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Urban dust load impact on gas-exchange parameters and growth of *Sophora japonica* L. seedlings

L. Bao[1,2], K. Ma[1], S. Zhang[1], L. Lin[1,2], L. Qu[1]

[1]State Key Laboratory of Urban and Regional Ecology, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing, P.R. China
[2]University of Chinese Academy of Science, Beijing, P.R. China

ABSTRACT

Green space interacts with particulate matters (PM) from urban atmosphere and road dust is a main source of urban PM pollution. We designed a dusting experiment of 21 days to examine the variations of pigments, gas exchange, height increments, and biomass of *Sophora japonica* seedlings (the most popular tree in Beijing) under 0, 1.45, 3.78, 9.02 and 16.64 g/m^2 urban road dust loads. Along with the rising dust loads, total chlorophyll and net photosynthetic rate had logarithmic droppings, from 1.37 ± 0.10 to 0.42 ± 0.03 mg/g and from 9.87 ± 0.18 to 5.69 ± 0.69 µmol/m^2/s, respectively. The root-shoot ratio of biomass logarithmically increased between 2.17 ± 0.51 and 2.73 ± 0.21. The height increments had a linear decline between 3.8 ± 1.0 and 1.5 ± 0.2 cm. Gas exchange parameters decreased fast in the initial dusting time and then reduced slowly or even increased. When dusted with the maximum foliar dust load of Beijing's street trees for 21 days, photosynthetic performance of *S. japonica* would decrease by 16%; height increment of *S. japonica* would decrease by 17%. Leaf biomass decline caused by dust might be a potential risk for street trees. Using plants like *S. japonica* as street trees or other green spaces could be a good measure to mitigate PM pollution.

Keywords: photosynthesis; emission; fly ash; heavy metals, plant stress

The emission of particulate matter (PM) is contributing to the decline of environmental quality in cities worldwide. Plants could reduce the airborne PM in urban areas (Nowak et al. 2006). The primary source of the PM is from the traffic (Tallis et al. 2011). Road dust can cause a series of negative effects on plant physiology, e.g. blocking the stomata and decreasing diffusive resistance (Farmer 1993), promoting the absorption of insolation and elevating the leaf temperature (Farmer 1993), lowering the photosynthetic pigments (Prusty et al. 2005, Prajapati and Tripathi 2008) and bringing down the net photosynthetic rate and stomatal conductance (Chaturvedi et al. 2013); negative effects of road dust on plant biomass and height were less studied.

Quantification studies of dust load impacts on plants were much fewer than those qualitative dust effect studies. It is not clear how the dust load of urban road dust impact urban plants. Loam and fly ash was applied on plant foliage to study the dust load impacts on photosynthesis or biomass (Armbrust 1986, Hirano et al. 1995, Singh and Siddiqui 2003). Iron dust was applied to soil surface to investigate the dust load impacts on stem heights and biomass of plants (Kuki et al. 2009). In those studies, photosynthesis or growth of plants were inhibited at high dust load, but may be promoted at low dust load. Therefore, it is important to find the limit of dust load that starts to restrain plant growth. Moreover, in most of the artificial simulations (Hirano et al. 1995, Singh and Siddiqui 2003), dust load treatments were arranged as doubled and redoubled that were unrelated to the actual field circumstance. It is necessary to detect the response of plant physiology and growth to the actual urban road dust load.

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Table 1. The amount of maximum dust load

	No. of samples	Mean	Minimum	Maximum	Standard deviation
Dust load (g/m^2)	10	16.97	13.84	20.49	0.68
Applied dust (g/pot)	10	73.08	61.27	84.85	2.43

Urban road dust carries high concentration of heavy metals, such as Cd, Cr, Cu, Pb and Zn (Charlesworth et al. 2003, Tang et al. 2013). Trace elements in PM can cause increasing concentration of those elements in plant shoots by covering foliage (Uzu et al. 2010, Pavlik et al. 2012). Those contaminants may enhance the dust load impacts of urban road dust on plants, therefore being different from impacts of other dusts.

