DOES CO$_2$ AMELIORATE PHYTOTOXIC EFFECTS OF O$_3$? A STUDY CASE ON THEIR INTERACTIVE EFFECTS ON PHYSIOLOGY, AND YIELD OF POTATO (*SOLANUM TUBEROSUM* L. CV. KARA) PLANTS

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Abstract: Potato (*Solanum tuberosum* L. cv. Kara) was grown in Open-Top Chambers (OTCs) in Northern Egypt at ambient (ca 350 ppm) or doubled CO$_2$ (ca 690 ppm) either in charcoal-filtered air (15 nl l^{-1}) or in non-filtered ambient air (78 nll^{-1} O$_3$) to investigate the changes in physiology and yield under long-term elevated CO$_2$ and/or O$_3$ throughout 100 days. Ambient O$_3$ level reduced net photosynthetic rates, number and weight of tubers by 18%, 41% and 21%, respectively, whereas elevated CO$_2$ caused the opposite effect where it increased the same parameters by 44%, 37% and 20%, respectively. Significant O$_3$ x CO$_2$ interactions were detected. However, O$_3$ caused an increase in GR and POD by 18% and 35%, respectively, while CO$_2$ caused an increase in POD only by 46%, and there was no effect of O$_3$ and/or CO$_2$ on other enzymes. The results of this study are discussed in relation to predicted atmospheric changes.

Keywords: Open-Top Chambers; OTCs; Potato (*Solanum tuberosum*); O$_3$; CO$_2$; photosynthesis; Stomatal Conductance; antioxidant enzymes; growth and yield. Superoxide Dismutase; SOD; Glutathione Reductase; GR; Ascorbate Peroxidase; APX; Guaiacol peroxidase; POD.

INTRODUCTION

Ozone (O$_3$) is the most phytotoxic pollutant in the USA, Europe and other industrialised areas of the world imposing the great threat on Vegetation (Gardner et al., 2005; Hassan, 2006; Hayes et al., 2009; Keutgen, 2005; Oksanen et al., 2005; Ranford and Reiling, 2007). Its ambient concentrations in the troposphere are increasing due to global industrialisation and excessive anthropogenic emission of its precursors. Moreover, CO$_2$ concentrations

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are expected to continue rising in the 21st century to levels that significantly increase plant growth and yield (Booker and Fiscus, 2005; Morison & Morecrof, 2008; Prather et al., 2001; Rämo et al., 2005).

Atmospheric CO_2 enrichment generally increases net photosynthesis, biomass and yield in many plant species, including potato plants (Ainsworth and Long, 2005; Donnelly et al., 2001a, b; Heagle et al., 2003; Schapendok et al., 2000). In contrast, exposure to O_3 induces leaf loss and decreased leaf production resulting in reduction in net photosynthetic rates and ultimately, reductions in net carbon gain and yield of crops (Calvo et al., 2009; Rämo et al., 2005; Ranford and Reiling, 2007; Topa et al., 2001; Yonekura et al., 2004).

Response of plants to O_3 and CO_2 singly and in combination vary among species and results are contradictory, while Booker and Fiscus (2005) reported that elevated CO_2 caused an increase in the threshold of O_3 flux for yields of soybean, Heagle et al. (2003) found difficulty in predicting effects of both gases on potato. However, Finnan et al. (2002) found that CO_2 ameliorated foliar injury caused by O_3 in potato. These variations in response may be due to differences in methodologies, duration of exposure and species and cultivar used.

Booker et al. (2005), found that exposure of soybean plants to elevated CO_2 prevent the deleterious effects of O_3 and they ascribed such protection to the fact that high CO_2 concentrations would lower O_3 flux into leaves. Similar results were found with potato, as elevated CO_2 caused an increase in tuber yield and ameliorated foliar injury caused by O_3 (Donnelly et al., 2001; Finnan et al., 2002).

McKee et al. (2000) stated that elevated CO_2 causes stomatal closure, lowering stomatal conductance and hence amount of O_3 flux into leaves and subsequently O_3 injury. Recently, this suggestion was supported by the results of Cardoso-Vilhena et al. (2004), who found similar results on wheat. However, this was not the case in the study of Pole and Pell (1999), who did not detect any reduction in stomatal conductance in some tree species in response to high levels of CO_2, although O_3 injury was diminished. They ascribed the protective effects of CO_2 against O_3 injury to increased photosynthate availability that could be used for repair and detoxification processes. Nevertheless, Olszyk and Wise (1997) revealed that elevated CO_2 did not suppress the deleterious effect of O_3 on biomass of tomato plants and they stated that this effect was due to partial stomatal closure. This finding was supported recently with the results of Cardoso-Vilhena et al. (2004).

Plants grown in elevated CO_2 might also be able to increase or maintain pools of antioxidants that confer resistance to O_3 injury on soybean plants (Booker and Fiscus, 2005; Polle and Pell, 1999; Reid and Fiscus, 1998).

However, Pritchard et al. (2000) found lower antioxidative enzyme activity in CO_2 – treated soybeans, they stated that this may reflect a decrease in oxidative stress resulting from growth in a CO_2 – enriched atmosphere. These results supported the earlier study of McKee et al. (1997) and Sgherri et al. (2000), who found no significant effect of elevated CO_2 on superoxide dismutase (SOD) and Glutathione Reductase (GR) in wheat and alfalfa plants, respectively.

