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# Effects of elevated $O_3$ concentration on winter wheat and rice yields in the Yangtze River Delta, China

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ABSTRACT

The effects of a continuing rise of ambient ozone on crop yield will seriously threaten food security in China. In the Yangtze River Delta, a rapidly developing and seriously air polluted region in China, innovative open-top chambers have been established to fumigate winter wheat and rice in situ with elevated $O_3$. Five years of study have shown that the yields of wheat and rice decreased with increasing $O_3$ concentration. There were significant relationships between the relative yield and AOT40 (accumulated hourly $O_3$ concentration over 40 ppb) for both winter wheat and rice. Winter wheat was more sensitive to $O_3$ than rice. $O_3$-induced yield declines were attributed primarily to 1000-grain weight and harvest index for winter wheat, and attributed primarily to grain number per panicle and harvest index for rice. Control of ambient $O_3$ pollution and breeding of $O_3$ tolerant crops are urgent to guarantee food security in China.



## 1. Introduction

Ozone ($O_3$) is a strong oxidative pollutant that is detrimental to crop yields, tree production and human health. Trends in tropospheric $O_3$ concentration have leveled off or decreased slightly in North America and Europe (IPCC, 2007), but the trends are increasing in east Asian countries, such as Japan (Naja and Akimoto, 2004), and China (Shao et al., 2006; Wang et al., 2007). Long-term monitoring has shown that annual mean ambient $O_3$ concentration increased by 0.23 ppb per year from 1995 to 2005 over Mt. Waliguan in Qinghai province, China (Deliger and Zhao, 2007). Results from 26 established global atmospheric chemistry-transport models driven by analyzed meteorological fields have shown that global annual mean surface $O_3$ concentrations will increase by 1.5 ppb (CLE scenario)–4.3 ppb (IPCC SRES A2 scenario) over the period 2000–2030 (Dentener et al., 2006). In China, rapid development of the economy and society and fast urbanization will cause ambient $O_3$ concentration to rise faster than in other countries due to increasing emissions of trace gases such as nitrogen dioxide and volatile organic compounds.

Investigations into the effects of $O_3$ on crops have been carried out over four decades, especially in developed countries. For example, the US National Crop Loss Assessment Network (NCLAN) and European Open Top Chamber Programme (EOTCP) have shown significant yield losses of major crops in USA and Europe. Investigations of crop responses to elevated $O_3$ concentration in terms of phenology, physiology, production and yield have confirmed that elevated $O_3$ could accelerate crop senescence, decrease photosynthesis, reduce biomass and carbon allocation into roots, and cause yield loss (Morgan et al., 2003; Andersen, 2003; Fiscus et al., 2005; Ashmore, 2005; Feng et al., 2008; Feng and Kobayashi, 2009). Although there are differences in $O_3$ sensitivity among crops, there are a considerable number of crops sensitive to elevated $O_3$ concentrations and subject to yield loss. The effects of elevated $O_3$ concentrations on crop growth and yield are assessed generally by a cumulative exposure over a threshold concentration for a given length of time (Nussbaum et al., 1995; Fuhrer et al., 1997). The index AOT40 (accumulated exposure over a threshold ozone concentration of 40 ppb) (USEPA, 1996) was widely adopted in Europe to assess the effects of $O_3$. Mills et al. (2007) compiled AOT40-yield response functions for 19 crops in European regions from data available in 700 published papers and proceedings. Of these crops, wheat is an $O_3$-sensitive crop and rice is a moderately $O_3$-sensitive crop. By analyzing the

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quantified relationship between crop yield and AOT40, this study can provide valuable data to optimize the coefficient of ozone dose–response in China and improve regional modeling studies that adopted the AOT40 index (e.g., Tian et al., 2011; Ren et al., 2007, 2011).

Wheat and rice are two globally important crops that are sensitive to $O_3$ stress (Mills et al., 2007). Rice is consumed by more than half of the world's current population (IRRI, 2002). Wheat is grown on about 200 million hectares in a wide range of environments, with an annual production of more than 619 million metric tons (FAO, 2007). Because they are widely planted, the responses of wheat and rice to elevated $O_3$ concentrations have been assessed in many countries (Pleijel et al., 1991; Kobayashi et al., 1995; Rai et al., 2007). Recently, the physiological, growth and yield responses of wheat and rice to elevated $O_3$ have been reviewed by Feng et al. (2008), Feng and Kobayashi (2009) and Ainsworth (2008), respectively. Ainsworth (2008) has reported that the mean rice yield would be decreased by 14% when exposed to 62 ppb $O_3$ (mean concentration) compared with charcoal-filtered air after meta-analyzing 18 studies. Feng et al. (2008) concluded that an elevated $O_3$ concentration would decrease wheat grain yield by 26% at a mean $O_3$ concentration of 77 ppb based on a database containing 53 peer-reviewed studies. Even with lower $O_3$ exposure (average 43 ppb, mean concentration), there was also a significant decrease in the grain yield of wheat (18%) (Feng and Kobayashi, 2009).

To model crop yield response to environmental change, and to facilitate breeding crop cultivars for high yields, the yield components are of interest. Available investigations have shown that elevated $O_3$ concentrations decreased the mass of individual grains and the grain number of rice by 5% and 20%, respectively (Ainsworth, 2008), and individual grain weight and grain number per ear of wheat by 18% and 11%, respectively (Feng et al., 2008).

As the most populous country in the world, China's grain supply is critical to global food security. If crop yields are threatened by elevated $O_3$ concentrations, there would be a negative impact on future grain production both in China and worldwide. Available ambient $O_3$ monitoring data have implied that the $O_3$ concentrations were sufficiently high to reduce rice and wheat yield significantly in some developed regions of China (Wang et al., 2007), such as the Yangtze River Delta (Zhou, 2004; Wang et al., 2005) and the Pearl River Delta (Shao et al., 2006). Crop losses due to $O_3$ concentrations in China have been estimated to be as much as 23% (Chameides et al., 1999; Aunan et al., 2000; Wang and Mauzerall, 2004; Wang et al., 2005). However, these estimates are usually questionable because the dose–response relationships used in those estimations were derived from experiments conducted in USA and Europe where crop cultivars, soil and climate environments, and management practices are different from those in China.

Recently, a few studies have been carried out to investigate the crop responses to elevated $O_3$ concentrations in China. Feng et al. (2003) studied the effects of elevated $O_3$ concentration on pot-planted wheat and rice in North China. Shi et al. (2009) and Zhu et al. (2011) investigated the effects of $O_3$ on rice and winter wheat yields, respectively, for four cultivars under open-air field conditions. However, it is not clear whether the dose–response relationships based on single season investigations or only two treatments are reliable predictors of long-term crop losses under elevated $O_3$ concentrations. In this study, by analyzing the experimental data for 5 years, we assessed wheat and rice losses under elevated $O_3$ concentrations and their relationship to changes in yield components. This is the first multiple-year study to establish dose–response relationships for wheat and rice under field conditions in order to assess and predict crop loss in China more reliably.

