

Effects of O₃ stress on photosynthesis of trees

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Abstract. The effects of elevated O₃ (O₃) on photosynthetic characteristics of urban trees were studied in an open-top air chamber. The results showed that net photosynthetic rate (*P_n*), transpiration rate (*E_t*), stomatal conductance (*G_s*) and water use efficiency (WUE) of different tree species decreased significantly with the increase of O₃ concentration, while intercellular CO₂ (*C_i*) concentration of different tree species increased significantly with the increase of O₃ concentration. Under different O₃ concentrations, the net photosynthetic rate, transpiration rate, stomatal conductance and intercellular CO₂ concentration of *Ginkgo biloba* (*G. biloba*) and *Koelreuteria paniculata* (*K. paniculata*) were higher than those of *Pinus bungeana* (*P. bungeana*) and *Platycladus orientalis* (*P. orientalis*), and the WUE of *P. bungeana* and *P. orientalis* was higher than that of *G. biloba* and *K. paniculata*. The effect of elevated O₃ concentration on plant photosynthesis is more obvious in *K. paniculata* and *G. biloba*.

Key words: open-top air chamber; O₃ stress; Photosynthesis; Stomatal conductance

I. Introduction

Ozone (O₃) concentration in many regions and cities in China is still rising continuously, and the extreme value of hourly O₃ concentration in big cities such as Beijing and Shanghai has reached over 300ppb, exceeding the third-class air quality standard of 120ppb in China (Li, 2016). Photosynthesis is a necessary process for plant growth (Wang et al., 2017). O₃ can inhibit the photosynthesis of most plants, and the increase of O₃ concentration will lead to the decrease of photosynthesis of plants (Li, 2016). Scholars found that the increase of O₃ concentration destroys the membrane system of plants, reduces the source of atmospheric CO₂, photoelectron transmission and plant photosynthesis (Xiong et al., 2017; Fu et al., 2014); The net photosynthetic rate of *Pinus tabulaeformis* leaves under high concentration O₃ treatment decreased significantly with the extension of treatment time (Zhang et al., 2007); Niu (2012) studied the photosynthesis of *Cinnamomum camphora* seedlings under O₃ stress and found that the photosynthetic rate and stomatal conductance of leaves decreased significantly. Xiong et al. (2017) studied the photosynthetic physiological characteristics of *Catalpa catalpa* under O₃ stress, and found that the net photosynthetic rate of *Catalpa catalpa* leaves gradually decreased with the extension of O₃ concentration treatment time, while the intercellular CO₂ concentration, transpiration rate and stomatal conductance showed a trend of first decreasing and then increasing with the increase of O₃ concentration.

It can be seen that most of the above studies are single species of broad-leaved tree species, and few studies on photosynthesis under O₃ stress have been done on needle-leaved tree species of multiple tree species at the same time. Therefore, in this study, the open-top air chamber method was used to measure the photosynthesis indexes of plants under different O₃ concentrations, and four landscaping tree species were taken to explore the effects of elevated O₃ concentration on the photosynthetic system of plant leaves, so as to provide reference for the study of adaptation mechanism of plants under O₃ stress.

2 Research method

2.1 Study area

The study area is located in Beijing Botanical Garden.

2.2 Experimental setup

The experimental setup method is the same as that of Chen et al (2021) .

2.3 Assay of photosynthetic characteristics

Net photosynthetic rate ($P_n/\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), transpiration rate ($E_t/\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), intercellular CO₂ concentration ($C_i/\mu\text{mol}\cdot\text{mmol}^{-1}$), stomatal conductance ($G_s/\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and photosynthetic active radiation ($\text{PAR}/\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)

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$\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) were measured by using CI340 photosynthetic apparatus. From May to October, we selected the relatively complete leaves of each seedling, and measured them from 9:00 to 15:00 every month on sunny days. The leaves of two different parts were selected for determination twice in three repetitions under each treatment. At the same time, water use efficiency (WUE) use the following formula to calculate.

$$WUE = \frac{P_n}{E_t}$$

Where WUE is water use efficiency ($\mu\text{mol}\cdot\text{mmol}^{-1}$), P_n is net photosynthetic rate ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), E_t is transpiration rate ($\text{nmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$).