Sophora japonica L. is the dominate street tree species in Beijing and takes up 81% of the urban street trees (Zheng and Zhang 2011). To understand the effects of road dust on the growth and photosynthesis of *S. japonica*, we set up a control experiment by artificially loading road dust collected from the surface street of Beijing. We suppose that with the increase of dust load, *S. japonica* seedlings will generate less pigments and gas exchange, leading to less accumulation of biomass. The detailed purposes of our work were to detect: (1) are *S. japonica* seedlings stressed significantly by the mean and max dust load of the street?; (2) are there some trends in the variations of photosynthesis and growth along with rising dust load and dusting time? Relevant knowledge would benefit the management of urban green space.

MATERIAL AND METHODS

Plant material and road dust collection. Seed germination was performed under greenhouse conditions at the Beijing Forestry University Forest Science Co, Ltd., in April 2012. *S. japonica* seedlings were obtained by the germination of seeds provided by the nursery of the Qingyuan county in Hebei province. 20 days after emergence, the seedlings were transferred to plastic pots containing cultivation soil (sand + peat + vermiculite in a proportion of 2:2:1). Each pot contained one plant. In August 2012, seedling heights were 30–50 cm, and the crown widths were about 30 cm.

The road dust used in the experiment was collected from the surface of some busy traffic streets between the 2nd Ring and 3rd Ring roads of Beijing. The road dust was sieved with a nylon fine screen to get the particles smaller than 106 μm in diameter (Armbrust 1986).

Maximum dust load experiment and treatment setting. The seedlings were irrigated and their leaves were washed three days before artificial dusting. We placed plastic wrap on the soil surface to avoid the dust retention. Road dust (diameter < 106 μm) was applied by using a dust sprayer until the leaf surface could not accommodate any more dust. The maximum applied dust (A_m) was then recorded. Leaves of 10 replications were collected to count the maximum dust load (D_m) (Table 1).

In September 2012, the dusting experiment was conducted including five treatments, and each treatment had nine replicate seedlings. In our previous study, the dust load of *S. japonica* in streets was significantly higher than that of *S. japonica* in parks, and the dust load of street tree *S. japonica* had a maximum value of 2.812 g/m^2 and an average value of 0.683 g/m^2 (Dai et al. 2012). Therefore, we regarded the average dust load of street trees as A_1 treatment, and according to the ratio of maximum applied dust and maximum dust load, $A_1 = 0.683A_m/D_m = 2.94$ (g/pot). Likewise, we regarded the maximum dust load of street trees as A_2, $A_2 = 2.812A_m/D_m = 12.11$ (g/pot). We defined half of the maximum dust load as A_3, $A_3 = 1/2A_m = 36.53$ (g/pot). Maximum dust load was regarded as A_4, $A_4 = 73.08$ (g/pot). We took A_0 treatment

Table 2. The dust load amounts of treatments in experiment

Treatment	Applied dust (g/pot)	Expected dust load (g/m^2)	Actual dust load (g/m^2)
A_0	0	0	0
A_1	2.94	0.683	1.45
A_2	12.11	2.812	3.78
A_3	36.53	8.485	9.02
A_4	73.08	16.97	16.64

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with no dust as control. The dust load amounts of treatments are shown in Table 2.

The seedlings were dusted by the method of maximum dust load experiment and kept stable for 21 days. The irrigation water was only applied to the soil surface directly by tubule. Immediately after dusting, three seedlings per treatment were cut down to test their actual dust load. Whole leaves from each seedling were washed in distilled water and ultrasonic oscillated for 3 min, and then vacuum filtered through pre-weighed filter membrane (Φ = 0.45 μm) (Freer-Smith et al. 1997). After drying, the leaf areas of these washed leaves were measured by the WinFOLIA Basic 2004a (REGENT Instruments Inc., Australia). The actual dust loads were similar to the expected dust loads, with a proximity of $r = 0.998$, so we used actual dust loads when curve fitting in this article.

Growth, pigments, and gas exchange measurements. The heights of seedlings were measured before dusting and after 21 days of dusting. When the experiment finished, three seedlings from each treatment were excavated and washed in water. The roots, stems, and leaves were separated and oven-dried at 80°C for at least 24 h to obtain dry weight biomass.

The second, third, and fourth leaves from the top of each sprig were collected and put into liquid nitrogen using three seedlings for each treatment. Fresh leaves were extracted with 95% ethanol and the absorbance of the leaf pigment extract were measured separately at 470, 649, and 665 nm. Chlorophyll and carotenoid were calculated according to the specific absorption coefficients of Lichtenthaler (1987).