## 2. Material and methods

### 2.1. Site description

The experimental site is located at the Shuangqiao Farm (31°53′N, 121°18′E) in Jiaxing City, Zhejiang Province, about 100 km southwest of Shanghai, China's largest city. This site is in the center of the Yangtze Delta and is one of the most important crop production areas in China. It is influenced by the Asian monsoon climate system, with cold dry winters and hot wet summers. Mean temperature and annual precipitation are 15.5 °C and 1200 mm, respectively. The soil developed from peatland 30–40 years ago is silt clay. Generally, rice and winter wheat were grown in fields in rotation, rice being grown from July to November, and wheat from November to May of the following year.

### 2.2. Open top chamber

The octagonal open-top chamber (OTC, 2.2 m in height and 2 m in diameter) had aluminum alloy frames covered with polyethylene plastic film, with an improved innovative $O_3$ distribution system (Zheng et al., 2007). The air or charcoal-filtered air was delivered by a centrifugal blower into a vertical plastic duct (diameter of 11 cm), where $O_3$ was added, then into a horizontally rotatable transparent pipe (diameter of 5 cm) above the crop canopy and released from many small holes (diameter of 10 mm at an interval of 10 cm) situated facing 45° downward on the pipe. The rotatable pipe was connected to the duct by a custom-designed shaft, propelled by the reaction force from released air and raised with crop growth. $O_3$ was supplied from an $O_3$ generator from pure oxygen by the high-voltage electric discharge method (Yuyao Shenglete Company, Zhejiang, China). The target $O_3$ concentration was obtained by a mass flow controller (custom-made by Shengye Company, Beijing, China) regulating oxygen entering the $O_3$ generator based on the established relationship between $O_3$ concentration within the OTC and oxygen volume. A series of solenoid valves, linked online with a programmable Log Controller (PLC K80S, LG, Korea) was employed to switch gas entering the $O_3$ analyzer (Model 9810B, Monitor Labs, Australia) from each OTC sequentially. The $O_3$ concentration within each chamber was monitored sequentially for 5 min. A data logger (CR1000, Campbell Scientific Inc., UT, USA) was connected to store $O_3$ concentration data and to monitor temperature within the OTCs using constantan-copper thermocouples. We checked the $O_3$ concentration data recorded by the data logger every day and regulated the oxygen volume entering the $O_3$ generator if the target $O_3$ concentrations within the OTC were not achieved. Monitoring has shown that $O_3$ concentrations were evenly distributed vertically and horizontally within the OTC. The difference in mean temperature between the inside and outside of the OTC was less than 1.87 °C (Zheng et al., 2007). Measurements with a Li-190 quantum sensor (Li-Cor Environmental, Lincoln, Nebraska, USA) showed that photosynthetic active radiation was reduced by less than 15% within the OTC compared with the outside.

### 2.3. Ozone treatments

There were 12 OTCs established in the field with four treatments of $O_3$ and three replicates randomly assigned to each treatment. All chambers were separated by a guard block (2 m × 3 m). Charcoal filtered air (CF) and non-filtered air (NF) were the control treatments. Two levels of elevated $O_3$ treatments were applied by adding $O_3$ into charcoal filtered air as the $O_3$-1 and $O_3$-2 treatments. The target $O_3$ concentrations were 70–100 ppb for the $O_3$-1 treatment and 150–200 ppb for the $O_3$-2 treatment, varying with year and crop (Table 1). Two modes of fumigation were applied: constant $O_3$ exposure and diurnal variation of $O_3$ exposure. In the former mode, constant $O_3$ concentrations were applied daily for 8 h (09:00–17:00) for the $O_3$-1 and $O_3$-2 treatments. In the latter mode, ambient diurnal variations in $O_3$ concentrations were simulated in the growing seasons for rice in 2006 and for winter wheat in 2006 and 2007. In the $O_3$-1 treatments: 50 ppb $O_3$ for 09:00–10:00, 100 ppb $O_3$ for 10:00–12:00, 150 ppb $O_3$ for 12:00–14:00, 100 ppb $O_3$ for 14:00–16:00, and 50 ppb $O_3$ for 16:00–17:00; in the $O_3$-2 treatments: 100 ppb $O_3$

**Table 1**
The crop management and $O_3$ exposure of winter wheat.

| | | 2004 | 2006 | 2007 | 2008 |
|---|---|---|---|---|---|
| Cultivar | | Yangmai185 | Jia002 | Jia002 | Jia002 |
| Crop management | Sowing | Oct. 15 | Nov. 7 | Nov. 15 | Nov. 18 |
| | Harvesting | May 23 | May 16 | May 28 | May 31 |
| Exposure | Mode | Constant | Diurnal variation | Diurnal variation | Constant |
| | Beginning | Mar. 15 | Mar. 13 | Mar. 26 | Mar. 20 |
| | End | May 18 | Apr. 28 | May 19 | May 13 |
| Target Concentrations (ppb) | $O_3$-1 treatment | 100 | 100 | 75 | 100 |
| | $O_3$-2 treatment | 200 | 150 | 150 | 150 |

**Table 2**
The crop management and $O_3$ exposure of rice.

| | | 2004 | 2005 | 2006 | 2007 | 2008 |
|---|---|---|---|---|---|---|
| Cultivars | | Jiahua 2 | Jiahua 2 | Fan 3694 | Fan 3694 | Fan 3694 |
| Crop management | Sowing | May 15 | May 27 | May 15 | May 25 | May 26 |
| | Transplanting | Jul. 2 | Jul. 1 | Jun. 20 | Jun. 29 | Jul. 4 |
| | Harvest | Oct.28 | Oct. 25 | Oct. 25 | Nov. 2 | Nov. 12 |
| Exposure | Mode | Constant | Constant | Diurnal variation | Constant | Constant |
| | Beginning | Jul. 21 | Jul. 25 | Jul. 17 | Jul. 29 | Jul. 25 |
| | End | Oct. 10 | Oct 14 | Oct. 10 | Oct. 11 | Oct. 19 |
| Target Concentration (ppb) | $O_3$-1 treatment | 100 | 75 | 100 | 100 | 100 |
| | $O_3$-2 treatment | 200 | 150 | 150 | 150 | 150 |

for 09:00–10:00, 150 ppb $O_3$ for 10:00–12:00, 200 ppb $O_3$ for 12:00–14:00, 150 ppb $O_3$ for 14:00–16:00, and 100 ppb $O_3$ for 16:00–17:00. Fumigations were performed every day except when it was raining. The timings of exposure are shown in Tables 1 and 2.

### 2.4. Crop management

Winter wheat (*Triticum aestivum* L.) and rice (*Oryza sativa* L.) seeds were provided by Jiaxing Academy of Agriculture using locally adapted cultivars (Tables 1 and 2). The cultivars of wheat and rice were changed in 2006 because local cultivars were also changed. All crop management practices, such as sowing, transplanting (rice), harvesting, and fertilizing, were adopted according to local agronomic customs so that water, fertilizer, pathogens, insects and weeds were not limiting factors. Compound fertilizer ($N/P_2O_5/K_2O$ = 15:15:15) was applied at a rate of 78 g/m$^2$ for each OTC before transplanting (rice) or sowing (winter wheat), and urea (N content, 46.5%) was applied at a rate of 21 g/m$^2$ for each OTC during the jointing stage and the booting stage (rice) or tillering stage (winter wheat).

