WILL INCREASING OZONE POLLUTION ASSOCIATED WITH GLOBAL CLIMATE CHANGE ALTER CROP TOLERANCE TO HERBICIDES ?

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ABSTRACT

The latest estimates have indicated that climate change will be associated with a 17-40% increase in ozone pollution over the next thirty years. Such an increase will cause a reduction in crop yield, and may also alter crop tolerance to pesticides. Studies in the 1970s and 1980s have provided clear indications of interactions occurring between herbicides and ozone. The nature of these interactions appears to vary between synergistic, additive and antagonistic depending on species, herbicide and ozone dose. For example, recent work in this laboratory has indicated an antagonistic interaction between ozone and phenmedipham in sugar beet. The possible significance of the interactions between ozone and herbicides are discussed in this paper.

INTRODUCTION

A prediction of the magnitude of changes in the global climate has been the subject of intensive research in the last decade. Recent estimates suggest that a small rise in temperature will be associated with increasing CO_2 concentration and altered rainfall patterns by the middle of the next century. These changes will necessitate a flexible approach to agriculture allowing change to crops more suited to the prevailing climatic conditions, and altered pesticide usage to control overwintering weeds, pests and diseases.

A frequently overlooked environmental change associated with global warming is that air pollution is also predicted to increase. For example, the concentration of the pollutant ozone is likely to increase by 17-40% over the next 30 years (Hough and Derwent, 1990). Such predictions are based on historical records of ozone concentrations, projected increases in the concentrations of ozone precursors (see below), and the impact of other 'greenhouse' gasses and global warming (Hough and Derwent, 1990; Ashmore and Bell, 1991). Experiments conducted in the USA and Europe have shown that a rise in ozone pollution would be associated with yield loss in sensitive crops such as beans, soybean and wheat (Heagle *et al.*, 1988; Fuhrer *et al.*, 1989). An area of further concern for agricultural production is the possibility that interactions between ozone and certain pesticides may lead to enhanced crop damage and add to this yield loss. This review will consider the interactions between ozone and herbicides. The effects of ozone alone will be briefly described, followed by a review of the literature on interactions between herbicides and ozone, and a description of recent work conducted in this laboratory.

SOURCES OF OZONE POLLUTION

The subject of ozone often leads to confusion by the general public. In the upper atmosphere (stratosphere), a layer of ozone is essential to protect the earth from damaging ultraviolet radiation from the sun. A reduction in the thickness of this layer by chemicals such as chloroflourocarbons is of major concern because of, for example, a potential increase in the number of skin cancers in humans. However, ozone is also present in the lower atmosphere (troposphere), due to either mixing from the stratosphere, or to photochemical production. In the latter case, ozone is regarded as a harmful pollutant and its production is closely associated with human activities.

The following simplified equation describes the atmospheric equilibrium between NO_2 (from car exhaust fumes and power station emissions), O_2, NO and O_3:

$$\underset{\text{Nitrogen Dioxide}}{NO_2} + \underset{\text{Oxygen}}{O_2} \overset{\text{sunlight}}{\rightleftharpoons} \underset{\text{Nitric Oxide}}{NO} + \underset{\text{Ozone}}{O_3}$$

The balance of this equation is shifted towards O_3 production when the action of sunlight on volatile organic hydrocarbons (VOCs, present in car exhaust fumes and in the emissions from the chemical and oil industry) produces peroxyl radicals which react with NO to prevent the back reaction (QUARG, 1993). Photochemical episodes occur when these reactions are enhanced, for example, during the hot, clear and still days associated with summer anticyclonic weather systems.

In the UK, ozone episodes of upto 150 nl l^{-1} are superimposed on a background concentration of 20-40 nl l^{-1} (QUARG, 1993). More usually, during a UK ozone episode, the concentration rises to a maximum of 60-90 nl l^{-1} by mid afternoon, and declines to the background concentration overnight (eg Figure 1). This diurnal cycle of ozone production persists for as long as the precursors (NOx and VOCs) are present, and the weather conditions are conducive to photochemical ozone production (usually 2-3 days). On a European scale, the mean daily maximum concentration for the period April to September has been predicted to exceed 70 nl l^{-1} in central areas of Europe, and 50 nl l^{-1} over most of continental Europe and the southern half of the UK (Simpson, 1993).