Results from studies investigating the interactive effects of O_3 and CO_2 are contradictory. While Elevated CO_2 alleviated some of the negative effects of O_3 on physiology

and growth of some plants (Booker and Fiscus (2005) and this was correlated to increase in the activity of antioxidant enzymes (e.g. SOD and GR), Heagle et al. (2003) found difficulties in predicting interactive effects of O_3 and CO_2 based on the effects of the individual gases in potato plants. Potato (*Solanum tuberosum* L.) is of major importance for human nutrition and, in terms of the tonnage produced, is the second most important crop in Europe after wheat. Potato ranks fourth on a global scale after wheat, rice and maize (FAO 2000, cited by Vandermeiren et al., 2005). The impact of future increases in atmospheric concentrations of gaseous pollutants such as O_3 on the yield and quality of potato tubers is therefore of major concern for scientists, policy-makers and farmers. The recent EU-funded collaborative research programme 'Changing Climate and Potential Impacts on Potato Yield and Quality' (CHIP) provided an extensive database of information on this topic. CHIP is one of the largest Open-Top Chamber (OTC) projects ever commissioned to examine the impact of rising tropospheric O_3 concentrations, alone or in combination with elevated CO_2, on the physiology, growth and productivity of crop species or natural vegetation, matched only in Europe by the earlier EU funded ESPACE-wheat and ESPACE-grass programs. Evaluations of the impact of future increases in tropospheric O_3 levels and accumulated exposure must take account of their interactions with other changing physical, chemical and environmental stresses which are directly affected by either human activities (e.g. fertilizer application) and/or by global change (rising CO_2 concentrations, water stress, UV-B radiation, other gaseous pollutants). The potato growth simulation model, LPOTCO, which has been improved using the comprehensive dataset by CHIP, suggests that the prevailing conditions under climate

scenarios for 2050 (including increasing in temperature, O_3 and CO_2 concentrations) would increase the yield of potato in most of European countries, primarily because of the beneficial effect of increased CO_2 (Wolf and Van Oijen, 2003). These predictions suggest that potato productivity partially protected from the potentially damaging impact of O_3 by increasing CO_2 – induced decreases in stomatal conductance (Fiscus et al., 1997; McKee et al., 1997; Vandermerien et al., 2005).

Although many studies addressed the effects of CO_2 and /or O_3 on plants in the developed world, no such study, to the best of knowledge, was conducted in developing world.

The present investigation was conducted to investigate the response to O_3 and CO_2, singly an in combination, of an important Egyptian crop, potato (*Solanum tuberosum* L. cv Kara) in terms of foliar injury, stomatal conductance, photosynthetic rates, activity of some antioxidant enzymes, growth and yield and to assess and to understand the mechanism by which CO_2 can protect plants against phytotoxic effects of O_3.

MATERIALS AND METHODS

Experimental area, cultural methods and exposure

The fieldwork was carried out in northern Egypt, at Abbis village (a 'rural' site), located 35 km to the south of Alexandria city in the Nile Delta. The experimental site was about 2 km from the nearest traffic road and was surrounded by Eucalyptus, Casuarina and Salix trees. This was being the main agricultural area of the country.

Whole tubers of an Egyptian cultivar of potato (*S. tuberosum* L. cv. Kara), obtained

from a commercial source (averaged 50 g), were sown in 20-L pots, 18 November 2007, filled with soil collected from the field (soil texture was Loamy clay) in accordance with Standardized Changing Climate and Potential Impacts on Potato Yield and Quality (CHIP) protocol (Donnelly et al., 2001). The soil already enriched with fertilizer (NPK as farmers used to mix soil with NPK at a rate of 200, 100 and 150 kg h^{-1}, respectively, to get a reasonable yield).

Potato plant is a winter crop, so they were sown and harvested during the winter season as normal agricultural practical in Egypt.

Pots transferred to eight OTCs (Hassan, 1998), and distributed randomly in a complete random factorial block design for reciprocal gas treatment. There were 12 pots per chamber. Plants were watered twice a week with tap water till field capacity. No pesticides or insecticides were applied.

The experiment was a two-factorial design that consisted of two O_3 treatments (ambient and charcoal-filtered) and two atmospheric CO_2 concentrations (ambient and double ambient) replicated twice.

The treatment combinations were:

1 charcoal-filtered air (CF) and ambient CO_2 (ca 350 ppm) (control)

2 CF plus double CO_2 concentrations (ca 650 ppm)

3 non-filtered air (NF) plus ambient CO_2

4 NF plus double CO_2.

CO_2 was dispensed from a 500-L receiver and its concentrations in the chambers were monitored with Infrared CO_2 analyser (Model 6252; Li-Cor, Inc., Lincoln, NE) calibrated with CO_2 standards. O_3 concentrations in the chambers were monitored using UV photometric O_3 analyser (TECO Model 49;

Thermo Environmental, Franklin, MA). Analyser was calibrated with TECO Model 49 PS calibrator. Air was sampled in each chamber at canopy level every 7.5 min.

Stomatal conductance and Net Photosynthetic measurements

Photosynthetic rates (A), stomatal conductance (g_s) and internal CO_2 (C_i) were measured on the first foliage expanded leaf, from all plants, during the entire course of the experiment using a portable Infra Red Gas Analyser (Li-Cor, Model 6200) under OTC conditions.