Gas exchange parameters (net photosynthesis rate (P_n), stomatal conductance (g_s), transpiration rate (E) and intercellular CO_2 concentration (c_i)) were measured using a portable photosynthesis system LI-6400 (LiCor Inc., Lincoln, USA). Four fully expanded healthy leaves (the third leaves from twig top) were measured for each seedling, and three seedlings were used for each treatment. Quantum flux was arranged as 800 μmol/m^2/s (6400-02B Red-Blue LED, LiCor Inc.), and ambient humidity (60 ± 10% relative humidity), temperature (25 ± 2°C) and atmospheric CO_2 concentration (400 ± 20 μmol/mol) were used. The measurements were made before dusting and after 2, 4, 6, 12, 16, and 20 days of dusting, and between 8:00 a.m. and 10:00 a.m. of those days.

Statistical analyses. Treatments were compared by the *LSD* (least significant difference) test of one-way ANOVA or the *LSD* test of repeated measures in GLM. Statistical analyses were done by the SPSS 15.0 (IBM, Armonk, USA).

RESULTS

Dust load impact on growth. Leaf biomass of all four dust load treatments was significantly lower than A_0 (Figure 1a). The leaf biomass of A_3 was the lowest. Shoot biomass (leaf biomass + stem biomass) of A_1 showed a significant decrease compared with A_0 (Figure 1b). The decrease of shoot biomass was greater than that of root biomass, which contributed to the increase of the root-shoot ratio. The root-shoot ratio had a logarithmic rising trend with dust loads (Figure 2a). In contrast, the height increment had a linear downtrend (Figure 2b).

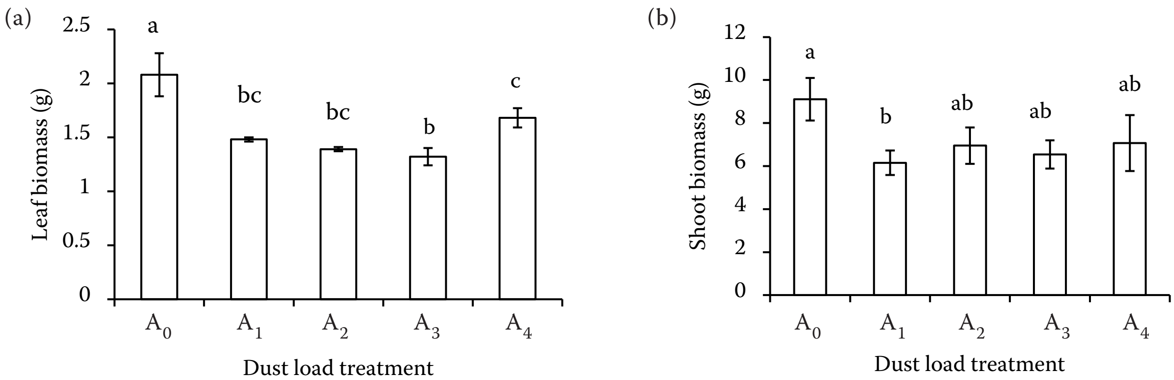

Figure 1. Biomasses of dust load treatments. (a) Leaf biomass; (b) shoot biomass. Different letters indicate significant differences by the least significant difference test ($P \leq 0.05$). A_0 – control; A_1 – 1.45; A_2 – 3.78; A_3 – 9.02; A_4 – 16.64 g/m^2