3 Results and analysis

3.1 Changes of net photosynthetic rate under ozone stress

It can be seen from the above that the net photosynthetic rate of *G. biloba* and *K. paniculata* is higher than that of *P. bungeana* and *P. orientalis* (Figure 1). Under different O_3 concentrations, the net photosynthetic rate of *P. bungeana* ranges from $3.30\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ to $4.06\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and *P. orientalis* ranges from $4.05\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ to $4.64\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$. The net photosynthetic rates of *K. paniculata* and *G. biloba* ranged from $6.05\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ to $7.46\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and from $5.90\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ to $7.19\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. With the increase of O_3 concentration, the net photosynthetic rate of plants decreased significantly, especially in *K. paniculata* and *G. biloba*. Compared with NF8040 O_3 concentration, the net photosynthetic rate of *P. bungeana* and *P. orientalis* decreased by 9.88% and 15.83%, respectively. The net photosynthetic rate of *K. paniculata* and *G. biloba* decreased by 10.33% and 18.46%, respectively.

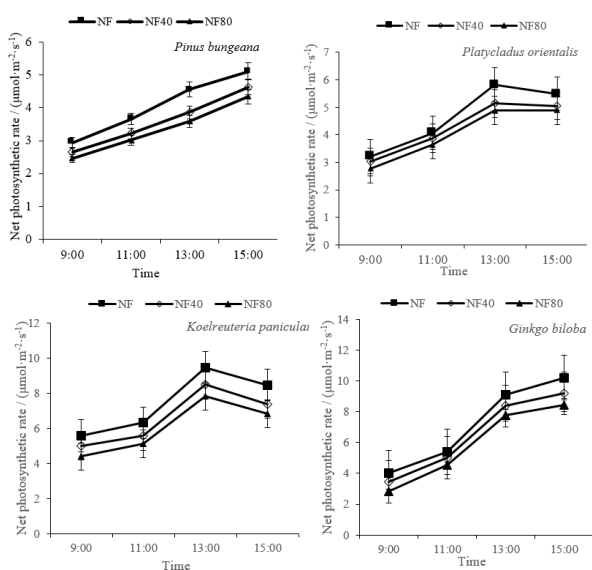
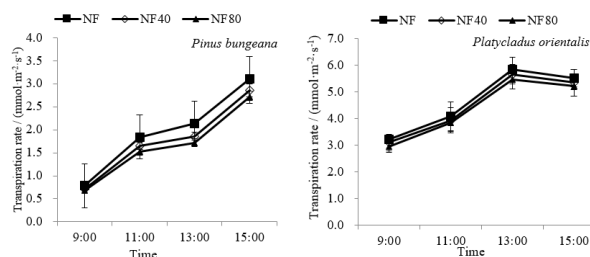


Figure 1. The variation of trees photosynthetic rate under different O_3 concentrations

3.2 Effects of O_3 stress on transpiration rate of trees

The effect of elevated O_3 concentration on transpiration rate of different trees is shown in figure 2. During the experiment, the transpiration rate of different tree species showed an increasing trend from 9:00, and the transpiration rate of different tree species decreased. The daily average transpiration rates of *P. bungeana* under different O_3 concentrations NF, NF40 and NF80 were $1.97\pm 0.74\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, $1.77\pm 0.68\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $1.66\pm 0.64\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. Compared with NF, the transpiration rates of *P. bungeana* under NF40 and NF80 decreased respectively. The daily average transpiration rates of *P. bungeana* under NF, NF40 and NF80 were $1.97\pm 0.74\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, $1.77\pm 0.68\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $1.66\pm 0.64\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. Compared with NF, the transpiration rates of *P. bungeana* under NF40 and NF80 decreased $0.20\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $0.31\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. After the experiment, the transpiration rates of *P. bungeana* decreased by 9.99% and 15.63% under NF40 and NF80, respectively. Under the O_3 concentrations of NF, NF40 and NF80, the daily average transpiration rates of *P. orientalis* were $4.67\pm 1.06\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, $4.52\pm 1.04\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $4.39\pm 1.03\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively; compared with the O_3 concentration of NF, they decreased by $0.16\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $0.29\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively, under NF40 and NF80. After the experiment, the transpiration rates of *P. orientalis* decreased by 3.34% and 6.12% under the O_3 concentration of NF40 and NF80, respectively. Under the O_3 concentrations of NF, NF40 and NF80, the daily average transpiration rates of *K. paniculata* were $7.47\pm 1.40\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, $6.66\pm 1.53\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $6.26\pm 1.38\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. Under the O_3 concentrations of NF40 and NF80, compared with NF, the transpiration rates of *K. paniculata* decreased $0.81\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $1.21\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. After the whole experiment period, the transpiration rates of *K. paniculata* decreased by 10.88% and 16.20% respectively under the O_3 concentration of NF40 and NF80. Under the O_3 concentrations of NF, NF40 and NF80, the daily average transpiration rates of *G. biloba* were $7.28\pm 2.56\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, $6.50\pm 2.55\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $6.25\pm 2.57\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. Under the O_3 concentrations of NF40 and NF80, compared with NF, the transpiration rates of *G. biloba* decreased $0.77\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $1.02\text{mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. After the whole experiment, the transpiration rates of *G. biloba* decreased by 10.64% and 14.07%, respectively, under the O_3 concentration of NF40 and NF80.



