Effect of Ozone fumigation on morpho-physiology and yield of field-grown rice in Pearl River Delta, China

Qiong Wang1,2,3, Bilige Sude2,3*, Ming Dong2,3,4, Yuan Shi2,3, Chunmei Geng2 and Xiaoke Wang5

1College of Water Sciences, Beijing Normal University, Beijing 100875, China.
2Chinese Research Academy of Environmental Sciences, Beijing 100012, China.
3State Environment Protection key Laboratory of Regional Eco-process and Function Assessment, Beijing 100012, China.
4College of Geographical Science, Inner Mongolia Normal University, Hohhot 010022, China.
5State Key Laboratory of Urban and Regional Ecology, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing 100085, China.

Received 3 January, 2013; Accepted 2 March, 2013

Ozone pollution may cause a reduction in grain crops production. The response relationship between plants and Ozone is an important factor to assess the effect of Ozone pollution. In order to learn the effect of Ozone pollution on major grain crops in Pearl River Delta, this study used Open-top chamber (OTC) to fumigate the rice in the field, and researched the effects on the physical manifestations, physiological form and yield of rice which was fumigated by different Ozone concentrations. According to the experimental results, with increase in Ozone fumigation gradient, the rice was damaged gradually. And the height, biomass and yield of rice were reduced. By fitting concentration response model and dose response model, it can be estimated that the Ozone concentration and Ozone AOT40 value for 10% rice yield loss of single cluster were 61.7 nL/L and 13.08 h nL/L respectively in this experimental area.

Key words: Modeling, Open-top chamber (OTC), Ozone, rice, production loss, AOT40.

INTRODUCTION

With development of urbanization and living condition of the Chinese people, urban air pollution become more and more serious, especially near ground-level Ozone pollution problems (Zhang et al., 1998). As a strong oxidant (Yang, 2001), Ozone has serious harmful effects on plants (Calatayud et al., 2003; Liang et al., 2010; Zheng et al., 2007, 2009; Fuhrer and Booker, 2003; Reich et al., 1986) and is threatening the yield of grain crops (Chameides et al., 1999; Wang et al., 2007; Chen et al., 2007) in China.

Both foreign and domestic scholars have conducted experiments on the effect of Ozone exposure on grain...
crops by means of laboratory control, field experiment, and open-top chamber (Felzer et al., 2007; Wang et al., 1993). The effect of Ozone on grain crops are mainly damaging physical characteristics (Yao et al., 2007) and physiological adjustment system (Zheng et al., 2007) of grain corps, reducing plant photosynthesis efficiency (Zheng et al., 2009, 2007), and affecting grain crop biomass and yield (Zheng et al., 2009, 2007; Xie et al., 2007). It is of significant importance to correctly assess the effect of Ozone pollution on grain crops. Wang and Guan (1995) improved the model of correlation between the Ozone concentration and crop loss rate, and assessed the effect of Ozone pollution on yield of major grain crops in China (Aunan et al., 2000). Scholars in America and Europe discovered that the effect of Ozone on grain crops was an accumulated result and put forward dose response model. United Nations Economic Commission for Europe (UNECE) determined that the critical Ozone concentration to harm grain crops is 40 nL/L and has established an AOT40 index. The method to calculate AOT40 (Fuhrer et al., 1997) is as follows (Equation 1):

$$AOT40 = \sum (C_0 - 40) \quad C_0 \geq 40 \text{nL/L}$$  \hspace{1cm} (1)

In the formula, AOT40 is the accumulated Ozone concentration exposure value when hour average Ozone concentration is greater than 40 nL/L, \((\text{h} \cdot \text{mL} / \text{L})\). \(C_0\) is the hour average Ozone concentration value when solar radiation is greater than 50 W·m⁻², (nL/L).