### 2.5. Yield

Before harvesting each crop, 15 plants were sampled from each OTC for yield determination. The yield components (floret, panicle and grain for wheat, and panicle, first and second rachis, and grain for rice) were separated for counting or weighing. Yield loss due to elevated $O_3$ concentrations for each year was calculated as the relative yield, which is the percentage of yield for each treatment compared with the yield under CF treatment.

### 2.6. Statistical analysis

An analysis of variance (ANOVA) was used to test for mean differences between treatments ($N = 3$ chambers). The Fishers Protected LSD test was used for post-hoc examination of treatment differences. Statistical analysis was performed with the statistical package SPSS (SPSS Inc., Chicago, IL, USA). The relationships among relative yield, yield component and AOT40 value were analyzed for each crop over 4 or 5 growing seasons using Excel 2003.

## 3. Results

### 3.1. Winter wheat

Elevated $O_3$ significantly reduced winter wheat yield (Table 3). Yields decreased by 8.5–58% and 40–73% compared with CF control for $O_3$-1 and $O_3$-2 treatments, respectively, with the annual variations in $O_3$ concentrations and AOT40. Nevertheless, significant exponential and linear regressions were fitted between relative yield and AOT40 for 4 years (Fig. 1A) for all data (Equation (1)) and for all data except the highest AOT40 in the first year (Equation (2)), respectively.

$$\mathrm{RY} = 100\exp(-0.0255\ \mathrm{AOT40}) \quad \left(R^2 = 0.846\right) \tag{1}$$

$$\mathrm{RY} = 100 - 2.2795\ \mathrm{AOT40} \quad \left(R^2 = 0.859\right) \tag{2}$$

The wheat yield components, such as number of florets, number of grains per panicle, 1000-grain weight and harvest index were also impacted by elevated $O_3$ (Table 3). Except for a lack of statistical analysis in 2004 due to handling of the harvested plants from chambers under the same treatment together, the number of grains

**Table 3**
The yield components of winter wheat after exposure to different $O_3$ treatments.

| Years | Treatments | AOT40 (ppm h) | No. of florets per panicle | No. of grains per panicle | 1000-grain weight (g) | Harvest index (%) | Yield per plant (g) |
|---|---|---|---|---|---|---|---|
| 2004 | CF | 0 | 15.6 | 25.81 | 45.75 | 50.43 | 1.29 |
| | NF | 3.82 | 15.67 | 27.82 | 46.5 | 55.19 | 1.41 |
| | $O_3$-1 | 22.61 | 14.13 | 23.37 | 27.18 | 29.28 | 0.54 |
| | $O_3$-2 | 61.91 | 13.21 | 19.65 | 12.56 | 31.16 | 0.35 |
| 2006 ($N = 3$) | CF | 0 | 17.93 ± 0.74 | 46.90 ± 3.93a | 37.43 ± 1.37a | 47.01 ± 0.53a | 1.52 ± 0.07a |
| | NF | 2.5 | 17.71 ± 0.95 | 46.24 ± 4.25a | 32.01 ± 0.78ab | 47.07 ± 2.21a | 1.35 ± 0.17ab |
| | $O_3$-1 | 14.27 | 16.87 ± 0.81 | 33.50 ± 3.29b | 29.71 ± 1.20b | 43.61 ± 3.42ab | 1.10 ± 0.15b |
| | $O_3$-2 | 24.22 | 16.71 ± 0.34 | 24.13 ± 3.75c | 24.05 ± 0.75c | 35.58 ± 3.20b | 0.91 ± 0.05b |
| ANOVA | *F* | | 2.063 | 5.47 | 18.256 | 2.875 | 3.286 |
| | *P-value* | | 0.184 | 0.024 | 0.001 | 0.103 | 0.079 |
| 2007 ($N = 3$) | CF | 0 | 15.45 ± 1.26a | 39.41 ± 4.03a | 43.62 ± 1.90a | 58.72 ± 5.13a | 1.65 ± 0.37a |
| | NF | 0.21 | 15.85 ± 1.30a | 40.58 ± 7.50a | 42.16 ± 0.73ab | 60.33 ± 2.67a | 1.61 ± 0.40a |
| | $O_3$-1 | 1.58 | 14.95 ± 0.81ab | 36.59 ± 4.03b | 41.74 ± 0.49b | 56.41 ± 5.69a | 1.51 ± 0.33a |
| | $O_3$-2 | 9.17 | 14.08 ± 1.13b | 32.39 ± 5.22b | 25.63 ± 1.03c | 43.35 ± 7.96b | 0.99 ± 0.26b |
| ANOVA | *F* | | 5.311 | 3.743 | 138.506 | 33.188 | 13.542 |
| | *P-value* | | 0.003 | 0.011 | 0 | 0 | 0 |
| 2008 ($N = 3$) | CF | 0.1 | 16.37 ± 0.73a | 36.55 ± 1.33a | 43.05 ± 2.98a | 50.45 ± 2.49a | 1.57 ± 0.11a |
| | NF | 0.15 | 16.65 ± 0.16a | 35.50 ± 1.59a | 44.69 ± 1.67a | 48.85 ± 1.17a | 1.55 ± 0.04a |
| | $O_3$-1 | 15.32 | 13.87 ± 1.14b | 27.49 ± 4.55b | 31.25 ± 2.74b | 39.12 ± 5.53b | 0.86 ± 0.15b |
| | $O_3$-2 | 27.67 | 13.73 ± 0.31b | 21.44 ± 2.21c | 27.25 ± 2.34b | 25.30 ± 4.25c | 0.58 ± 0.02c |
| ANOVA | *F* | | 15.233 | 20.402 | 36.265 | 28.492 | 84.129 |
| | *P-value* | | 0.001 | <0.001 | <0.001 | <0.001 | <0.001 |

Note: Data are "means ± standard errors"; and values with different letters in the same column for each year designate significant difference among the $O_3$ treatments ($P < 0.05$).

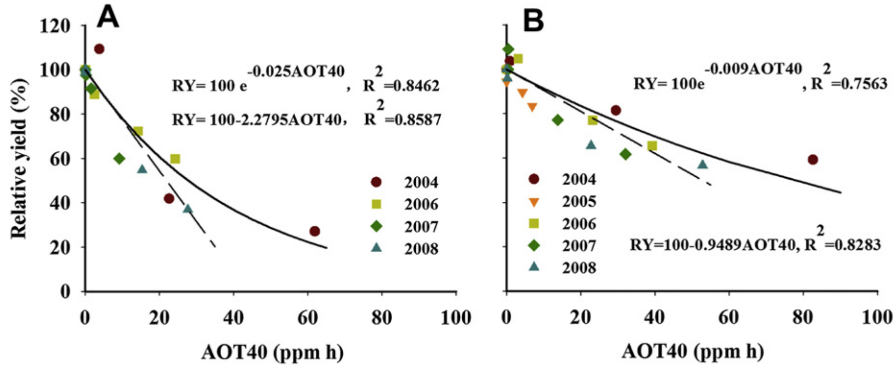


**Fig. 1.** The exponential (for all data) and linear regressions (for all apart from the highest AOT40 data in first year) of relative crop yield to CF control and AOT40 (accumulated daily mean $O_3$ concentration above 40 ppb during fumigation): (A) winter wheat, (B) rice.

per panicle, 1000-grain weight and harvest index significantly decreased with increasing $O_3$ exposure (Table 3). Significantly, linear relationships across cultivars and years between relative yield and relative yield components to CF treatment showed that 1000-grain weight ($P < 0.01$) and harvest index ($P < 0.01$) were the primary factors impacting yield loss under elevated $O_3$ (Fig. 2).