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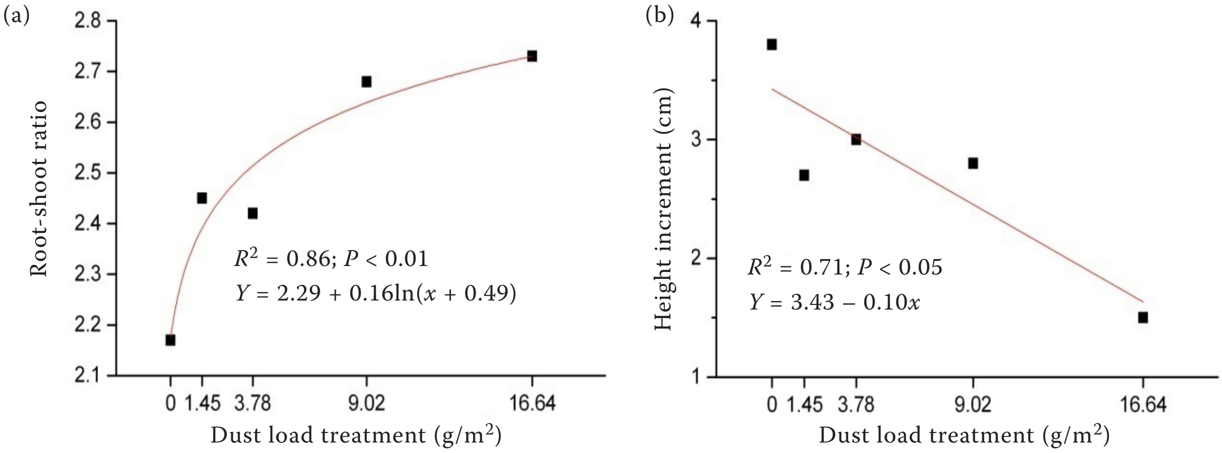
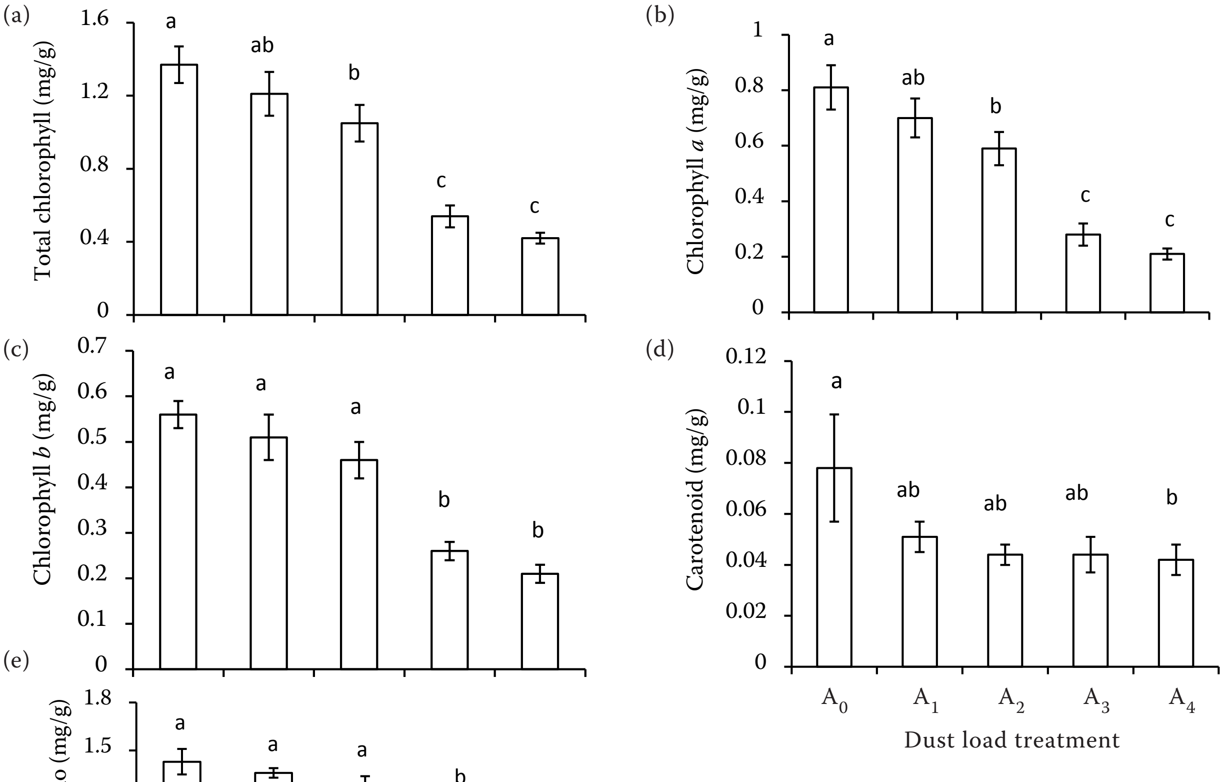


Figure 2. Dust load effects on growth. (a) Root-shoot ratio; (b) height increment

When compared to A_0, leaf biomass decreased by 29% in A_1 treatment, and by 33% in A_2 treatment; height increment decreased by 29% in A_1 treatment, and by 21% in A_2 treatment.