Measurement were made weekly between 10:00 and 14:00 h (Egyptian local time), from the 7th to the 15th week after planting (destructive harvest).

Destructive harvest and antioxidant assays

Destructive harvests were carried out five times; 20, 40, 60, 80 and 100 Days After Planting (DAP). Plants were randomly selected and divided into above- and below-ground biomass before determining the number and dry weight of green and senescent leaves, stem dry weight, tuber number and tuber dry weight.

Antioxidant enzymes assays

Leaves collected from the final destructive harvest (100 DAP) were subjected to biochemical analyses. Extractions of antioxidant enzymes from the leaves of the four treatments (CF and ambient CO_2 (control); CF plus double CO_2 concentrations; NF plus ambient CO_2; (iv) NF plus double CO_2), were performed according to Lee et al. (1997). Leaves were cut from each treatment and immersed in liquid nitrogen and kept in a deep freezer at 80°C until the analyses.

Samples were weighed and ground at about 0°C in 25 m Tris-HCl buffer containing 3 mM $MgCl_2$, then the homogenates were centrifuged at 20,000 for 15 min at 4°C (Centrifuge 17 S/RS, Heraeus Sepatech). The supernatants were used for the enzyme assays and the results were expressed on protein basis (Bradford, 1976).

All assays were performed using a final volume of 1 ml, with at least duplicate assays undertaken on each sample. Moreover, the assays were end point determinations (Hassan, 2006).

Superoxide dismutase (SOD, EC 1.15.1.1) activity was monitored according to Lee et al. (1997). The extraction contained 50 mM phosphate buffer solution (pH 7.8), 13 mM L-methionine, 63 μM nitro blue tetrazolium and 2 μM riboflavin. The ability of the extract to inhibit the photochemical reduction of nitro blue tetrazolium was determined at 560 nm (Schimadzu UV-1201 spectrophotometer). The amount of the extract resulting in 50% inhibition of nitro blue tetrazolium reaction is defined as one unit of SOD activity.

Guaiacol PX (GPX Glutathione Reductase (GR, EC, 1.11.1.7) activity was determined by adding 50 mM phosphate buffer (pH 6.1), 1% H_2O_2, 1% guaiacol to the extract and the absorbance was determined at 470 nm.

Ascorbate peroxidase (APX, EC, 1.11.1.11) activity was determined according to Maehly and Chance (1954). The reaction mixture contained 50 mM potassium phosphate, 0.5 mM ascorbate, 0.1 mM EDTA and 0.1 mM H_2O_2 and the absorbance was determined at 290 nm.

Glutathione Reductase (GR, EC, 1.6.4.2) activity was determined according to Lee et al. (1997). The enzyme activity was monitored by measuring a decrease in absorbance at 334 nm resulting from oxidation of NADH (6.2 mM⁻¹). The assay mixture contained 0.1 M Tris-HCl (pH 8.0), 1 mM EDTA, 0.1 mM NADH and 1 mM oxidized glutathione (GSSG) and the leaf extract.

Glutathione was analysed with a Schimadzu R.F. 1201 HPLC, and peaks were detected by a fluorescence detector using an excitation wavelength of 340 nm and an emission wavelength of 420 nm (Hassan, 2006).

RESULTS

Gas concentrations

Daily mean ambient CO_2 concentration was 350 + 20 ppm while elevated CO_2 (Doubled ambient) concentration was 690 + 25 ppm.

Ambient O_3 concentration was 78 nl l⁻¹ + 8 (8 hd⁻¹, seasonal average). The mean AOT40 (accumulated O_3 exposure above a threshold of 40 nl l⁻¹) value for the non-filtered air was 29600 nll⁻¹ (Table 1).

Table I Seasonal 8-h (09:00–17:00 h Egyptian Local Time) daily average of O_3 and CO_2 concentrations + I SE.

Treatment	O_3 (nl l⁻¹)	CO_2 ppm	AOT40
Control (Charcoal filtered)	15 + 3	350 + 20	00
Ambient (Non-filtered)	78 + 8	350 + 20	29,600
Elevated CO_2	15 + 3	690 + 30	00
Elevated CO_2 and O_3	78 + 8	690 + 30	29,600