### 3.2. Rice

Rice yields were reduced significantly under elevated $O_3$ across cultivars, years and exposure protocols (Table 4). The mean yields decreased by 10–34% and 16–43% compared with CF control for $O_3$-1 and $O_3$-2 treatments, respectively, with the annual variations in $O_3$ concentrations and AOT40. Significant exponential and linear regressions were fitted between relative yield to CF treatment and AOT40 for 5-year periods for all data (Equation (3)) and for all data except the highest AOT40 in the first year (Equation (4)) (Fig. 1B), respectively.

$$RY = 100\exp(-0.0092\ \mathrm{AOT40}) \quad \left(R^2 = 0.756\right) \tag{3}$$

$$RY = 100 - 0.9489\ \mathrm{AOT40} \quad \left(R^2 = 0.828\right) \tag{4}$$

The rice yield components were reduced under elevated $O_3$ concentrations (in $O_3$-1 and $O_3$-2 treatments), which varied with $O_3$ exposure and seasons (Table 4). The $O_3$ effects were significant for harvest index in 5 years, 1000-grain weight and number of second rachis in 4 years, and number of first rachis and number of grain per panicle in 3 years. For 5-year periods across cultivars and exposure protocols, the significant linear relationships between relative yield and relative yield components to CF treatment showed that the

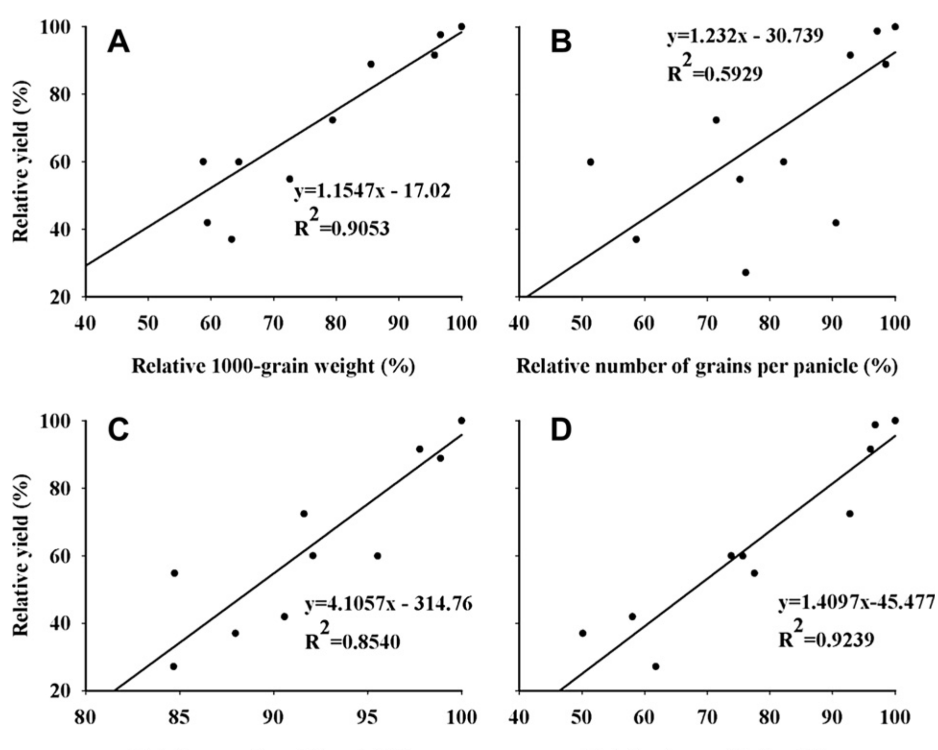


**Fig. 2.** The linear relationships between relative yield and relative yield components to CF control for winter wheat in 4 year $O_3$ experiments: (A) 1000-grain weight, (B) Number of grains per panicle, (C) Number of florets, and (D) Harvest index.