Dust load impact on pigment contents. Total chlorophyll and chlorophyll *a* of A_2, A_3, and A_4 were lower than those of A_0 (Figures 3a,b). Chlorophyll *b* and chlorophyll *a*/*b* of A_3 and A_4 showed significant



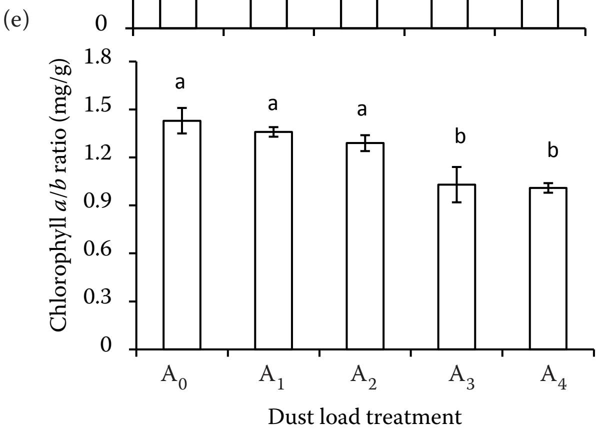


Figure 3. Pigments of dust load treatments. (a) Total chlorophyll; (b) chlorophyll *a*; (c) chlorophyll *b*; (d) carotenoid; (e) chlorophyll *a*/*b* ratio. Different letters indicate significant differences by the least significant difference test ($P \leq 0.05$). A_0 – control; A_1 – 1.45; A_2 – 3.78; A_3 – 9.02; A_4 – 16.64 g/m²

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differences from A_0 (Figures 3c,e). Carotenoid of A_4 was significantly lower than that of A_0 (Figure 3d). Chlorophyll *a* was more sensitive to dust treatment than chlorophyll *b* and carotenoid. A_4 treatment was the lowest group for all pigment parameters. A logarithmic function ($Y = 2.25 - 0.62\ln(x + 3.99)$, $R^2 = 0.94$, $P < 0.01$) was fitted to describe the relationships between total chlorophyll and dust load. When compared to A_0, total chlorophyll decreased by 12% in A_1 treatment, and by 23% in A_2 treatment.

Dust load and dusting time impact on gas exchange. P_n, g_s, and E of A_2, A_3, and A_4 were significantly lower than A_0 (Figures 4a,b,c), and c_i of all four treatments were significantly lower than A_0 (Figure 4d). P_n and E decreased fast in the initial four days and varied slightly afterwards. g_s and c_i kept declining rapidly during the initial 12 days, then varied smoothly. At the end of dusting, the minimum value of four parameters all appeared in A_4 treatment. Dusting for 6 days with A_1 treatment reduced P_n by 9%, and with A_2 treatment reduced P_n by 17%. Dusting for 20 days with A_1 treatment reduced P_n by 9%, and with A_2 treatment reduced P_n by 20%. A logarithmic function ($Y = 13.34 - 2.52\ln(x + 3.94)$, $R^2 = 0.99$, $P < 0.001$) was fitted to describe the relationships between P_n (the 20th day) and dust load. P_n varied with the dusting time in power functions (Table 3).

DISCUSSION

Dust load and dusting time impact on photosynthesis and growth of *S. japonica*. The dust load effect on total chlorophyll and net photosynthetic rate had a logarithmic declines with dust loads in our study. When cucumbers and kidney beans were dusted with Kanto loam, their P_n dropped in line with the dust loads (Hirano et al. 1995). Effects of fly ash on pigments of wheat increased and then deceased with the dust loads (Singh and