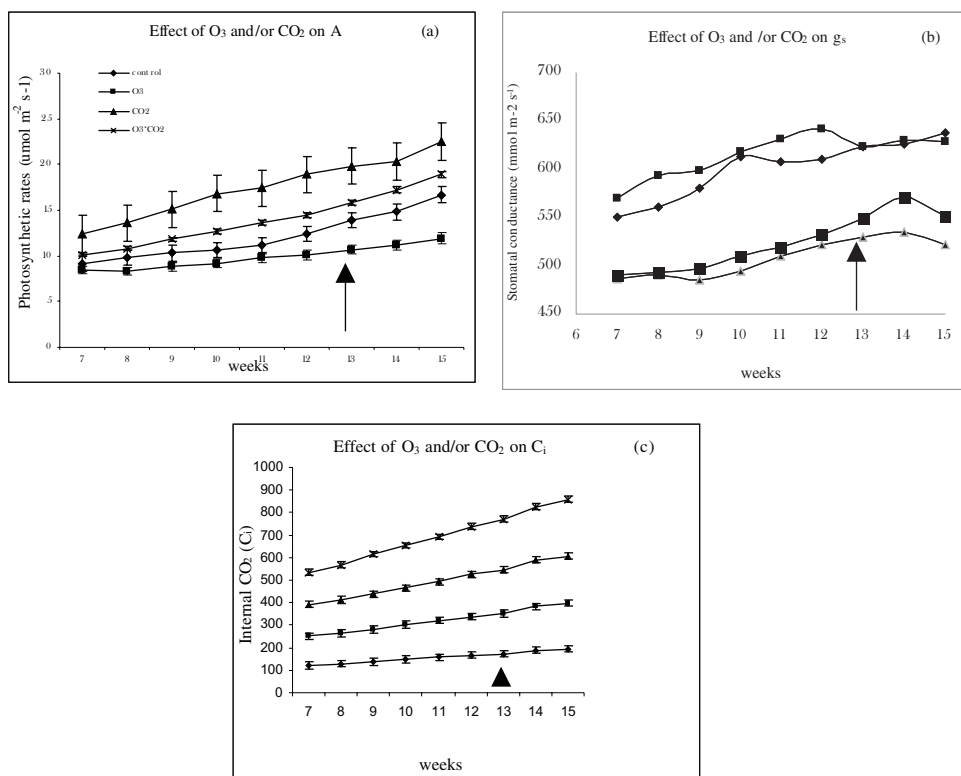**Figure 1** Effect of O_3 and/ or CO_2 on (a) Photosynthetic rates 'A' μmol m^{-2}s^{-1}, (b) stomatal conductance 'g_s' mmol m^{-2} s^{-1} and (c) Internal CO_2 'C_i' ppm. Each figure is a mean of 12 readings + 1 SE

Effects on A, g_s and C_i

O_3 decreased A by 18% while it had no significant effect ($P \geq 0.05$) on g_s and C_i (Figure 1 (a)–(c)). On the other hand, CO_2 caused increases in A, g_s and C_i by 44, 15 and 28%, respectively (Figure 1 (a)–(c)).

Interaction between O_3 and CO_2 was less than additive and CO_2 protected plants against adverse effect of O_3 on A as A, g_s and C_i were increased by 15, 13 and 12%, respectively due to this interaction (Figure 1 (a)–(c)).

Visible injury symptoms

Table 2 shows that O_3 increased Number and percentage of foliar injury by two and five fold, respectively, while exposure to elevated CO_2 had no significant ($P \geq 0.005$) on these parameters.

Exposure to O_3 and CO_2 protected potato plants against phytotoxic effect of O_3 on number of injured leaves and percentage of injury by 63 and 50%, respectively (Table 2).

Effect on yield

O_3 decreased number and weight of tubers by 41 and 21%, respectively, while CO_2 increased them by 37 and 20%, respectively (Table 3). However, interaction between CO_2 and O_3 increased number of tubers by 35% while it had no significant effect ($P \geq 0.005$) on weight of tubers (Table 3).

Table 2   Effect of CO$_2$ and O$_3$ on foliar injury symptoms of potato leaves

Parameter	Control	Ambient	Elevated CO$_2$	Elevated CO$_2$ and O$_3$
Number of injured leaves	16[a]	38[c]	14[a]	21[b]
Degree of injury	0.14[a]	0.82[c]	0.12[a]	0.52[b]

[a]Means not followed by the same letter are significantly different from each other at $P \leq 0.05$.
[b]Data presented show the mean values ($n = 25$) for the proportion of sampled leaves were indicated and the leaf area damaged assessed on a scale where 1 = no visible injury or damaged area.
[c]5 = complete infection or damaged area.

Table 3   Effect of CO$_2$ and O$_3$ on potato tuber yield

Parameter	Control	Ambient	Elevated CO$_2$	Elevated CO$_2$ and O$_3$
Weight (kg)	7.83[b]	6.21[a]	9.42[c]	8.23[b]
No. of tubers (per plant)	6.01[b]	3.56[a]	8.21[c]	8.10[c]

Legends as Table 2. Figures are means of ten plants per treatment.

Table 4   Activities of GR, guaicol peroxidase (POD), SOD and APX in extract of leaves (100 DAP), in response to O$_3$ and/ or CO$_2$

Treatment	GR activity (n mol cm^{-2}s^{-1})	POD activity (n mol cm^{-2}s^{-1})	SOD activity (μcm^{-2})	APX (μcm^{-2})
Control (Charcoal filtered)	0.28[a]	5.62[a]	4.61[a]	0.32[a]
Ambient (Non-filtered)	0.33[b]	7.63[b]	4.12[a]	0.29[a]
Elevated CO$_2$	0.25[a]	8.21[c]	4.23[a]	0.34[a]
Elevated CO$_2$ and O$_3$	0.24[a]	5.82[a]	2.35[a]	0.31[a]

Legends as Table 2. Figures are means of ten plants per treatment.

Antioxidant enzymes

It was found that O$_3$ caused significant ($P \leq 0.0\,1$) increase in GR and POD by 18 and 35%, respectively, while it had no significant effect ($P \geq 0.005$) on SOD and APX (Table 4)

POD activity was increased by 46% due to exposure to elevated CO$_2$, while other antioxidants showed no significant ($P \geq 0.005$) response (Table 4). Furthermore, exposure to both gases, simultaneously, had no significant ($P \geq 0.005$) effect on any of antioxidant enzymes (Table 4).