**Table 4**
The yield components of rice after exposure to different $O_3$ treatments.

| Years | Treatments | AOT40 (ppm h) | No. of grain per panicle | No. of first rachis per panicle | No. of second rachis per panicle | 1000-grain weight (g) | Harvest index (%) | Yield per plant (g) |
|---|---|---|---|---|---|---|---|---|
| 2004 ($N = 3$) | CF | 0 | 107.90 ± 8.12a | 11.50 ± 3.44a | 20.70 ± 3.65a | 27.70 ± 1.01a | 53.48 ± 2.04a | 2.72 ± 0.21a |
| | NF | 0.91 | 102.60 ± 8.41a | 10.00 ± 0.82a | 17.60 ± 4.59a | 27.12 ± 0.83a | 54.13 ± 1.34a | 2.78 ± 0.20a |
| | $O_3$-1 | 29.472 | 85.80 ± 9.33b | 9.10 ± 0.95b | 11.40 ± 3.19b | 25.74 ± 1.09b | 50.52 ± 2.11a | 2.21 ± 0.22b |
| | $O_3$-2 | 82.603 | 55.70 ± 6.49c | 8.10 ± 0.74c | 7.70 ± 2.85c | 24.39 ± 0.62c | 45.98 ± 2.01b | 1.62 ± 0.13c |
| ANOVA | *F* | | 41.355 | 5.572 | 25.678 | 1.782 | 2.545 | 5.195 |
| | *P-value* | | 0.000 | 0.003 | 0.000 | 0.228 | 0.129 | 0.028 |
| 2005 ($N = 3$) | CF | 0 | 153.00 ± 7.39 | 12.63 ± 0.41 | 25.13 ± 1.77a | 23.14 ± 0.26 | 52.66 ± 2.00a | 2.92 ± 0.14a |
| | NF | 0 | 145.40 ± 3.35 | 12.00 ± 0.53 | 23.17 ± 0.70ab | 22.71 ± 0.05 | 52.04 ± 1.00ab | 2.76 ± 0.13ab |
| | O3-1 | 4.3 | 139.10 ± 6.83 | 12.06 ± 0.11 | 22.46 ± 1.10ab | 22.83 ± 0.46 | 51.19 ± 0.01ab | 2.62 ± 0.04ab |
| | O3-2 | 6.93 | 132.50 ± 7.83 | 11.70 ± 0.36 | 20.63 ± 1.26b | 22.54 ± 0.25 | 48.99 ± 1.01b | 2.44 ± 0.10b |
| ANOVA | *F* | | 1.817 | 1.082 | 2.151 | 0.745 | 2.351 | 3.479 |
| | *P-value* | | 0.222 | 0.410 | 0.172 | 0.555 | 0.148 | 0.070 |
| 2006 ($N = 3$) | CF | 0 | 146.80 ± 7.56ab | 14.33 ± 0.48a | 22.88 ± 1.96ab | 25.61 ± 0.27a | 42.49 ± 0.02ab | 5.37 ± 0.60a |
| | NF | 3.14 | 155.20 ± 6.78a | 14.79 ± 0.26a | 25.33 ± 1.52a | 24.87 ± 0.46ab | 46.35 ± 0.01a | 5.63 ± 0.44a |
| | $O_3$-1 | 23.24 | 133.50 ± 5.96bc | 14.04 ± 0.34ab | 19.67 ± 1.61bc | 25.03 ± 0.33a | 42.38 ± 0.02ab | 4.14 ± 0.41ab |
| | $O_3$-2 | 39.28 | 118.20 ± 5.67c | 13.12 ± 0.70b | 16.65 ± 1.39c | 23.81 ± 0.28b | 37.08 ± 0.04b | 3.52 ± 0.59b |
| ANOVA | *F* | | 6.128 | 4.619 | 7.067 | 4.745 | 2.996 | 2.394 |
| | *P-value* | | 0.004 | 0.037 | 0.012 | 0.035 | 0.095 | 0.044 |
| 2007 ($N = 3$) | CF | 0.4 | 106.20 ± 15.30 | 11.44 ± 0.42 | 15.14 ± 2.03 | 24.61 ± 1.80a | 45.58 ± 1.63a | 2.41 ± 0.23a |
| | NF | 0.5 | 112.90 ± 11.00 | 11.65 ± 0.45 | 16.35 ± 2.67 | 23.81 ± 0.94a | 47.06 ± 2.03a | 2.63 ± 0.30a |
| | $O_3$-1 | 13.83 | 103.40 ± 8.87 | 10.69 ± 0.53 | 14.86 ± 2.04 | 17.91 ± 2.92b | 41.42 ± 1.78b | 1.86 ± 0.22ab |
| | $O_3$-2 | 32.08 | 99.70 ± 15.00 | 10.85 ± 0.80 | 13.75 ± 3.37 | 15.06 ± 1.08b | 39.49 ± 1.75b | 1.49 ± 0.28b |
| ANOVA | *F* | | 0.418 | 1.945 | 0.717 | 18.623 | 12.683 | 4.396 |
| | *P-value* | | 0.745 | 0.201 | 0.569 | 0.001 | 0.002 | 0.042 |
| 2008 ($N = 3$) | CF | 0.01 | 137.13 ± 14.66a | 13.40 ± 0.91a | 21.86 ± 2.21a | 23.43 ± 0.73a | 47.34 ± 2.21a | 3.17 ± 0.43a |
| | NF | 0.27 | 133.07 ± 3.21a | 13.36 ± 0.18a | 21.01 ± 0.43a | 23.77 ± 0.48a | 45.13 ± 2.69a | 3.04 ± 0.27a |
| | $O_3$-1 | 22.77 | 106.79 ± 11.40a | 12.56 ± 0.77ab | 15.63 ± 2.52b | 20.53 ± 0.14b | 40.90 ± 0.75b | 2.08 ± 0.33b |
| | $O_3$-2 | 52.84 | 104.11 ± 8.89b | 12.01 ± 0.69b | 14.14 ± 2.70b | 17.35 ± 1.36c | 40.65 ± 1.16b | 1.80 ± 0.04b |
| ANOVA | *F* | | 8.211 | 2.823 | 9.492 | 41.017 | 9.213 | 14.981 |
| | *P-value* | | 0.008 | 0.007 | 0.005 | 0.000 | 0.006 | 0.001 |

Note: Data are "means ± standard errors"; and values with different letters in the same column for each year designate significant difference among the $O_3$ treatments ($P < 0.05$).

number of grain per panicle ($P < 0.01$) and harvest index ($P < 0.01$) are the primary factors impacting yield loss under elevated $O_3$ concentrations (Fig. 3). Other components such as number of first rachis ($P < 0.05$), number of second rachis ($P < 0.05$) and 1000-grain weight ($P < 0.05$) were also affected by $O_3$, possibly influencing yield loss.

## 4. Discussion

### *4.1. Yield losses of rice and wheat due to elevated $O_3$ concentration*

The yield loss for winter wheat was higher than for rice under both $O_3$-1 and $O_3$-2 treatments (Tables 3 and 4). Mills et al. (2007) had classified wheat as a sensitive crop and rice as a moderately sensitive crop by general linear model analysis between yield loss and AOT40. Meta-analyses have shown that wheat yield loss was 26% at mean 7-h or 8-h $O_3$ concentrations of 72 ppb with a range of 30–200 ppb across all of the reported studies on effects of chronic $O_3$ treatment (Feng et al., 2008) and rice yield loss was 14% at a mean $O_3$ concentration of 62 ppb when compared with rice grown in charcoal filtered air (Ainsworth, 2008).

Although the climate, cultivars and exposure periods varied each year, the combined winter wheat dataset could be represented by a single regression equation of relative yield plotted against AOT40 (Fig. 1A). The same was true for the rice yield data (Fig. 1B). This may be because the soil conditions, water and nutrient supply were sufficient for winter wheat and rice growing in our study under all conditions. Although crop cultivars were switched in 2006 due to local practice, the two cultivars for wheat and rice were bred from the same strains and may have the same degree of sensitivity to $O_3$. In this study, we have developed an $O_3$ exposure system that simulates the diurnal variation of ambient $O_3$, and it was used in 2006 for wheat and rice and 2007 for wheat. Unfortunately, the system was not stable and was difficult to maintain. So the constant $O_3$ exposures were resumed in later experiments. It is difficult to assess the differences in the response of wheat and rice to AOT40 between the constant and diurnal pattern of $O_3$ exposures because of yearly variations in exposure duration and dose in this study. Emberson et al. (2009) have reported that there were no obvious differences between the sensitivities of Asian winter and spring wheat varieties planted in different countries. Analysis of variance and generalized linear modeling (GLIM) showed that there were no statistically significant differences between the slope of the regressions for the data from the USA and EU for each of the crops studied (Mills et al., 2007). When compared with the slope of Mills' functions, our results showed that wheat and rice were more sensitive to $O_3$ in the Yangtze Delta of China when AOT40 was larger than 1.5 ppm h for wheat and 10.7 ppm h for rice, respectively (Fig. 4). The greater sensitivity of wheat and rice to $O_3$ in the Yangtze Delta where wheat and rice grow without water and nutrient stresses than that predicted from North American dose–response relationships has been reported by Emberson et al. (2009). Compared with our study, the wheat and rice used in North China by Feng et al. (2003) were less sensitive to $O_3$ (Fig. 4). In addition to growing method (pot vs. field) and climate (semi-humid vs. humid), the difference in germplasm resource may be a factor in explaining the differences in $O_3$ sensitivities of cultivars from the Yangtze Delta versus North China. The contribution of genetic variation will require further investigation.