### Gradient design

The fumigation device which manufactured by our team included octahedral cylinder OTC, ventilation, Ozone production, concentration control and automatic monitoring systems. The volume of OTC is 10 m³, air exchange frequency is greater than twice per minute. There are four gradients for fumigation.

i) Non-filter gradient (NF): Directly connected to non-filter air.

ii) 40 gradient (NF+40): Based on connecting to the air, add some Ozone through Ozone generator to make the Ozone concentration equal to atmospheric Ozone +40 nL/L.

iii) 80 gradient (NF+80): Based on connecting to the air, add some Ozone to make the concentration equal to atmospheric Ozone +80 nL/L.

iv) 120 gradient (NF+80): Based on connecting to the air, add some Ozone to make the concentration equal to atmospheric Ozone +120 nL/L.

Every gradient has three parallel OTC, and there are altogether 12 OTC fumigation chambers. The concentrations of the four gradients for setting the highest value are no restriction, 80, 120 and 160 nL/L, respectively. Fumigation was conducted from 9:00 to 17:00 every day beginning from April 21, 2009. There are 44 days for fumigation.

### Measuring method and time

**Morphologic measurement:** 10 clusters of rice were selected from every chamber and marked. Their heights in different phenophases were measured and their leaf colors observed.

**Biomass measurement:** Three clusters of rice growing in the same condition were selected from each chamber to measure their number and weight; they were separated according to organs (stem, healthy leaf, withering leaf and ear) and their dry weight after drying at 65°C for 48 h obtained.

**Yield measurement:** Rice of 1 m² at every chamber in mature stage were selected and their number of clusters recorded. From their ears, we obtained their dry weight after drying at 65°C for 48 h. The data was analysed by SPSS13.0 statistical software. The measured phenophase and time of various indexes are shown at Table 1.

### MATERIALS AND METHODS

#### Experimental region

Dongguan city is selected as experimental region in Guangdong Province. Experimental region is located in the east of Pearl River Delta, the average annual temperature of Dongguan is 23.1°C with rich sunlight and rainfall 20 (Chen, 2005). The experimental material is the Yuejingsi Seed No. 2 Rice that has been widely planted in Pearl River Delta. These were sowed on March 7 and transplanted to field on March 28, 2009. The area of experimental field is about 2 MU. Rice seedlings were transplanted manually to ensure they were evenly planted.

#### Gradient design

The fumigation device which manufactured by our team included octahedral cylinder OTC, ventilation, Ozone production, concentration control and automatic monitoring systems. The volume of OTC is 10 m³, air exchange frequency is greater than twice per minute. There are four gradients for fumigation.

i) Non-filter gradient (NF): Directly connected to non-filter air.

ii) 40 gradient (NF+40): Based on connecting to the air, add some Ozone through Ozone generator to make the Ozone concentration equal to atmospheric Ozone +40 nL/L.

iii) 80 gradient (NF+80): Based on connecting to the air, add some Ozone to make the concentration equal to atmospheric Ozone +80 nL/L.

iv) 120 gradient (NF+80): Based on connecting to the air, add some Ozone to make the concentration equal to atmospheric Ozone +120 nL/L.

Every gradient has three parallel OTC, and there are altogether 12 OTC fumigation chambers. The concentrations of the four gradients for setting the highest value are no restriction, 80, 120 and 160 nL/L, respectively. Fumigation was conducted from 9:00 to 17:00 every day beginning from April 21, 2009. There are 44 days for fumigation.

### Measuring method and time

**Morphologic measurement:** 10 clusters of rice were selected from every chamber and marked. Their heights in different phenophases were measured and their leaf colors observed.

**Biomass measurement:** Three clusters of rice growing in the same condition were selected from each chamber to measure their number and weight; they were separated according to organs (stem, healthy leaf, withering leaf and ear) and their dry weight after drying at 65°C for 48 h obtained.

**Yield measurement:** Rice of 1 m² at every chamber in mature stage were selected and their number of clusters recorded. From their ears, we obtained their dry weight after drying at 65°C for 48 h. The data was analysed by SPSS13.0 statistical software. The measured phenophase and time of various indexes are shown at Table 1.