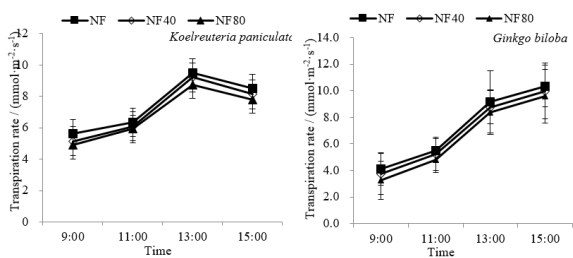


Figure 2. The variation of trees transpiration rate under different O₃ concentrations

3.3 Changes of stomatal conductance under ozone stress

It is found that the values of stomatal conductance of *G. biloba* and *K. paniculata* were greater than that of *P. bungeana* and *Pl. orientalis*(Figure 3). The increase of O₃ concentration significantly reduced the stomatal conductance of plants, which was more obvious in *K. paniculata* and *G. biloba*. Compared with the O₃ concentration of NF, the average decrease rates of stomatal conductance of *P. bungeana* and *P. orientalis* were 7.21% and 15.30%, while those of *K. paniculata* and *G. biloba* were 15.62% and 33.28%, respectively. Under O₃ concentrations of NF40 and NF80, the decrease rates of stomatal conductance of *K. paniculata* and *G. biloba* were 2.17 and 2.18 times that of *P. bungeana* and *P. orientalis*, respectively.

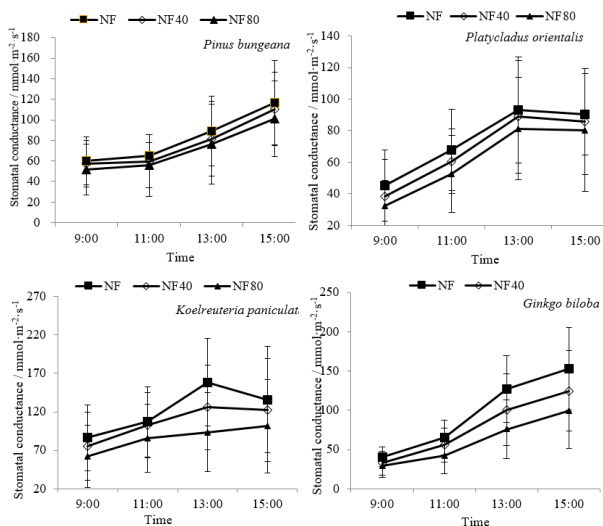


Figure 3. The variation of trees stomatal conductance under different O₃ concentrations

3.4 Effects of O₃ stress on intercellular CO₂ concentration in trees

The effect of elevated O₃ concentration on intercellular CO₂ concentration of different tree species was shown in figure 4. During the experiment, the intercellular CO₂ concentration of different tree species showed an increasing trend from 9:00. Under different O₃ concentration, the daily average values of intercellular CO₂ concentration of *P. bungeana* were

233.43±106.92μmol·mmol⁻¹, 260.86±106.02 μmol·mmol⁻¹ and 310.83±94.04 μmol·mmol⁻¹, respectively. Compared with the O₃ concentration of NF, the intercellular CO₂ concentration of *P. bungeana* increased by 27.43μmol·mmol⁻¹ and 77.40 μmol·mmol⁻¹, respectively. The values of intercellular CO₂ concentration of *G. biloba* and *K. paniculata* were higher than that of *P. bungeana* and *P. orientalis*. Under O₃ concentrations (NF, NF40 and NF80), the intercellular CO₂ concentrations of *P. bungeana*, *P. orientalis*, *K. paniculata* and *G. biloba* ranged from 233.43μmol·mmol⁻¹ to 245.89μmol·mmol⁻¹, from 197.63 μmol·mmol⁻¹ to 417.36μmol·mmol⁻¹ and from 219.48μmol·mmol⁻¹ to 355.38μmol·mmol⁻¹, respectively. The increase of O₃ concentration significantly increases the intercellular CO₂ concentration of plants, which is more obvious in *K. paniculata* and *G. biloba*. Compared with the O₃ concentration of NF, the average increase rates of intercellular CO₂ concentration of *P. bungeana* and *P. orientalis* were 10.65% and 28.79%, while those of *K. paniculata* and *G. biloba* were 19.70% and 50.99%, respectively. It can be seen that under the O₃ concentration of NF40 and NF80, the increase rates of intercellular CO₂ concentration of *K. paniculata* and *G. biloba* are 1.85 and 1.77 times that of *P. bungeana* and *P. orientalis*, respectively.