### *4.2. Responses of yield components to elevated $O_3$ concentrations*

Crop yield is affected by a series of components depending on crop architecture and inflorescence structure. For winter wheat, the number of florets and number of grains per plant were significantly reduced by elevated $O_3$ concentrations (Table 3). Compared with

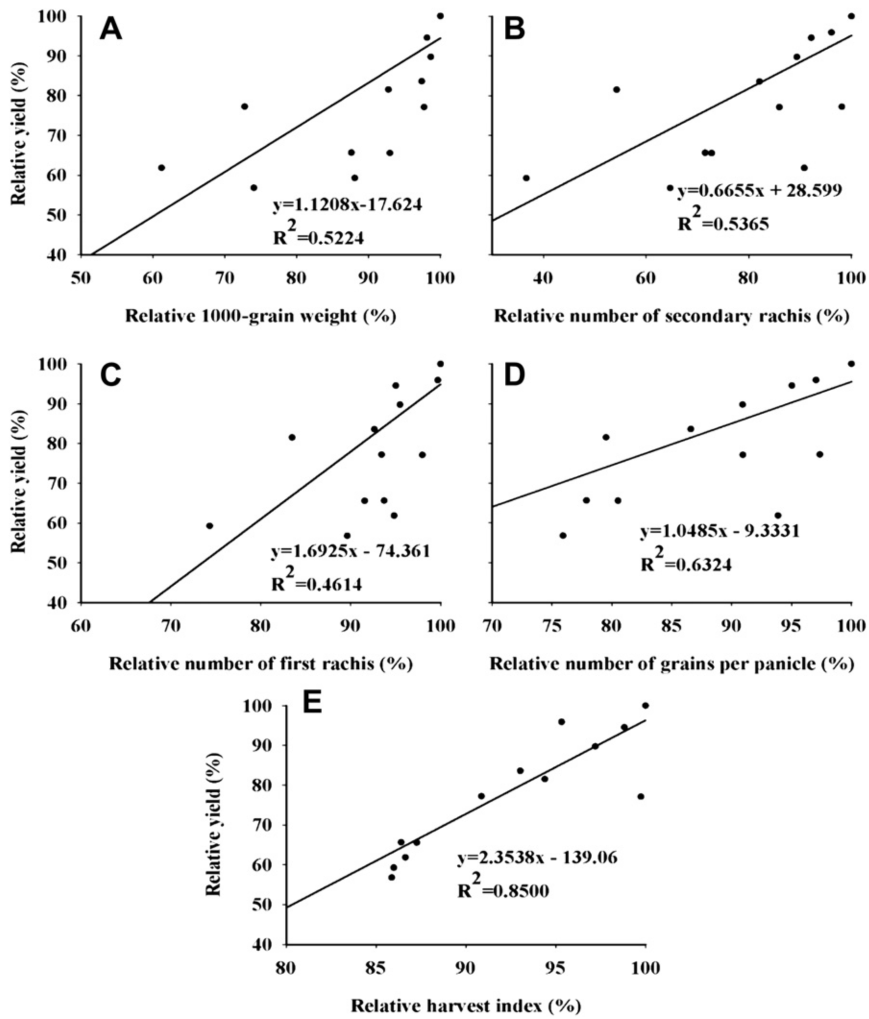


**Fig. 3.** The linear relationships between rice relative yield and relative yield components to CF control for rice in 5 year $O_3$ experiments. (A) 1000-grain weight, (B) Number of secondary rachis, (C) Number of first rachis, (D) Number of grains per panicle, and (E) Harvest index.

the CF control, the number of florets and number of grains per panicle were reduced for all 4 years, by 8.5% and 17.5% for $O_3$-1 treatment, and by 11.8% and 32.9% for $O_3$-2 treatment, respectively. In the review by Feng and Kobayashi (2009), the number of florets was reduced by 6% at a mean $O_3$ concentration of 65 ppb. For rice, the numbers of grains, first rachis and second rachis per panicle were reduced for all 5 years, by 12.7%, 8.0%, and 20.0% for $O_3$-1 treatment, and by 22.3%, 12.2%, and 30.5% for $O_3$-2 treatment, respectively (Table 4). In the review by Ainsworth (2008), for rice, the number of grains was reduced by 20% at a mean $O_3$ concentration of 62 ppb. Shi et al. (2009) reported that the number of florets per panicle was significantly reduced, but individual grain mass was only slightly reduced for sensitive rice cultivars under elevated $O_3$ exposure in fully open-air field conditions.

Grain weight per plant is one of the yield components most critical to crop yield. The elevated $O_3$ concentration could significantly reduce grain weight because of decreased photosynthesis and increased oxidative stress. This study showed that the 1000-grain weights decreased by 18.6% and 37.2% for winter wheat (Table 3), and by 10.1% and 17.3% for rice (Table 4), in $O_3$-1 and $O_3$-2

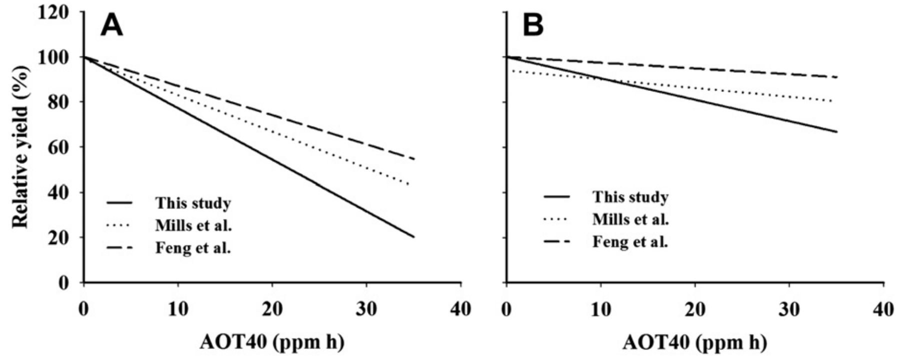


**Fig. 4.** Comparison of exposure–response relationships among this study, Feng et al. (2003) and Mills et al. (2007) for (A) winter wheat, and (B) rice.

treatments, respectively. In recent reviews, grain weights losses of 18% for wheat (Feng and Kobayashi, 2009) and 5% for rice (Ainsworth, 2008) were reported.

Many yield components were reduced along with total yield under elevated $O_3$ conditions (Tables 3 and 4). Based on the relationships between relative yield and relative component value (Figs. 2 and 3), the results indicated that, for wheat, 1000-grain weight and harvest index were more sensitive to $O_3$ concentration than other components, and, for rice, the number of grains per panicle and harvest index were also seriously impacted by elevated $O_3$ concentrations.