DISCUSSION

The mean AOT40 value for ambient O$_3$ levels throughout the entire course of the experiment (29,600 nl l^{-1}) was very comparable to that recorded in Southern Europe (30,000 nl l^{-1}) and much higher than that recorded in central and northern Europe (Donnelly et al., 2001; Fowler et al., 1999). Moreover, Heagle et al. (2003) found a similar AOT40 in the USA (27,600). Very Recently, Calvo et al. (2009) reported that AOT40 in Spain was 25,283 nl l^{-1} and this is very comparable to the AOT40 recorded

in the present study and this is may be due to that both countries have a Mediterranean climate.

This AOT40 is known to cause foliar injury symptoms on many plants including potato (Calvo et al., 2007; 2009; Hayes et al., 2009; Heagle et al., 2003). Reports on foliar injury of potato caused by O_3 and/or CO_2 are fairly common.

It was found in the present study that elevated CO_2 confer some protection against O_3, and it was clear that stress caused by CO_2 predisposed leaves to injury caused by O_3 and not vice versa and this explain how they interact to alleviate foliar injury. These results are in agreement with the results of Donnelly et al. (2001) and Finnan et al. (2002), who found that elevated CO_2 (680 ppm) has protected Potato plants (Cv. Binje) from toxic effects of O_3 (65 nl l[-1]). However, this was not the case in the study of Heagle et al. (2003) who indicated that exposure to elevated CO_2 (715 ppm) did not protect Potato plants (Cv. Dark Red Norland) from toxic effects of elevated O_3 (80 nl l[-1]), they found that at 80 nl l[-1] O_3 foliar injury increased as CO_2 concentration increased and they could not explain why interaction between these pollutants exacerbate foliar injury.

Gas exchange measurements indicated that non-filtered air tended to reduce stomatal conductance in Cv. Kara and this is in agreement with the general trend in the literature (e.g. Calvo et al., 2009; Dann and Pell, 1989; Emberson et al., 2000; Landolt et al.,1997) on many crops including potatoes. In agreement with the results of Calvo et al. (2009) the results indicated more efficient photosynthesis and less need for stomata to be open to allow CO_2 influx. Booker and Fiscus (2005) found increases in A at elevated CO_2 and they suggested that this could compensate for the inhibitory

effect of O_3 on plant metabolism and growth by increasing carbon assimilation over that at ambient CO_2. This would be affected at elevated CO_2 by increased photoassimilation and decreased photorespiration (Long and Naidu, 2002; Ziska and Bunce, 2008).

CO_2-induced reduction in O_3 flux is an important component in the prevention of O_3 injury (Booker et al., 2005; Fiscus et al., 2005). However, Cardaso-Vilhena et al. (2004) stated that there are other factor(s) in addition to decreased O_3 flux are involved in the protective effect of elevated CO_2 against O_3 damage. Exact mechanism whereby elevated CO_2 protects plants fro phytotoxic effects of O_3 remain unknown (Heagle et al., 2003). Protection may result from increased rates of repair of incipient O_3 injury, but measures to identify biochemical mechanisms to explain this possibility are rudimentary (Booker and Fiscus, 2005; Heagle et al., 2003).

Although elevated CO_2 did not alter GR, SOD and APX activities in ways that appeared instrumental in O_3 detoxification processes, POD activity was increased in response to elevated CO_2. However, McKee et al. (1997) concluded that amelioration of O_3 injury by elevated CO_2 was not primarily related to effects on antioxidant metabolism, but rather decreased O_3 flux. Responses of antioxidant metabolism to elevated CO_2 are variable but it appears that growth at elevated CO_2 reduces oxidative stress, although it is unclear whether the accompanying metabolic changes affect plant susceptibility to oxidative stress from additional factors such as O_3, drought and salinity (Booker and Fiscus, 2005; Sgherri et al., 2000; Wustman et al., 2001). Kurz et al. (1998) found a decrease in glutathione concentration at elevated concentrations of CO_2, and they attributed this to a lower mineral:C content because glutathione functions as a reserve of reduced S

(Booker and Fiscus, 2005). However, this was not the case in the present study as elevated CO_2 did not affect significantly ($P \geq 0.05$) glutathione concentration, and this is in agreement with the results of McKee et al. (1997) and Sgherri et al. (2000) on wheat and alfalfa, respectively. Moreover, there was no significant effect of CO_2 on SOD and AA concentrations, while exposure to elevated CO_2 caused significant increase in POD activity. Schwanz and Polle (2001) found a lower level of AA, while Sgherri et al. (2000) found higher AA concentrations at elevated CO_2. Increased POD activity was possibly the result of greater carbohydrate supply at elevated CO_2 (e.g. Booker and Fiscus, 2005; Sgherri et al., 2000).

In the present study, increased GR concentration and POD activity in ambient air can be interpreted as response to oxidative stress imposed by O_3 (Chernkova et al., 2000). However, the lack of significant changes in activities of SOD and AA in ambient air, which also observed in other O_3 studies (e.g. Azevedo et al., 1998), differed from studies where activities of these enzymes increased in response to O_3 (Chernkova et al., 2000).

The variability in the response of antioxidants to elevated O_3 and CO_2 among studies reflects differences in the magnitude of the perceived oxidative stress, the species-specific mechanisms involved in responses to changes in redox status, the plant capacity to cope with additional stress(s), experimental protocols and environmental conditions. Therefore, further studies are needed to further the understanding of the response of antioxidant metabolism to elevated O_3 and CO_2 singly and in combination.