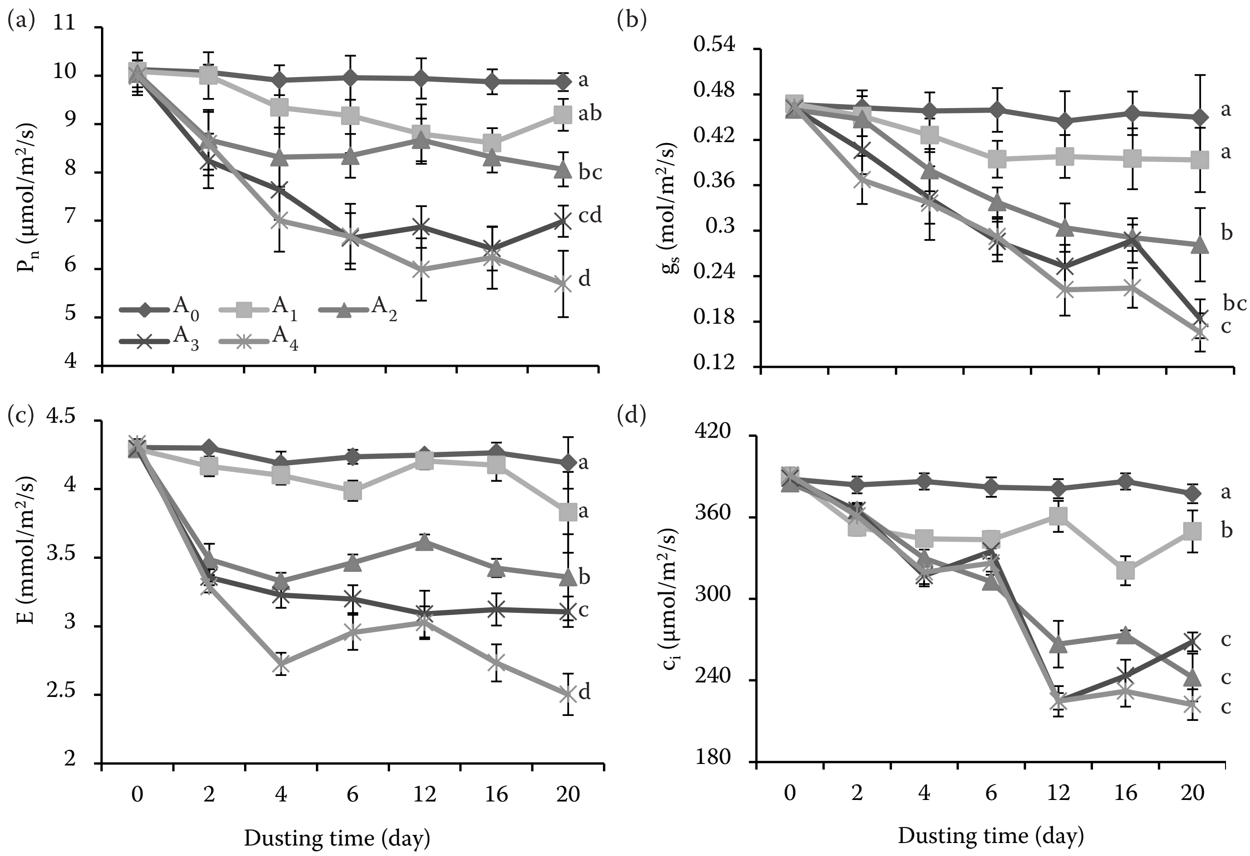

Figure 4. Gas exchange variation of dust load treatments with dusting time. (a) Net photosynthesis rate (P_n); (b) stomatal conductance (g_s); (c) transpiration rate (E); (d) intercellular CO_2 concentration (c_i). Different letters indicate significant differences by the least significant difference test of repeated ANOVA ($P \leq 0.05$). A_0 – control; A_1 – 1.45; A_2 – 3.78; A_3 – 9.02; A_4 – 16.64 g/m²

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Table 3. Net photosynthetic rate variations with dusting time

	Dust load treatment (g/m^2)	Probability > *F*	Adj. *R*-square	Regression function
Net photosynthetic rate	A_0	< 0.001	0.80	$Y = 10.12(1 + x)^{-0.01}$
	A_1	< 0.001	0.73	$Y = 10.19(1 + x)^{-0.05}$
	A_2	< 0.001	0.66	$Y = 9.64(1 + x)^{-0.06}$
	A_3	< 0.001	0.86	$Y = 9.71(1 + x)^{-0.14}$
	A_4	< 0.001	0.95	$Y = 10.02(1 + x)^{-0.19}$

A_0 – control; A_1 – 1.45; A_2 – 3.78; A_3 – 9.02; A_4 – 16.64 g/m^2

Siddiqui 2003). Armbrust (1986) tried to set up a linear regression equation between cotton photosynthetic rate and dust concentration of loam (Armbrust 1986).