### *4.3. Implications for estimating $O_3$-induced crop loss*

The 5-year studies strongly support the hypothesis that winter wheat and rice yields are decreasing with increasing $O_3$ concentrations in China. The AOT40-based exposure–response equations derived from these experiments could be used to estimate current and future $O_3$-induced grain losses confidentially. Higher yield losses relative to AOT40 were found in our study compared with those in the study by Feng et al. (2003) and in the synthesis by Mills et al. (2007). This indicates that yield loss due to increased $O_3$ concentration might be higher than previous estimates.

The exposure–response equation, as an essential tool for estimating crop loss due to increasing $O_3$ concentration, has generally been obtained from open-top chamber (OTC) experiments because OTCs can be built economically, operated easily, and have stable $O_3$ concentrations. Nevertheless, there are some deficiencies when estimating crop loss based on OTC-derived exposure–response functions.

The primary errors come from altered growing conditions within the OTC relative to ambient conditions. The greenhouse effects of OTC have been criticized for many years. The OTC systems we used in this study could distribute $O_3$ evenly and stably, and exchange air between the inside and outside two to three times per minute (Zheng et al., 2007), so the environmental differences between the OTC and actual growing condition have been partially mitigated. The air temperature inside the OTC was 2 °C higher than outside when ambient temperature was above 30 °C in summer (Zheng et al., 2007). The light inside the OTC was also reduced by about 10% and air humidity was altered sometimes. Because our results were expressed as the relative yield and their component values under $O_3$ treatment compared with those under CF treatment using the same-designed OTCs, the OTC effects were avoided to some degree. $O_3$-FACE (Ozone-free air control enrichment) experiments have been carried out in USA, Europe and China (Shi et al., 2009), but unfortunately, it is difficult to derive dose–response functions from these data because only one treatment and one control are deployed in most FACE experiments.

In the first year experiment, the target $O_3$ concentration was set as high as 200 ppb above the ambient $O_3$ concentration. Excluding the first year's results, significant linear relationships were obtained between wheat and rice yields and AOT40, and would be better than the logarithmic relationships derived from all ranges of $O_3$ exposure. The exposure–response functions based on a large span of $O_3$ concentrations probably introduced errors in estimation of crop loss.

Because of rainy days, unstable ambient $O_3$ concentrations and the high activity of $O_3$ molecules, $O_3$ concentrations within the OTCs were achieved with small bias to the set concentrations. From Table 5, it is seen that the mean $O_3$ concentration under CF treatment ranged from 7.0 ppb to 20.3 ppb after ambient air had been charcoal-filtered, less than that under NF treatment (in the range 17.4 ppb–27.7 ppb). As compared with the target concentrations in Tables 1 and 2, mean $O_3$ concentrations achieved in $O_3$-1 and $O_3$-2 treatments (Table 5) are less than the target concentrations. Because of frequent rainy days, the mean $O_3$ concentrations achieved were as much as 36% less than the target concentration in 2007. So, exposure–response functions based on accumulated $O_3$ concentrations (e.g., AOT40) would be more meaningful.

**Table 5**
Mean (±SD) $O_3$ concentration (ppb) within the OTCs during the $O_3$ fumigation in 2007 and 2008.

| Year | Crop | CF | NF | $O_3$-1 | $O_3$-2 |
|---|---|---|---|---|---|
| 2007 | Wheat | 11.6 ± 5.1 | 27.7 ± 6.4 | 51.5 ± 3.5 | 98.4 ± 3.5 |
| 2008 | Wheat | 20.3 ± 9.5 | 27.7 ± 2.8 | 96.5 ± 4.1 | 142.7 ± 8.3 |
| 2007 | Rice | 19.7 ± 4.6 | 22.6 ± 1.3 | 69.6 ± 4.9 | 118.6 ± 5.3 |
| 2008 | Rice | 7.0 ± 3.7 | 17.4 ± 4.5 | 82.2 ± 4.3 | 138.3 ± 6.6 |

Our experiments were conducted without stresses such as nutrient or water deficiencies, diseases and pests, etc. It is not clear what the changes in $O_3$-induced crop yield loss under natural stresses would be. So studies of the effects of $O_3$ on crops in combination with multiple climatic, soil and water factors are necessary in the future.

## 5. Conclusions

Yields of winter wheat and rice, the most important grain crops in China and the world, are being threatened by rising ambient $O_3$ concentrations caused by rapid economic development and urbanization. Our experiments over 5 years have confirmed that winter wheat and rice were sensitive to elevated $O_3$ levels in China. The AOT40-based dose–response relationships showed that wheat and rice yield losses in our study were more serious than in previous reports both domestically and from abroad, which means that there would be more crop yield loss under $O_3$ stress in China and $O_3$ pollution would cause more damage to Chinese food security than previously assumed.

Our study also confirmed that winter wheat was more sensitive to $O_3$ than rice. In addition, the response of yield components to $O_3$ concentration would change with crop type. Grain mass is the most $O_3$-sensitive component for winter wheat but grain number is the most sensitive component for rice. This would make more sense for crop selection, cultural practices and breeding to reduce $O_3$-caused crop loss.

Because of the wide variability in crops, cultural practices and climate overall of China, it is not sufficient to estimate national $O_3$ effects on crop yield based on the experimental data obtained from one or two sites. In our experimental site, water and fertilizer were optimal for crop growth, and winter wheat and rice were likely more sensitive to $O_3$. In order to scale up the experimental results to the national scale, the response of winter wheat and rice to $O_3$ under stresses such as poor nutrient availability, drought, or temperature stress should be investigated. In addition, other crops such as maize, soybean, etc. should be studied to evaluate the effects of $O_3$ on the overall Chinese cropping system.

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## References

Ainsworth, E.A., 2008. Rice production in a changing climate: a meta-analysis of responses to elevated carbon dioxide and elevated ozone concentration. Global Change Biology 14, 1642–1650.

Andersen, C.P., 2003. Source sink balance and carbon allocation belowground in plants exposed to ozone. New Phytologist 157, 213–228.

Ashmore, M.R., 2005. Assessing the future global impacts of ozone on vegetation. Plant, Cell and Environment 28, 949–964.

Aunan, K., Berntsen, T., Seip, H., 2000. Surface ozone in China and its possible impact on agricultural crop yields. AMBIO: A Journal of the Human Environment 29, 294–301.

Chameides, W.L., Li, X.S., Tang, X.Y., Zhou, X.J., Luo, C., Kiang, C.S., John, J.St., Saylor, R.D., Liu, S.C., Lam, K.S., Wang, T., Giorgi, F., 1999. Is ozone pollution affecting crop yields in China? Geophysical Research Letters 26, 867–870.

Deliger, Zhao, Y.C., 2007. The variation of atmospheric background compounds in recent 10 years at Mt. Waliguan, Qinghai Province, China. Environmental Chemistry 26, 241–244 (In Chinese).