Pell et al. (1988) found a reduction in number and weight of Norchip potato tubers by 31 and 14% respectively, when exposed to 80 nl l⁻¹ and this in agreement with the results of the present study where both parameters were reduced by 41 and 21% at 78 nl l⁻¹ O_3. Heagle et al. (2003) reported 31% reduction in number of potato tubers grown in open top chambers in the USA when exposed to 80 nl l⁻¹ O_3. Moreover, Booker et al. (2005) found that tuber yield decreased by 31 and 14% in Cv. Dark Red Norland when exposed to 80 and 45 nl l⁻¹ O_3, respectively. Very Recently, Calvo et al. (2009) found that ambient non-filtered air (16.3 nl l⁻¹ O_3) and non-filtered air supplied with O_3 (58.8 nl l⁻¹) significantly decreased tuber fresh weight by 29 and 44%, respectively in Potato plants Cv. Desiree grown in Spain. Moreover, O_3 alters crop quality as it reduces starch content of potato (Vorne et al., 2002). Such alteration in nutritional quality may become increasingly important in developing countries where demand for food is stable or increasing slowly. However, it is unclear whether agricultural production can meet the growing demand for the food in developing countries where human populations are increasing rapidly (Chameides et al., 1999; Hassan, 2006; Hassan et al., 1995; WHO/UNEP, 1992). In such areas, potato offers great potential to meet food requirements due to its high ratio of edible to non-edible components.

In general, the results of the present study are in agreement with the most previous work on potato and this indicate that O_3 levels in Egypt are high enough to cause reductions in growth and yield of economic plants, moreover, potato plants cv. Kara are sensitive to O_3 as Western cultivars.

Overall, increasing concentrations of ambient O_3 will suppress the increase in growth and yield of crops resulting from rising CO_2 concentrations. On the other hand, O_3 injury is lessened by elevated CO_2 due to reduced O_3 influx, increased carbon assimilation and to as yet undetermined additional factor(s) (Booker and Fiscus, 2005).

BIOGRAPHY

Prof. Ibrahim Hassan received his PhD from Imperial College, London University in 1994 in the field of air pollution effect on plants. He is a Professor of plant physiology and environmental botany at Alexandria University, Egypt. Currently, he is in a sabbatical leave to KSA. He is principal investigator of a project entitled Ozone Effects on Egyptian Crops in Egypt funded by the Department of International Development (DFID) and the British Council (BC) in collaboration with Imperial College, London.

REFERENCES

Ainsworth, E.A. and Long, S.P (2005) 'What we have learned from 15 years of free-air CO_2 enrichment (FACE)? A meta-analytic review of the response of photosynthesis, canopy properties and plant production to rising CO_2', *New Phytology*, Vol. 165, pp.351–372.

Azevedo, R.A., Alas, R.M., Smith, R.J. and Lea, P.J. (1998) 'Response of antioxidant enzymes to transfer from elevated CO_2 to air and O_3 fumigation, in the leaves of wild-type and a catalse deficient mutant of barely', *Physiology Plant*, Vol. 104, pp.280–292.

Booker, F.L. and Fiscus, E.L. (2005) 'The role of O_2 flux and antioxidants in the suppression of O_3 injury by elevated CO_2', *Environmental and Experimental Botany*, Vol. 56, pp.2139–2151.

Bradford, M.M. (1976) 'A rapid and sensitive method for the utilizing the principle of protein dye binding', *Analytical Biochemistry*, Vol. 72, pp.248–254.

Calvo, E., Martin, C., and Sanze, M.J. (2007) 'Ozone sensitivity differences in five tomato cultivars: visible injury and effects on biomass and fruits', Water, Air and Soil Pollution, Vol. 186, pp. 167–181

Calvo, E., Calvo, I., Jimenez, A., Porcino, J.L. and Sanze, M.J. (2009) 'Using manure to compensate O_3-yield loss in potato plants cultivated in the east of Spain', *Agriculture, Ecosystem and Environment*, Vol. 131, pp.185–192.

Cardoso-Vilhena, J., Balaguer, L., Eamus, D., Ollerenshaw, H. and Barnes, J. (2004) 'Mechanisms underlying the amelioration of O_3-induced damage by elevated atmospheric concentrations of CO_2', *Journal of Experimental Botany*, Vol. 55, pp.771–781.

Chameides, W.L., Li, X. and Tang, X. (1999) 'Is O_3 pollution affecting crop yield in China?' *Geophysical Research Letters*, Vol. 26, pp.867–870.

Chernkova, T., Robinson, J.M., Lee, E.H. and Mulchi, C.L. (2000) 'O_3 tolerance and antioxidant enzyme activity in soybean cultivars', *Photosynthtica Research*, Vol. 64, pp.15–26.

Dann, M.S. and Pell, E.J. (1989) 'Decline of activity an quantity of ribulose bisphosphate carboxylase/oxygenase and net photosynthesis in O_3-treated potato foliage', *Plant Physiology*, Vol. 91, pp.427–432.

Donnelly, A., Craigon, J., Black, C.R., Colls, J.J. and Landon, G. (2001a) 'Does elevated CO_2 ameliorate the impact of O_3 on chlorophyll content and photosynthesis in potato (*Solanum tuberosum* L.)', *Physiology Plant*, Vol. 111, pp.501–511.