Several parameters are used to describe the concentration of ozone. Long-term averages reflect the episodic nature of ozone pollution, and include parameters such as the 12h mean (0600-1800h) and the mean concentration during daylight hours. To gain more information about the concentration during episodes, the concentration is also averaged during the 7 or 8 hours of the day with the highest ozone concentration (eg 1100 - 1800h, figure 1). The choice of 7 or 8 hours as an averaging period depends on several factors including the breadth of the ozone curve and the daylength at the location being studied. International research is currently focused on the use of the accumulated ozone dose above a threshold concentration (eg dose above 30 nl l^{-1} during the growing season), which allows the cumulative frequency and severity of ozone episodes to be compared between locations.

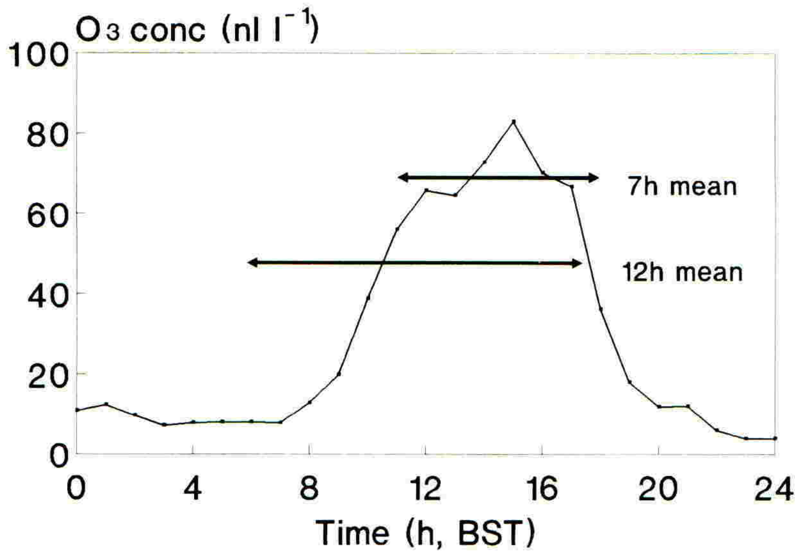

Figure 1: A typical diurnal curve for ozone concentration during a photochemical episode. The data were recorded at Sutton Bonington, a rural site in the English East Midlands on 25 July 1989.

OZONE POLLUTION IN A CHANGING CLIMATE

Predictions of future tropospheric ozone concentrations are based on estimates of the likely increases in ozone precursors (NOx and VOCs), and on retrospective modelling of changes in ozone concentration during the last century. Volz and Kley (1988) reinvestigated a set of measurements of ozone which were made at the Montsouris laboratory near Paris between 1876 and 1910. They concluded that the ozone concentrations 100 years ago averaged 10 nl l^{-1}, and were less than half of the current rural concentrations. This rise in concentration was strongly influenced by photochemical production due to increased levels of NOx. Hough and Derwent (1990) were able to simulate this change using a global tropospheric model which linked the chemistry of the pre-industrial atmosphere with that of the present day. Estimates of the future growth in emissions of NOx were incorporated into the model to predict the ozone concentration in the year 2020. Hough and Derwent based their predictions on the upper and lower bands of estimated NOx emissions and predicted that tropospheric ozone will increase in concentration by 17 - 40% (from 30 to 35-42 nl l^{-1}) in the Northern hemisphere. Regional trends in ozone concentration are more difficult to determine due to the lack of long-term data from continuous monitoring sites (Lefohn *et al.*, 1992).

A further consideration for the inclusion of predictions of ozone concentrations in a discussion of the effects of global climate change, is that ozone is also a 'greenhouse' gas (Hough and Derwent, 1990). Although less important than carbon dioxide and methane, ozone traps long-wave radiation emitted from the earth's surface and thus an increase in ozone concentration will contribute to global warming. The implications of global warming for pesticide usage are considered elsewhere in these Proceedings.

