by Radoglou and Jarvis, (1990). Imprints of upper (adaxial) and lower (abaxial) epidermis were isolated from central vein at

“maximum leaf width. Colorless nail polish and adhesive transparent tape was used for impression. Replica imprints isolated from the leaf epidermis were observed by using a microscope (Ernst Leitz GmbH Wetzlar, Type 20-446.023, Germany) at various magnifications (*50, *125 or *1250) for the studying of stomata. Minimum 5 microscopic regions was haphazardly chosen for each replica. Entire numbers of closed stomata/mm² were counted under the microscope and mean %age of 20 observations from each species was calculated.

Photosynthetic pigments

The chlorophyll contents (Chl. “a”, “b” and Total Chl.) was determined through Ultra-violet spectrophotometer and concentration was calculated as method used by Arnon, (1949) and carotenoid contents was noted as noted by Davis, (1976).

Gaseous exchange characteristics measurement

Few parameters of gaseous exchange i.e. rate of photosynthesis (A) stomatal conductance” (g_s), rate of transpiration (E) and sub-stomatal carbon dioxide concentration (C_i) were observed from younger and expanded foliage by utilizing LCA-4 ADC movable

infra-red gas analyzer (IRGA). All the measurements were taken during 10:00 am - 2:00pm.

Result and discussion

Effects on growth characteristics

Data depicted in the Table 1 revealed that the plant height (110.6, 120.4, 144 and 313 cm), Stem diameter size (18.42, 20.19, 24.06 and 36 cm) and tree canopy (108.6, 119.2, 142.0 and 217 cm²) varied significant at all four locations 200 m 400 m 600m and 5000 m.

This variation is directly proportional to the distance from the crushing units. Similar observation was also reported by Leghari *et al.*, (2019a) they found significant effects of stone crushing dust pollution on three fruit plant species (*Vitis vinifera* L., *Morus alba* L., and *Prunus armeniaca* L.) cultivated near the stone crushing units at three different distance sites (500, 1000, and 1500 m).

Similarly Mughal *et al.*, (2018) reported that the road side dust pollution causes reduction in plants height, stem size and tree canopy. A decline in tree cover of *Vitisvenifera*L was also observed by Leghari *et al.*, (2014) near road side plantation.

Table 1. Summary of pair sample t-test at 95% Confidence level for the variation in Plant growth characteristics.

Parameters	Distance from stone crusher			
		200m	400m	(600m) 5000 m (Control)
Plant Height(cm)	Mean	110.6±3.2	120.4±8.9	144±11 313±1.5
	T value	24.32	16.22	4.3
	Sig. (2 tailed)	0.001	0.002	0.012
Stem Size(cm)	Mean	18.42±1.04	20.19±1.67	24.06±2.06 36±2.1
	T value	18.22	9.33	3.2
	Sig. (2 tailed)	0.002	0.003	0.005
Tree Canopy(cm ²)	Mean	108.6±6.26	119.2±10.03	142.0±11.94 217±4.2
	T value	40.22	33.2	5.34
	Sig. (2 tailed)	0.002	0.012	0.018

Leaf morphological characteristics and dust accumulation

The leaf morphological characteristics are directly proportional to the distance from the crushing plant. Data illustrated in Table 2 exposed that the petiole length (0.3, 0.42, 0.6 and 0.8 cm), leaf length (5.3, 5.7, 6.6 and 7.3cm), Leaf breadth (1.9, 2.3, 2.6 and

2.9cm), Leaf area (6.2, 6.9, 7.2 and 9.6cm²) have significantly different at all four sites 200, 400, 600 and 5000m significantly. These result resemble with the findings of Pyatt and Haywood, (1989); Leghari *et al.*, (2019b) and Mughal *et al.*, (2018). Whereas, the dust load on plant leaf surface was (1.16, 0.84, 0.62 and 0.13µg/cm²) which is inversely proportional to

the distance from the crusher. Similar findings have been reported by Prajapati and Tripathi, (2008); Leghari *et al.*, (2019a); Shafiq *et al.*, (2009) and Preeti, (2000). Various plants have shown decrease in leaf length and breadth in highly polluted areas as reported by Leghari and Zaidi, (2013). Other

researchers Bahttiand Iqbal, (1988); Gupta and Ghous, (1988); Iqbal and Shafiq, (1999); Shafiqand Iqbal, (2003). Dineva, (2004) have also reported that the dust deposition causes decrease in leaf length, breadth and petiole length.

Table 2. Summary of pair sample t-test at 95% Confidence level for variation in leaf morphological characteristics and dust load on leaf area.