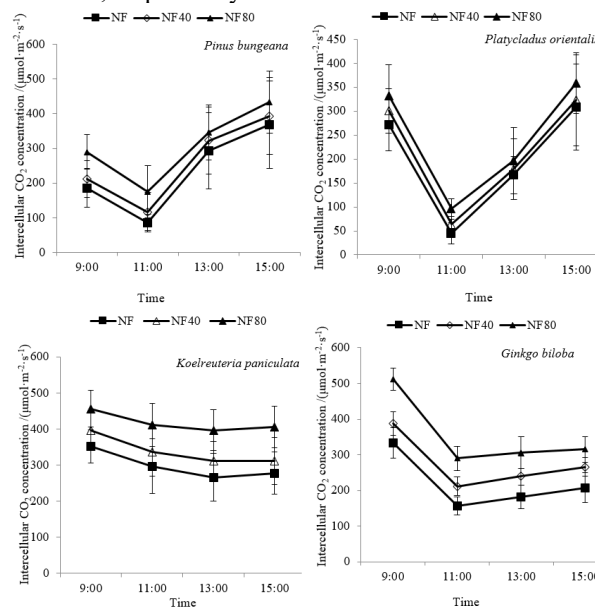


Figure 4. The variation of trees intercellular CO₂ concentration under different O₃ concentrations

3.5 Effects of O₃ stress on WUE of trees

It can be seen from the above that the *WUE* of *P. bungeana* and *P. orientalis* is greater than that of *G. biloba* and *K. Paniculata* (figure 5). The *WUE* values of *P. bungeana* and *P. orientalis* were greater than that of *G. biloba* and *K. paniculata*. Under different O₃ concentrations (NF, NF40 and NF80), the *WUE* of *P. bungeana* ranged from 2.29μmol·mmol⁻¹ to 2.37μmol·mmol⁻¹; The *WUE* of *P. orientalis* ranged from 0.93 μmol·mmol⁻¹ to 0.99μmol·mmol⁻¹, the *WUE* of *K.*

paniculata ranged from $0.92\mu\text{mol}\cdot\text{mmol}^{-1}$ to $1.00\mu\text{mol}\cdot\text{mmol}^{-1}$, the *WUE* of *G. biloba* ranged from $0.92\mu\text{mol}\cdot\text{mmol}^{-1}$ to $0.99\mu\text{mol}\cdot\text{mmol}^{-1}$. The *WUE* of plants decreased significantly with the increase of O_3 concentration, which was more obvious in *K. paniculata* and *G. biloba*. Compared with the O_3 concentration of NF, the average decrease rates of *WUE* of *P. bungeana* and *P. orientalis* were 2.87% and 5.32%, while that of *K. paniculata* and *G. biloba* were 4.05% and 7.11%, respectively. Under the O_3 concentration of NF40 and NF80, the *WUE* reduction rates of *K. paniculata* and *G. biloba* were 1.41 and 1.34 times that of *P. bungeana* and *P. orientalis*, respectively.