OZONE POLLUTION AND CROP YIELD

Predictions of the future implications for crop production of rising ozone concentrations necessitates an understanding of the current effects of ozone on crop yield. Early research (eg Heagle *et al.*, 1973) established that ozone concentrations in many areas of the USA, such as North Carolina and California, were sufficiently high to induce visible injury and yield loss in sensitive crops. This led to extensive research to establish the magnitude of crop loss. A programme of research, NCLAN (National Crop Loss Assessment Network), was established in the 1980s which resulted in the production of ozone dose response functions to estimate the magnitude of yield loss, and sensitivity of several crops (Heck *et al.*, 1988).The results of this work were used to determine the economic benefits for agricultural production of a reduction in ozone pollution (Adams *et al.*, 1988). It was estimated that 25% and 40% decreases in ozone pollution would be associated with 1.9% and 2.8% increases, respectively, in total agricultural revenue (representing 1.9 or 3 billion $US in 1982). The estimated benefit of a 25% decrease in pollution ranged from a 1.1% increase in revenue in the northern plains, through 4.8% in the Corn belt to 19.8% in the north east.

More recent work in Europe (EOTC, European Open-Top Chamber Programme) has resulted in the production of ozone dose-response relationships for European cultivars of crops such as wheat and beans (Fuhrer *et al.*, 1989; Skarby *et al.*, in press; Colls *et al.*, in press). Examples of the effects of ozone on crop yield are provided in Table 1. This information is currently being used to determine the extent of crop losses in Europe due to ozone pollution. The emphasis of this research is based on the determination of a critical ozone dose above a threshold concentration which when exceeded results in yield loss. Ultimately, European maps will be produced which will show the magnitude of yield loss in areas of Europe where exceedance of the critical ozone dose coincides with the growth of sensitive crops. Models of increases in ozone pollution associated with climate change could then be used to predict future increases in yield losses in these areas.

In many parts of Europe and the USA, ozone also induces visible injury in crops. The symptoms are typically bronze flecking or chlorosis, depending on species (Figure 2). These symptoms are associated with disruption of cellular membranes and inhibition of photosynthesis (Farage *et al.*, 1991; Ojanpera *et al.*, 1992; Sanders *et al.*, 1992), and increases in the levels and activity of cellular antioxidants (Heath, 1988). At the whole plant level, these cellular changes are manifested by decreases in leaf area and leaf area duration, resulting in a reduction in the amount of assimilate partitioned into the fruit of the crop (Unsworth *et al.*, 1984).

Table 1: Yield change in bean (*Phaseolus vulgaris*) and wheat (*Triticum aestivum*) with increasing ozone concentration

Growing Season Mean O_3 Concentration* (nl l^{-1})	Percentage Yield (relative to 10 nl l^{-1}) Bean	Wheat
10	100	100
20	98	97
30	92	93
40	85	87
50	75	81
60	63	73

* 7h for bean and 8h for wheat

The data are calculated from Colls *et al.*, in press, and Skarby *et al.*, in press, and are based on five years of field experiments conducted in 8 European countries. The seasonal mean concentration of ozone in ambient air ranged from 20 - 44 nl l^{-1} at the bean experimental sites, and from 23 - 49 nl l^{-1} at the wheat sites.

OZONE AND HERBICIDE INTERACTIONS

The previous section illustrated the effects of increasing ozone *per se* on crop yield without consideration of the effects of ozone on pesticide action. However, interactions have been identified between ozone and pesticides such as the fungicide benomyl (Taylor and Rich, 1973) and the herbicide EPTC (Hatzios, 1983). This section will consider the potential importance of herbicide:ozone interactions in a changing climate.

Research into the potential for interactions between ozone and herbicides was initiated in the 1970s and coincided with an increasing awareness of the potential effects of ozone and other pollutants (SO_2, NO_x and acid deposition) on agricultural crops. Early experiments were limited to a small range of crops and herbicides which were of relevance at that time (Table 2). In most cases, the exposure of plants pre-treated with herbicides to a simulated ozone episode resulted in additive or more than additive crop responses. For example, a synergistic interaction occurred when tobacco pre-treated with pebulate was exposed to ozone (Carney *et al.*, 1973), and additive interactions were observed between ozone and EPTC in maize (Hatzios, 1983a).