Dentener, F., Stevenson, D., Ellingsen, K., van Noije, T., Schultz, M., Amann, M., Atherton, C., Bergmann, D., Bey, I., Bouwman, L., Butler, T., Cofala, J., Collins, B., Drevet, J., Doherty, R., Eickhout, B., Eskes, H., Fiore, A., Gauss, M., Hauglustaine, D., Horowitz, L., Isaksen, I.S.A., Josse, B., Lawence, M., Krol, M., Lamargue, J.F., Montanaro, V., Miler, J.F., Peuch, V.H., Pitari, G., Pyle, J., Rast, S., Rodriquez, J., Sanderson, M., Savage, N.H., Shindell, D., Strahan, S., Szopa, S., Sudo, K., Van Dingenen, R., Wild, O., Zeng, G., 2006. The global atmospheric environment for the next generation. Environmental Science and Technology 40, 3586–3594.

Emberson, L.D., Burker, P., Ashmore, M.R., Mills, G., Jackson, L.S., Agrawal, M., Atikuzzaman, M.D., Cinderby, S., Engardt, M., Jamir, C., Kobayashi, K., Oanh, N.T.K., Quadir, Q.F., Wahid, A., 2009. A comparison of North American and Asian exposure-response data for ozone effects on crop yields. Atmospheric Environment 43, 1945–1953.

FAO, 2007. Statistical Database. No. 4, July 2007. http://www.fao.org.

Feng, Z.Z., Kobayashi, K., 2009. Assessing the impacts of current and future concentrations of surface ozone on crop yield with meta-analysis. Atmospheric Environment 43, 1510–1519.

Feng, Z.W., Jin, M.H., Zhang, F.H., 2003. Effects of ground-level ozone ($O_3$) pollution on the yields of rice and winter wheat in the Yangtze River Delta. Journal of Environmental Sciences (China) 15, 360–362.

Feng, Z.Z., Kobayashi, K., Ainsworth, E.A., 2008. Impact of elevated ozone concentration on growth, physiology and yield of wheat (*Triticum aestivum* L.): a meta-analysis. Global Change Biology 14, 2696–2708.

Fiscus, E.L., Booker, E.L., Burkey, K.O., 2005. Crop responses to ozone: uptake, modes of action, carbon assimilation and partitioning. Plant, Cell and Environment 28, 997–1011.

Fuhrer, J., Skarby, L., Ashmore, M.R., 1997. Critical levels for ozone effects on vegetation in Europe. Environmental Pollution 97, 91–106.

IPCC (The Intergovernmental Panel on Climate Change), 2007. Climate Change 2007: the Physical Science Basis. http://www.ipcc.ch/ipccreports/ar4-wg1.htm.

IRRI (International Rice Research Institute), 2002. Riceweb. http://www.riceweb.org.

Kobayashi, K., Okada, M., Nouchi, I., 1995. Effect of ozone on dry matter partitioning and yield of Japanese cultivars of rice (*Oryza sativa* L.). Agriculture, Ecosystems and Environment 53 (2), 109–122.

Mills, G., Buse, A., Gimeno, B., Bermejo, V., Holland, M., Emberson, L.D., Pleijel, H., 2007. A synthesis of AOT40-based response functions and critical levels of ozone for agricultural and horticultural crops. Atmospheric Environment 41, 2630–2643.

Morgan, P.B., Ainsworth, E.A., Long, S.P., 2003. How does elevated ozone impact soybean? A meta-analysis of photosynthesis, growth and yield. Plant, Cell and Environment 26, 1317–1328.

Naja, M., Akimoto, H., 2004. Contribution of regional pollution and long range transport to the Asia-Pacific region: analysis of long-term ozonesonde data over Japan. Journal of Geophysical Research 109, D21306.

Nussbaum, S., Geissmann, M., Fuhrer, J., 1995. Ozone exposure-response relationships for mixtures of perennial ryegrass and white clover depend on ozone exposure patterns. Atmospheric Environment 29, 989–995.

Pleijel, H., Skärby, L., Wallin, G., Selldén, G., 1991. Yield and grain quality of spring wheat (*Triticum aestivum L., cv. Drabant*) exposed to different concentrations of ozone in open-top chambers. Environment Pollution 69, 151–168.

Rai, R., Agrawal, M., Agrawal, S.B., 2007. Assessment of yield losses in tropical wheat using open top chambers. Atmospheric Environment 41, 9543–9554.

Ren, W., Tian, H.Q., Liu, M., Zhang, C., Chen, G., Pan, S., Felzer, B., Xu, X., 2007. Effects of tropospheric ozone pollution on net primary productivity and carbon storage in terrestrial ecosystems of China. Journal of Geophysical Research – Atmosphere 112, D22S09.

Ren, W., Tian, H.Q., Xu, X.F., Liu, M.L., Lu, C.Q., Chen, G.S., Melillo, J., Reily, J., 2011. Spatial and temporal patterns of $CO_2$ and $CH_4$ fluxes in China's croplands in response to multifactor environmental changes. Tellus B 63B, 222–240.

Shao, M., Tang, X.Y., Zhang, Y.H., Li, W.J., 2006. City clusters in China: air and surface water pollution. Frontiers in Ecology and the Environment 4, 353–361.

Shi, G., Yang, L., Wang, Y., Kabayashi, K., Zhu, J., Tang, H., Pan, S., Chen, T., Liu, G., Wang, Y., 2009. Impact of elevated ozone concentration on yield of four Chinese rice cultivars under fully open-air field conditions. Agriculture, Ecosystems and Environment 131, 178–184.

Tian, H., Melillo, J., Lu, C., Kicklighter, D., Liu, M., Ren, W., Xu, X., Chen, G., Zhang, C., Pan, S., Liu, J., Running, S., 2011. China's terrestrial carbon balance: contributions from multiple global change factors. Global Biogeochemical Cycles 25, GB1007.

USEPA (U.S. Environmental Protection Agency), 1996. Air Quality Criteria for Ozone and Related Photochemical Oxidants. US EPA. Report No. EPA/600/P-93/004bF. (Online).

Wang, X.P., Mauzerall, D.L., 2004. Characterizing distributions of surface ozone and its impact on grain production in China, Japan, and South Korea: 1990 and 2020. Atmospheric Environment 38, 4383–4402.

Wang, H.X., Kiang, C.S., Tang, X.Y., Zhou, X.J., Chameides, W.L., 2005. Surface ozone: a likely threat to crops in Yangtze delta of China. Atmospheric Environment 39, 3843–3850.

Wang, X.K., Manning, W.J., Feng, Z.W., Zhu, Y.G., 2007. Ground-level ozone in China: distribution and effects on crop yields. Environmental Pollution 147, 394–400.

Zheng, Q.W., Wang, X.K., Feng, Z.Z., Song, W.Z., Feng, Z.W., Ouyang, Z.Y., 2007. Effects of elevated ozone on biomass and yield of rice planted in open-top chamber with revolving ozone distribution. Environmental Science 28, 170–175 (In Chinese).

Zhou, X.J., 2004. The Interaction between the Atmosphere and Ecosystems in Yangtze Delta Region. Meteorological Press, Beijing (In Chinese).

Zhu, X., Feng, Z., Sun, T., Liu, X., Tang, H., Zhu, J., Guo, W., Kabayashi, K., 2011. Effects of elevated ozone concentration on yield of four Chinese cultivars of winter wheat under fully open-air field conditions. Global Change Biology 17, 2697–2706.