Donnelly, A., Craigon, J., Black, C.R., Colls, J.J. and Landon, G. (2001b) 'Elevated increase biomass and tuber yield in potato even at high O_3 concentrations', *New Phytologist*, Vol. 149, pp.265–274.

Emberson, L., Ashmore, M., Cambridge, H.M., Simson, D. and Tuovinen, J.P. (2000) 'Modeling stomatal O_3 flux across Europe', *Environmental Pollution*, Vol. 109, pp.403–413.

Finnan, J.M., Donnelly, A., Borke, J.I. and Jones, M.B. (2002) 'The effects of elevated CO_2 and O_3 on potato (*Solanum tuberosum* L.) yield', *Agricultural Ecosystems and Environment*, Vol. 88, pp.11–22.

Fiscus, E.L., Booker, F.L. and Burkey, K.O. (2005) 'Crop responses to O_3: uptake, modes of action, carbon assimilation and partitioning', *Plant Cell Environment*, Vol. 28, pp.1365–3040.

Fiscus, E.L., Reid, C.D., Miller, J.E. and Heagle, A.S. (1997) 'Elevated CO_2 reduces O_3 flux and O_3-induced yield losses in soybean: possible implications for elevated CO_2

studies', *Journal of Experimental Botany*, Vol. 48, pp.307–313.

Gardner, S.D.L, Freer-Smith, P.H., Tucker, J. and Taylor, G. (2005) 'Elevated CO_2 protects poplar (*Populous trichocarpa* x p.deltious) from damage induced by O_3: identification of mechanism', *Environmental Pollution*, Vol. 32, pp.221–235.

Hassan, I.A. (1998) 'Effects of O3 on crop quality: a study case from Egypt', in L.J. De Kok and I. Stulen (Eds). *Responses of Plants to Air Pollution and Global Change*, pp.323–328, Leiden, the Netherlands: Backhuys Publishing.

Hassan, I.A. (2006) 'Physiological and biochemical responses of potato (*Solanum tuberosum.* cv Kara) to O_3 and antioxidant chemicals: possible roles of antioxidant enzymes', *Annals of Applied Biology*, Vol. 148, pp.197–206.

Hassan, I.A., Ashmore, M.R. and Bell, J.N.B. (1995) 'Effects of O_3 on radish and turnip under Egyptian field conditions', *Environmental Pollution*, Vol. 89, pp.107–114.

Hayes, F., Mills, G. and Asmore, M. (2009) 'Effects of inter- and intra-species competition and photosynthesis in mesocosms of *Lolium perenne* and *Trifolium repenes*', *Environmental Pollution*, Vol. 157, pp.208–214.

Heagle, A.S., Miller, J.E. and Pursley, W.A. (2003) 'Growth and yield responses of potato to mixtures of CO_2 and O_3',. *Journal of Environmental Quality*, Vol. 32, pp.1603–1610.

Keutgen, A.K. (2005) 'Photosynthetic response of plants to O_3 and its effects on nutrient status', *Environmental Expert Botany*, Vol. 53, No. 3, pp.271–290.

Kurz, C., Schmieden, U., Strobel, P. and Wild, A. (1998) 'The combined effects of CO2, O3 and drought on the radical scavenging system of young oak trees (Quecus petraea)- a phyothron study. Chemosphere, 36, pp. 783–788.

Landolt, W., Gunthardt-Gorg, M.S., Pfenniger, I., Eining, W., Hampp, R., Maurer, S. and Matyssek, R. (1997) 'Effect of fertilization on O_3-induced changes in metabolism of birch (*Betula pendula*) leaves', *New Phytologist*, Vol. 137, pp.389–397.

Lee, E.H., Upadhyaya, A., Agrawal, M. and Rowland, R.A. (1997) 'Mechanism of

ethylenediurea (EDU) induced O_3 protection: re-examination of free radical scavenger systems in snap bean exposed to O_3', *Environmental and Experimental Botany*, Vol. 38, pp.199–209.

Long, S.P. and Naidu, S.L. (2002) 'Effects of oxidants at the biochemical, cell and physiological levels, with particular references to O_3', in J.N.B. Bell and M. Treshow (Eds). *Air Pollution and Plant Life*, Chichster: John Wiley & Sons, pp.69–98.

Maehly, A.C. and Chance, B. (1954) 'The assay of catalase and peroxidase', in D. Glick (Ed). *Methods of Biochemical Analysis*, pp.357–424, New York: Interscience.

McKee, I.F., Eibmeier, M. and Polle, A. (1997) 'Enhanced O_3 tolerance in wheat grown at elevated CO_2 concentration: O_3 exclusion and detoxification', *New Phytologist*, Vol. 137, pp.275–284.

McKee, I.F., Mulholland, B.J., Craigo, J., Black, C.R. and Long, S.P. (2000) 'Elevated concentrations of CO_2 protects against and compensate for O_3 damage to photosynthetic tissues of field-grown wheat', *New Phytologist*, Vol. 146, pp.427–435.

Morison, J.L. and Morecrof, M.D. (2008) (Eds) *Plant Growth and Climate Change*, London: Blackwell Pub. Ltd.

Oksanen, E., Friewald, V., Prozherrina, N. and Rousi, M. (2005) 'Photosynthsis of birch (*Betula pedula*) is sensitive to springtimae and O_3', *Canadian Journal of Forest Research*, Vol. 35, No. 3, pp.703–712.