#### Table 1. Records of phenophase and time of different project.

<table>
<thead>
<tr>
<th>Time</th>
<th>Phenophase</th>
<th>Determination of project</th>
</tr>
</thead>
<tbody>
<tr>
<td>April 10</td>
<td>Reviving stage</td>
<td>Morphologic measurement</td>
</tr>
<tr>
<td>April 17</td>
<td>Tillering stage (1)</td>
<td>Morphologic measurement</td>
</tr>
<tr>
<td>April 29</td>
<td>Tillering stage (2)</td>
<td>Morphologic and biomass measurements</td>
</tr>
<tr>
<td>May 12</td>
<td>Jointing stage</td>
<td>Morphologic and biomass measurements</td>
</tr>
<tr>
<td>May 30</td>
<td>Booting stage</td>
<td>Morphologic measurement</td>
</tr>
<tr>
<td>June 11</td>
<td>Booting and heading stage</td>
<td>Biomass measurement</td>
</tr>
<tr>
<td>June 16</td>
<td>Heading stage</td>
<td>Morphologic measurement</td>
</tr>
<tr>
<td>June 24</td>
<td>Filling stage</td>
<td>Morphologic and biomass measurement</td>
</tr>
<tr>
<td>July 15</td>
<td>Mature stage</td>
<td>Morphologic, biomass and yield measurements</td>
</tr>
</tbody>
</table>
Table 2. Records of visible damage on rice under Ozone fumigation.

<table>
<thead>
<tr>
<th>Phenophase</th>
<th>NF</th>
<th>NF+40</th>
<th>NF+80</th>
<th>NF+120</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reviving stage and tillering stage (1)</td>
<td>Healthy leaves</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tillering stage (2)</td>
<td>Healthy leaves</td>
<td>Healthy leaves</td>
<td>Speckles appear on few leaves</td>
<td>Speckles appear on some leaves</td>
</tr>
<tr>
<td>Jointing stage</td>
<td>Healthy leaves</td>
<td>Speckles appears on few leaves</td>
<td>Speckles appears on some leaves</td>
<td>Speckles appears on 10% of the leaves</td>
</tr>
<tr>
<td>Booting stage</td>
<td>Speckles appears on few leaves</td>
<td>Speckles appears on some leaves</td>
<td>Speckles appears on 10% of the leaves</td>
<td>Speckles appears on 15% of the leaves</td>
</tr>
<tr>
<td>Heading stage</td>
<td>Few leaves wither</td>
<td>10% of the leaves wither</td>
<td>15% of the leaves wither</td>
<td>20% of the leaves wither</td>
</tr>
<tr>
<td>Filling stage</td>
<td>10% of the leaves wither</td>
<td>15% of the leaves wither</td>
<td>20% of the leaves wither</td>
<td>30% of the leaves wither</td>
</tr>
<tr>
<td>Mature stage</td>
<td>30% of the leaves wither</td>
<td>50% of the leaves wither</td>
<td>80% of the leaves wither</td>
<td>90% of the leaves wither</td>
</tr>
</tbody>
</table>

Table 3. Height of rice under Ozone fumigation(cm).