Parameters		Distance from stone crusher			
		200m	400m	600m	5000 m (Control)
Petiole Length (cm)	Mean	0.3±0.01	0.42±0.02	0.6±0.02	0.8±0.2
	T value	19.1	12.22	4.6	
	Sig (2 tailed)	0.003	0.006	0.034	
Leaf Length (cm)	Mean	5.3±0.28	5.7±0.34	6.6±0.46	7.3±0.01
	T value	22.2	18.23	8.21	
	Sig (2 tailed)	0.002	0.006	0.009	
Leaf Breadth (cm)	Mean	1.9±0.04	2.3±0.05	2.6±0.11	2.9±0.2
	T value	18.2	13.2	7.44	
	Sig (2 tailed)	0.003	0.005	0.013	
Leaf Area cm ²	Mean	6.2±0.18	6.9±0.19	7.2±0.33	9.6±1.5
	t value	22.1	16.4	8.2	
	Sig (2 tailed)	0.002	0.005	0.021	
Leaf Dust Content µg/cm ²	Mean	1.16±0.21	0.84±0.23	0.62±0.22	0.13±0.02
	t value	18.1	8.1	3.1	
	Sig (2 tailed)	0.003	0.019	0.166	

Fruit yield and quality

The facts reported in Table 3 revealed that number of fruits/tree (29.64, 56.52, 87.2 and 149) and Fruit Size (5.14, 5.64, 6.73 and 9.32cm) was varied significantly at all four locations 200, 400, 600 and 5000m respectively. Similarly, Mughal *et al.*, (2018) observed that the number of fruits/ plant and fruit size is increased as the dust deposition is decreased. The excessive influence of CO₂ content and leaf injuries

cause reduction in the yield and quality of fruits. A significant decrease was observed in fruit diameter of *Malus pumila*. Analogous findings in the biomass, number of fruit/tree, number of branches/tree, fresh weight of fruit and number of fruit/ branches in *Prunus armeniaca* L. due to stone crushing dust. Leghari *et al.*, (2018). Furthermore, Adbel-Rehman and Ibrahim, (2012) reported decrease in number of fruits in *Ficus carica* due to cement dust load.

Table 3. Summary of pair sample T-test at 95% Confidence level for the variation in yield and fruit quality.

Parameters		Distance from stone crusher			
		200m	400m	600m	5000m Control)
No of Fruits/ Tree	Mean	29.64±3.23	56.52±11.64	87.2±14.3	149±2.32
	t value	9.33	7.2	3.35	
	Sig (2 tailed)	0.004	0.016	0.034	
Fruit Size	Mean	5.14±0.25	5.64±0.41	6.73±0.51	9.32±1.3
	t value	20.4	15.22	4.3	
	Sig (2 tailed)	0.002	0.011	0.041	

Leaf physiological characteristics

The data given in Table 4 regarding leaf relative water content (75.23, 62.77, 56.54 and 41.83%) and fruit moisture content (58.21, 63.36 and 77.43%) were significantly different at all four investigated sites (200, 400, 600 and 5000 m) respectively. Moreover,

it was found that as the distance from the stone crushing increases the LRWC and FMC% also increases. Whereas the number of closed stomata/cm² (70.2, 50.88, 37.64 and 2.54) at location A, B, C and D in the leaves lower epidermis was decreased with respect to the distance.

Table 4. Summary of pair sample T-test at 95% confidence level for the physiological characteristics.

Parameters		Distance from stone crusher			
		200m	400m	600m	5000m Control)
Leaf relative water Content %	Mean	75.23±14.54	62.99±15.12	56.54±14.03	41.83±1.34
	t value	22.4	10.3	6.8	
	Sig (2 tailed)	0.002	0.018	0.033	
Fruit moisture content %	Mean	58.21±3.2	63.36±4.7	72.82±2.9	77.43±1.32
	t value	39.33	18.4	9.21	
	Sig (2 tailed)	0.001	0.002	0.006	
No; of closed stomata/cm ²	Mean	70.2±12.79	50.88±13.60	37.64±13.09	2.54±0.92
	t value	24.2	12.11	2.66	
	Sig (2 tailed)	0.001	0.009	0.018	

Table 5. Summary of pair sample T-test at 95% confidence level for photosynthetic, transpiration rate and water use efficiency characteristics.

Parameters		Distance from stone crusher			
		200m	400m	600m	5000m Control
Photosynthetic Rate µmole CO ₂	Mean	7.27±0.41	7.92±0.58	8.72±0.36	9.32±0.32
	t value	25.22	18.32	8.33	
	Sig (2 tailed)	0.002	0.004	0.008	
Transpiration Rate mmole H ₂ O	Mean	0.78±0.08	0.82±0.08	0.92±0.08	1.02±0.03
	t value	27.22	14.76	6.67	
	Sig (2 tailed)	0.004	0.013	0.062	
Water Use Efficiency µmole CO ₂ /mmole H ₂ O	Mean	9.36±0.47	9.68±0.41	9.47±0.69	9.15±0.32
	t value	5.37	5.2	5.11	
	Sig (2 tailed)	0.033	0.035	0.031	

These results of this experimental research agrees with the reports of Leghari *et al.*, (2011). They resulted that the relative moisture content increased at polluted site in *Fraxinus xanthoxyliods* and *Vitis vinifera*. Plants have shown variation in the RWC of from season to season and specie to specie Ogunkunle *et al.*, (2015) and Saura-Mas and Lloret, (2007). Similarly, Paulsamy *et al.*, (2000) reported that the increase in RWC may help the plant to maintain their physiological balance under dust pollution stress. The alteration number of opened and closed stomata

cause damage in the leaf and this alteration in stomatal density may reduce the physiological processes in the plant as described by Leghari *et al.*, (2015) during study plants near house hold waste burning sources in six plants.