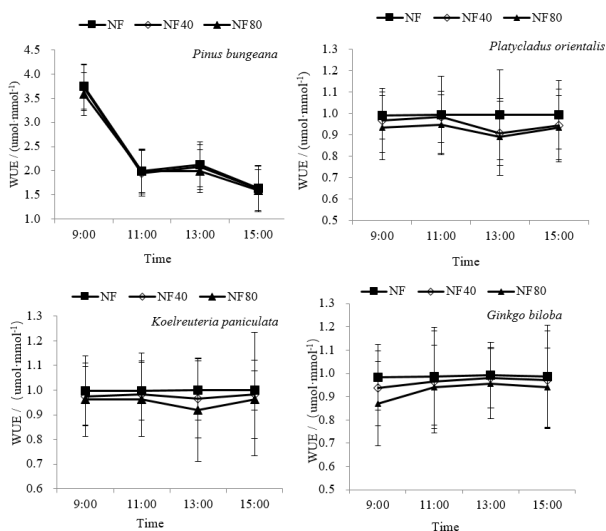


Figure 5. The variation of trees WUE under different O_3 concentrations

4 Discussion

The effect of O_3 on photosynthesis of trees has attracted much attention. Leaf is the main place for photosynthesis. Photosynthesis is a physiological process that plants are sensitive to the increase of O_3 concentration. Under high O_3 concentration, the net photosynthetic rate, transpiration rate, stomatal conductance and water use efficiency of trees decrease obviously. Compared with the air filtration control, continuous treatment with 80ppb O_3 for 90 days resulted in a decrease in photosynthetic rate of *Quercus mongolica* by more than 50% (Yan et al., 2010); The net photosynthetic rate of *Metasequoia glyptostroboides* seedlings fumigated with 100ppb O_3 decreased significantly by 41% (Feng et al., 2008). Richardson particularly pointed out that the photosynthesis rate will obviously decrease seasonally in summer (Xin, 2016). Wittig et al. (2007) found that with the increase of O_3 concentration, the photosynthetic rate and stomatal conductance of tree leaves decreased by 11% and 13% respectively. Guo (2001) set up four OTCs with different gradients, and found that when the O_3 concentration increased to 50nL/L, 100nL /L and 200nL/L, the photosynthetic rate of different plant leaves decreased by 2%, 19% and 46% respectively, compared with that under normal environmental O_3 concentration,

indicating that the photosynthetic rate of plants decreased significantly with the increase of O_3 concentration. Similar studies have reached similar conclusions in wheat (Akhtar et al., 2010). The above results are consistent with the results of this study, which shows that the increase of O_3 concentration can obviously inhibit the photosynthesis of leaves of trees. Elevated O_3 will first cause the stomata of plant leaves to close. When the stomata of plant leaves close, the amount of CO_2 entering the plant decreases correspondingly. When the amount of CO_2 decreases, the amount of CO_2 absorbed by plant photosynthesis also decreases, thus reducing the photosynthetic rate of plants. At this time, it also causes changes in other photosynthetic factors. The decline rate of photosynthesis parameters among different tree species is different, which is caused by the difference of different tree species (Cao et al., 2012; Sarkar et al., 2010) is determined by the sensitivity of different species to O_3 (Bussotti et al., 2007). In this study, the net photosynthetic rate of *P. bungeana* decreased by 11.71% and 18.81% respectively under the O_3 concentration of NF40 and NF80, while that of *G. biloba* decreased by 9.39% and 18.01% respectively. This is also consistent with Li (2014) finding that there are significant differences in O_3 stress resistance of seedlings of *Du Ying apiculus*, *Mytilaria laosensis* and *Castanopsis fissa* under different O_3 concentrations.

In this study, other photosynthetic parameters decreased with the increase of O_3 concentration, and only intercellular CO_2 concentration increased significantly with the increase of O_3 concentration. The reason was that non-stomatal factors of plants also affected the change of photosynthesis. The results are consistent with the research of Xin et al. (2016) on the relationship between photosynthetic characteristics and O_3 dose of different poplar genotypes, and it is found that the intercellular CO_2 concentration of three poplar genotypes decreases with the increase of O_3 concentration under different O_3 concentrations. With the increase of O_3 concentration, the relationship between intercellular CO_2 concentration (C_i) and net photosynthesis was inversely correlated, and the higher the intercellular CO_2 concentration, the lower the photosynthetic rate. When the intercellular CO_2 concentration is high, the photosynthetic rate decreases. Relevant studies have pointed out that when the stomatal conductance of plants decreases and the intercellular CO_2 concentration remains unchanged or increases, the decrease of net photosynthetic rate should be caused by non-stomatal factors such as the reduction of mesophyll cell assimilation ability (Xin et al., 2016); Only when stomatal conductance and intercellular CO_2 concentration decrease at the same time can the decrease of net photosynthetic rate be mainly caused by stomatal limitation (Farquhar G D and Sharkey T D., 1982). The results of this study showed that when O_3 concentration increased, stomatal conductance of different tree species decreased, but the change law of intercellular carbon dioxide concentration is opposite, which indicated that the decrease of photosynthetic rate was also affected by non-stomatal limitation. Therefore, the influence of O_3 concentration increase on plant photosynthetic rate was the result of combined action of stomatal limitation and non-stomatal limitation.

5 Conclusion

The net photosynthetic rate, transpiration rate, stomatal conductance and WUE of different tree species decreased significantly with the increase of O₃ concentration, while the intercellular CO₂ concentration of different tree species increased significantly with the increase of O₃ concentration. Under different O₃ concentrations, the net photosynthetic rate, transpiration rate, stomatal conductance and intercellular CO₂ concentration of *K. paniculata* and *G. biloba* were higher than those of *P. bungeana* and *P. orientalis*, and their water use efficiency was higher than that of *K. paniculata* and *G. biloba*. The effect of O₃ concentration on plant photosynthesis is more obvious in broad-leaved trees.

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