<table>
<thead>
<tr>
<th>Phenophase</th>
<th>NF</th>
<th>NF+40</th>
<th>NF+80</th>
<th>NF+120</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reviving stage</td>
<td>15.1±2.8b*</td>
<td>15.0±2.6b</td>
<td>16.8±3.1a</td>
<td>15.7±3.3ab</td>
</tr>
<tr>
<td>Tillering stage (1)</td>
<td>27.7±3.3b</td>
<td>30.6±4.0a</td>
<td>30.2±3.7a</td>
<td>30.6±3.7a</td>
</tr>
<tr>
<td>Tillering stage (2)</td>
<td>52.2±5.4b</td>
<td>56.7±5.7a</td>
<td>54.1±4.8ab</td>
<td>55.2±7.3a</td>
</tr>
<tr>
<td>Jointing stage</td>
<td>77.3±4.2a</td>
<td>76.4±5.2a</td>
<td>76.5±5.1a</td>
<td>75.1±3.1a</td>
</tr>
<tr>
<td>Booting stage</td>
<td>109.3±6.3a</td>
<td>105.6±5.0ab</td>
<td>102.5±6.0bc</td>
<td>99.0±8.2c</td>
</tr>
<tr>
<td>Heading stage</td>
<td>117.8±5.6a</td>
<td>118.7±6.1a</td>
<td>112.4±5.3b</td>
<td>105.4±5.5c</td>
</tr>
<tr>
<td>Filling stage</td>
<td>121.8±4.8a</td>
<td>120.3±5.2a</td>
<td>112.9±4.9b</td>
<td>107.0±6.0c</td>
</tr>
<tr>
<td>Mature stage</td>
<td>115.4±6.6a</td>
<td>113.3±6.3ab</td>
<td>106.3±13.9bc</td>
<td>101.1±7.6c</td>
</tr>
</tbody>
</table>

*The letters in the figure mean the comparing results at 5% level. Different letters indicate distinctive difference and the same letters indicate indistinctive difference.

RESULTS AND ANALYSIS

Effect of Ozone on morphologic physiology of rice

Leaves are important photosynthesis organs for plants and are playing an important role in the growth of plant. Constant Ozone fumigation makes obvious visible signs of damage on leaves. According to Table 2, in the period of reviving and tillering stages (1), the leaves of the rice at all the four gradients are peak green. In tillering stage 2 (fumigating for 9 days), there are different signs of damage on leaves at different gradients. Leaves are healthy at NF and NF+40. There are some speckle in leaves at NF+80. The speckles are very obvious on rice leaves at NF+120. With continuity of Ozone fumigation and increase of concentration gradient, the morphological damages of the rice are becoming more and more serious. At NF+120 of the mature stage, most of the rice withered. 30% of the rice at NF withered. In the whole growth period, there are distinctive differences in morphologic damages of rice leaves at different Ozone fumigation gradients.

Ozone affects rice height to certain extent. It can be concluded that the heights at NF+80 and NF+120 are distinctively greater than those at NF and NF+40 in reviving stage and tillering stage(1) (Table 3). In tillering stage(2), the heights of rice at high fumigation gradients is still greater than those at NF. The heights among gradients are no significant different in jointing stage. The rule begins to change in rice booting stage. The heights of rice at high fumigation gradients is still less than those at low gradients. After heading stage, rice at NF+120 is much less than that at other gradients; In filling stage and mature stage, the trend of height in different fumigation gradients is that: NF > NF+40> NF+ 80> NF+ 120. The above show that high-concentration Ozone fumigation has dwarfing effect on rice. The followings have the same rule.

Effect of Ozone fumigation on rice biomass

According to Table 4, rice biomass does not have
distinctive differences at different gradients in tillering stage (2), jointing stage, and booting and heading stage. However, rice biomass declines with the increase of Ozone gradients after being fumigated for a certain time. In filling stage, rice biomass at NF is obviously higher than that at other gradients. The trend of rice biomass in different gradients is: NF > NF+40> NF+ 80> NF+ 120. Ozone fumigation can affect allocation of biomass in various organs (Figure 1). The allocation percentage of biomass in different organs has no significant difference in tillering stage (2) and jointing stage. In booting and heading stage, the dry weight of ear at NF+80 and NF+120 is higher than that at NF and NF+40. On the contrary, the dry weight of stem and leaf at NF+80 and NF+120 is lower than that at NF and NF+40. In filling stage, the dry weight of stem and green leaf at NF and NF+40 is higher than that at NF+80 and NF+120. The trend of weight in ear, stem and green leaf in different gradients is that: NF > NF+40> NF+ 80> NF+ 120. The high-concentration Ozone fumigation reduced the weight of each organ.