Larcher, (1995) reported that the oxides of sulphur, Nitrogen and toxic particulate matters pierce in the photosynthetic tissues through stomatal pores and clog them which further effects the physiological processes.

Table 6. Summary of pair sample T-test at 95% confidence level for the plant leaf photosynthetic pigment Composition.

Parameters		Distance from stone crusher			
		200m	400m	600m	5000m (Control)
Chlorophyll a (mg/g.f.wt)	Mean	0.66±0.06	0.69±0.07	0.80±0.05	1.01±0.2
	t value	8.31	5.5	1.6	
	Sig (2 tailed)	0.016	0.021	0.137	
Chlorophyll b (mg/g.f.wt)	Mean	0.33±0.03	0.37±0.03	0.42±0.01	0.49±0.01
	t value	9.2	6.1	3.2	
	Sig (2 tailed)	0.02	0.028	0.000	
Total Chlorophyll (mg/g.f.wt)	Mean	0.99±0.06	1.06±0.10	1.22±0.05	1.5±0.3
	t value	8.2	5.1	1.2	
	Sig (2 tailed)	0.014	0.103	0.211	
Carotenoids (mg/g.f.wt)	Mean	0.29±0.3	0.36±0.02	0.41±0.03	0.49±0.02
	t value	11.5	7.86	3.99	
	Sig (2 tailed)	0.026	0.050	0.14	

Gas exchange characteristics

The findings in Table 5 illustrated that the photosynthetic rate (7.27, 7.92 8.92 and 9.32 $\mu\text{mole CO}_2$) and transpiration rate (0.78, 0.82, 0.92 and 1.02mmole H_2O) are significantly different in site 200, 400, 600 and 5000 m respectively. It is found that the photosynthetic rate and transpiration rate has increased as the distance has increased from the crushing plant. However, Water use Efficiency (9.36,

9.68, 9.47 and 9.15) at site 200, 400, 600 and 5000 m have not shown any significant alteration in plants at all sites. These findings are in the lines of the findings many researchers including Bhatti and Iqbal, (1988); Gupta and Ghous, (1988); Iqbal and Shafiq, (2000); Dineva, (2004); Mughal *et al.*, (2018) have reported that dust particles penetrate in the leaf photosynthetic tissues and result decrease in the gaseous exchange and morphological characteristics.

**Fig. 1.** Abbaseen stone crushing plant at Kuchlak Quetta.

Photosynthetic pigment composition

The consequences in Table 6 regarding Chlorophyll a at Site 200, 400, 600 and 5000 m were (0.66, 0.69, 0.80 and 1.01 mg/g.f.wt) chlorophyll b (0.33, 0.37, 0.42 and 0.49 mg/g.f.wt) and Total Chlorophyll 0.99, 1.06, 1.22 and 1.5 mg/g.f.wt), carotenoid content (0.29, 0.36, 0.41 and 0.49 mg/g.f.wt) at location 200 m, 400m, 600m and 5000m respectively. It was observed during the study that the chlorophyll content has increased with respect to the decrease in dust load on the leaf surface. Similarly many

researchers as Chauhan, (2010) found 43.36% reduction in chlorophyll a concentration in *Ficus religiosa*. Leghari *et al.*, (2011; 2014; 2015) found reduction in chlorophyll a, b in many plants. Missanjo *et al.*, (2015); Saini *et al.*, (2011) stated that it is due to decline in to phaeophtin by loss Mg ion.

Moreover, decrease in carotenoid is due to pollutants from releasing sources. Sharma and Tripathi, (2009) reported that carotenoid is a photo protective agent in chloroplast.



Fig. 2. Stone crushing dust on leaves and fruits *Punica granatum* L.

This chemical protects the chloroplast machinery against photo oxidative destruction. Many plants have shown decrease in Chlorophyll a, b and carotenoids as *Ficus religiosa* Mir *et al.*, (2008), *E. angustifolia* L. *Vitis vinifera* L. Leghari *et al.*, (2011; 2014), *Prosopis juliflora* Chauhan, (2010), *Artimisia maritime* Leghari *et al.*, (2015) and *Duranta repens* Leghari *et al.*, (2018).

Conclusion

It may be concluded that stone crushing dust cause substantial alteration to physiological, morphological and growth by destruction of leaf area and stomata. The high dust load with toxic pollutants and heavy metal influences the photosynthetic pigments and gaseous exchange process of *Punica granatum* L. These pollutants affect the qualitative and quantitative yield of trees. The vegetation grown in the vicinity of crushing plants has highest effects as

compared to the distanced vegetation.

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