Olszyk, D,M., Wise, C.(1997). 'Interactive effect of elevated CO2 and O3 on rice and flacca tomato' Agriculture, Ecosystem and Environment, 66, pp. 1–10.

Prather, M., Ehalt, D. and Dentener, F. (2001) 'Atmospheric chemistry and greenhouse gases', in J.T. Houghton, Y. Ding, D.J. Griggs, M. Noguer, P.J. Van der Linden, X. Dai, K. Maskell and C.A. Johnson (Eds). *Climate Change 2001: The Scientific Basis. Contribution of working group I to the third Assessment report of IGP Climate Change*, UK: Cambridge University Press, pp.239–287.

Pell, E.J., Pearson, N.S. and Vinten-Johansen, C. (1988). 'Qualitative and quantitative effects of O3 and/or SO2 on field- grown

potato plants. Environmental Pollution, 53, pp. 171–186

Pole, A. and Pell, E.J. (1999) 'Role of CO_2 in modifying the plant response to O_3', in Y. Luo and H.A. Monney (Eds). CO_2 *and Environmental Stress*, San Diego, USA: Academic Press, pp.93–123.

Pritchard, S.G., Ju, Z., Van-Santen, E., Qiu, J., Weaver, D.B., Prior, S.A. and Rogers, H.H. (2000) 'The influence of elevated CO_2 on the activities of antioxidative enzymes in two soybean genotypes', *Australian Journal of Plant Physiology*, Vol. 27, pp.1061–1068.

Rämo, K., Slotte, H., Kanerva, T., Ojanpera, K. and Manninen, S. (2006) 'Growth and visible injuries of four *Centaurea jacea* L. ecotypes exposed to elevated O_3 and CO_2', *Environmental and Experimental Botany*, Vol. 58, pp.287–298.

Ranford, J. and Reiling, K. (2007) 'O_3 induced leaf loss and decreased leaf production of European holly (*Ilex aquifoium* L.) over multiple seasons', *Environmental Pollution*, Vol. 145, pp.355–364.

Reid, C.D. and Fiscus, E.L. (1998). Effects of elevated CO2 and/or O3 on limitations of CO2 assimilation in soybean (Glycine max). Journal of Experimental Botany, 49, pp. 885–995.

Schapendok, A.H., Van Oijen, M.M., Dijkstra, P., Post, C.S., Jordi, W.R.M. and Stoopen, G.M. (2000) 'Effects of elevated CO2 concentration on photosynthetic accumulation and productivity of two potato cultivars grown in OTCs', *Australian Journal of Plant Physiology*, Vol. 27, pp.875–886.

Sgherri, C.L.M., Salvateci, P., Menconi, M., Raschi, A. and Navari-Izzo, F. (2000). Interaction between drought and elevated CO2 in the response of alfalfa plants to oxidative stress. *J. Plant Physio.*, 156: 360 366.

Schwanz, P. and Polle, A. (2001) 'Differential stress response of antioxidative systems to drought in pendunculate oak (*Quercu robur*) and maritime pine (*Pinus pinaster*) grown under high CO2 concentration',

Journal of Experimental Botany, Vol. 52, pp.133–134.

Topa, M.A., Vanderklein, D.W. and Corbin, A. (2001) 'Effects of elevated O_3 and Low light on diurnal and seasonal carbon gain in sugar maple', *Plant, Cell and Environment*, Vol. 24, No. 7, pp.663–667.

Vandermeiren, K., Black, C., Pleijel, H. and DeTemmerman, L. (2005) 'Impact of rising O_3 on potato: effects on photosynthesis, growth, productivity and yield quality', *Plant Cell and Environment*, Vol. 28, pp.982–996.

Vorne, V., Ojanperä, K., De Temmerman, L., Bindi, M., Högy, P., Jones, M.B., Lawson, T. and Persson, K. (2002) 'Effects of elevated CO_2 and O_3 on potato tuber quality in the European multiple-site experiment "CHIP- Project"', *European Journal of Agronomy*, Vol. 17, pp.369–381.

WHO/UNEP (1992) *Air Pollution in Megacities of the World*, London: Blackwell Pub. Ltd.

Wustman, B.A. Oksanene, E., Karnosky, D.F., Noormets, A., Isebrands, J.G., Pregitzar, K.S., Henrey, G.R., Sober, J., Podila, G.K. (2001). 'Effects of elevated CO2 and O3 on aspen clones varying in O3 sensitivity: can CO2 ameliorate the harmful effects of O3? ' Environmantal Pollution, 115, pp. 473–481.

Wolf, J. and Van Oijen, M. (2003) 'Model simulation of changes in climate and atmospheric CO_2 and O_3 on tuber yield potential of potato (cv. Bintje) in the European Union', *Agricultural Ecosystems and Environment*, Vol. 94, pp.141–157.

Yonekura, T., Yoshidone, M., Waltanabe, M., Honda, Y., Ogiwara, I. and Izuta, T. (2004) 'Carry-over effects of O_3 and waters stress on leaf phonological charcterstics and bud frost hardiness of *Fagus crenata* seedlings', *Tree Structure and Function*, Vol. 18, No. 5, pp.581–588.

Ziska, L.H. and Bunce, J.A. (2008) 'Plant response to rising CO2', in J.L. Morison and M.D. Morecrof (Eds). *Plant Growth and Climate Change*, pp.17–47, London: Blackwell Pub. Ltd.