**Effect of Ozone fumigation on rice yield**

High-concentration Ozone fumigation can obviously decrease the single rice yield and unit area yield of grain crops and vegetables (Zheng et al., 2007). In experiment, with the increase of Ozone fumigation gradients, the yield of single cluster rice declines greatly. In particular, the effect of high Ozone fumigation gradients on rice yield is more obviously (Figure 2).

**Rice yield loss model assessment**

There are three type of models for assessing the relationship between Ozone pollution and yield loss, statistic model, mechanism model, and photochemical model (Zheng and Wang, 2004; Yao et al., 2007). Statistic model mainly includes concentration response model and dose response model. In reference to the research result (Heck et al., 1982) improved by Wang and Guan (1995), we establish the relation model between yield loss of a single cluster rice and Ozone pollution, the model is that:

\[ Y = aX + b \]

In this formula, Y is yield loss percentage of single cluster of rice; X is the hour average concentration of Ozone in growth season of grain crop; a and b are regression parameters.
Figure 2. Yield of single cluster of rice under Ozone fumigation.

Figure 3. Concentration response model of yield loss of single cluster of rice under Ozone fumigation.

Figure 4. Dose response model of yield loss of single cluster of rice under Ozone fumigation.

Conclusions

In the experiment, high-concentration Ozone fumigation has harmful morphologic effect on rice leaves such as making them wither and bringing speckles to them. As a result, rice height decreases with increase in fumigation gradient. The oxidability of Ozone destroys the normal physical structure of rice leaves directly, and decreases activity of relevant enzymes and chloroplast pigments in leaves. Ozone also can affect photosynthetic efficiency of plants and reduce the yield of crops finally.

Physical damage on leaves appeared firstly. It can be observed from tillering stage (2) to mature stage. The effect of Ozone fumigation on height appears later than physical damage. High-gradient have distinctive effect on crop height in booting stage. The dwarfing effect by high-concentration Ozone has not been observed until booting stage. The influence of organ biomass allocation start from booting and heading stage.

Effect of Ozone fumigation on rice biomass postpones to filling stage and eventually affects the rice yield. It can be concluded that the high-concentration Ozone fumigation on grain crops is caused by accumulation effect and it will have better effect using dose response model to predict yield loss of crops (Massman, 2004).

Average values of atmospheric Ozone concentration and Ozone AOT40 surveyed in experimental area in rice growth season are 50 nL/L and 3.19 h·μL/L, which do not exceed model fitted values (61.7 nL/L and 13.08 h·μL/L). Also, Rice production is not seriously threatened by Ozone pollution in Dongguan city.

The maximum of Ozone concentration of atmospheric in experimental area is 99.7 nL/L. This situation suggests that extremum of Ozone concentration still exceed the critical concentration that can cause rice yield decreasing by 10%. Therefore, Ozone pollution in this area affect...
rice yield to certain extent and will become more and more serious with the development of social economy.

In the summer of 2006, the large field comprehensive air quality survey experiment was conducted by Lu et al. (2010) in Pearl River Delta. In this survey, the average Ozone concentration value is 40.53 nL/L at 17 surveying stations that covered the whole Pearl River Delta. The maximum average value they surveyed is 132.18 nL/L. It suggests that the Ozone pollution condition is very severe in Pearl River Delta. The economic loss incurred by regional grain crop yield decline shall not be neglected. Therefore, studies on the effect mechanism of Ozone on rice should be conducted further and various research methods should be enriched in order to accurately and scientifically estimate the effect of increase of Ozone concentration on rice yield loss.

Conflict of Interests

The author(s) have not declared any conflict of interests.

ACKNOWLEDGEMENT

The authors would like to acknowledge the support of the Special Fund for Public Service Sector of National Environmental Protection of China (No. 200809152; No. 200809077).

REFERENCES