In the case of the dust load effect on growth, the root-shoot ratio of biomass had a logarithmic increase and the height increment was a linear decline with dust loads. The increase of the root-shoot ratio exhibited a change of biomass allocation between above- and underground at road dust stress. When road dust stress was aggravated, *S. japonica* distributed more biomass to root than to crown, so growth of stem height may be delayed. Thus the height increment dropped with the dust load. In Kuki's research (2009), stem height of *S. tomentosa* significantly declined when soil iron dust load was elevated, but *S. terebinthifolius* did not (Kuki et al. 2009). Biomass increased and then deceased with the rising dust load in wheat and *S. tomentosa* experiments (Singh and Siddiqui 2003, Kuki et al. 2009). Biomass fluctuated and then decreased, and a linear regression equation was set up to describe the cotton' s dry weight and dust concentration of loam (Armbrust 1986).

As for the dusting times effects, the gas exchange parameters decreased fast during the initial dusting time and then reduced slowly or even increased. This was probably because in the later period the leaves had an expanded growth so the dust effects were reduced a little. In Pereira's iron dust research (2009), the g_s decreased fast in the initial 10 days, but the P_n reduced only a little in 30 days (Pereira et al. 2009). Cotton photosynthetic rates of 1, 3, 7, and 14 days were compared in Armbrust's study (1986).

Implications to urban green space management. Though researchers found that pigment content, P_n, and g_s of roadside plants negatively correlated with dust loads (Prusty et al. 2005, Prajapati and Tripathi 2008, Chaturvedi et al. 2013), how much dust load could significantly affect the photosynthesis and growth has not been explored so far.

In our research, net photosynthetic rate declined with dust loads by function of $Y = 13.34 – 2.52\ln(x + 3.94)$, according to which we could calculate the consequence of different dust loads. When dusted with the average foliar dust load (0.683 g/m^2) of Beijing's street trees, P_n of *S. japonica* would decrease by 6% in 20 days; and dusted with the maximum dust load (2.812 g/m^2), P_n of *S. japonica* would decrease by 16%. Reductions in photosynthesis of street trees may decrease the regulating services of green space.

We substituted dust loads of the Beijing city into the regression functions of height increment $Y = 3.43 – 0.1x$. We concluded that height increment of *S. japonica* would decrease by 12% with average foliar dust load of Beijing in 20 days; and decrease by 17% with maximum foliar dust load of Beijing. Furthermore, dust loads of 1.45 g/m^2 reduced leaf biomass by 29%; dust loads of 3.78 g/m^2 reduced leaf biomass by 33%. Though there was no obvious trend, leaf biomass decline caused by dust might be a potential risk for street trees. Loss of street tree crowns may influence the supporting service of green space.

In conclusion, along with the rising urban dust load, total chlorophyll and net photosynthetic rate had logarithmic droppings, from 1.37 ± 0.10 to 0.42 ± 0.03 mg/g and from 9.87 ± 0.18 to 5.69 ± 0.69 $\mu mol/m^2/s$, respectively. The root-shoot ratio of biomass logarithmically increased between 2.17 ± 0.51 and 2.73 ± 0.21. The height increments had a linear decline between 3.8 ± 1.0 and 1.5 ± 0.2 cm. Gas exchange parameters decreased fast in the initial dusting time and then reduced slowly or even increased. When dusted with the maximum foliar dust load of Beijing's street trees for 21 days, photosynthetic performance of *S. japon-*

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ica would decrease by 16%; height increment of *S. japonica* would decrease by 17%. Leaf biomass decline caused by dust might be a potential risk for street trees. Using plants like *S. japonica* as street trees or other green spaces could be a good measure to mitigate PM pollution.

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Corresponding author:

Mr. Keming Ma, Chinese Academy of Sciences, Research Center for Eco-Environmental Sciences, State Key Laboratory of Urban and Regional Ecology, 18 Shuangqing Road, Haidian District, 100 085 Beijing, P.R. China
e-mail: mkm@rcees.ac.cn