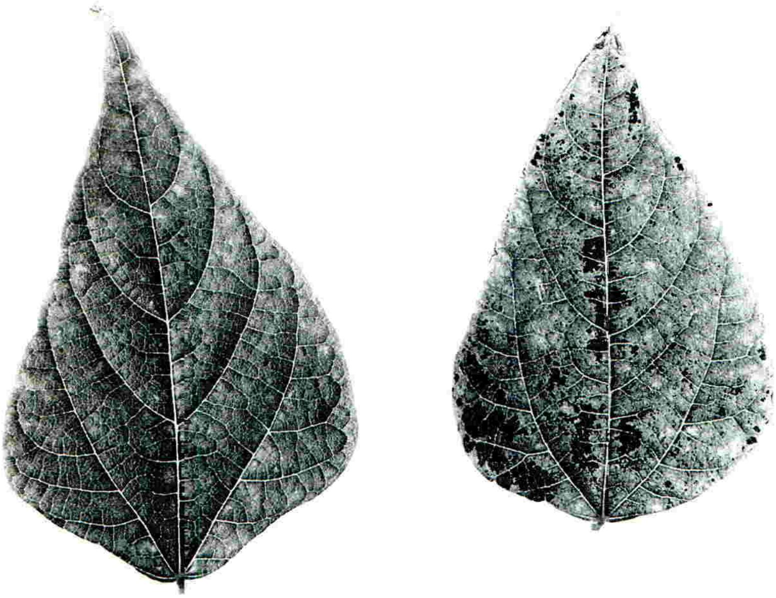

Figure 2: Ozone injury on beans (*Phaseolus vulgaris*). The leaf on the left is from a plant exposed to charcoal-filtered air and is undamaged, and the leaf on the right demonstrates typical bronze flecking which developed after exposure to ozone.

The concentration of ozone during an episode appears to be of importance in determining the nature of the interaction. Thus, exposure of metolachlor-treated sorghum to 200 nl l^{-1} of ozone for 6h generated an additive interaction, whereas exposure to 300 nl l^{-1} indicated an antagonistic response (Hatzios, 1983b) . The situation is further complicated when the timing of ozone episodes is considered relative to herbicide treatment. This factor was well illustrated in the effects of chlorsulfuron on the weed velvetleaf (Hatzios and Yang, 1983). These authors demonstrated that pre-treatment of seedlings with 0.06 or 0.12 kg ha^{-1} of chlorsulfuron resulted in an antagonistic interaction following a 6h episode of 200 nl l^{-1} ozone. However, if the ozone episode occurred before chlorsulfuron treatment, the interaction was additive. Thus, the precise nature of the interaction is dependant upon several factors, including the mode of action of the herbicide, the species and cultivar of the crop, ozone concentration and dose, and the timing of the ozone episode.

Table 2: Examples of herbicide:ozone interactions cited in the literature.

Herbicide	Ozone conc (nl l^{-1})	Interaction*	Crop	Reference
Atrazine	200 300 (6h d^{-1}, 6d)	+ A	Maize	Mersie, *et al*, 1990
EPTC	200 (6h d^{-1}, 2d)	+	Maize	Hatzios, 1983a
Metolachlor	200 (6h)	+	Sorghum	Hatzios, 1983b
Chlorsulfuron	200 (6h d^{-1}, 2d)	+	Sorghum	Hatzios and Yang, 1983
Pebulate	300 (2d)	S	Tobacco	Carney, *et al*, 1973
Benefin	300 (2d)	A	Tobacco	Carney, *et al*, 1973
Trifluralin	300 (2d)	+	Tomato	Carney, *et al*, 1973

* key: + additive; A: antagonistic; S: synergistic

Interpretation of these results in terms of European crop production is difficult due to the relatively high ozone concentrations used in the past, and to a lesser extent to the choice of crops in these early experiments. Even with the predicted increases in ozone in a changing climate, it is unlikely that episodes as high as 300 nl l^{-1} will be a common occurrence. Work in progress in this laboratory is focusing on the potential for interactions in spring-sown crops (barley, oil seed rape and sugar beet) at ozone concentrations which could be experienced now, and in the 21st century. Of particular interest in this study are the interactions between post-emergence herbicides and ozone episodes of 80 - 150 nl l^{-1} .

The central principle of our experiments is that young crop plants are sprayed with field-rate herbicide and then exposed to a simulated ozone episode in controlled environment chambers (see paper by Dixon *et al.*, these Proceedings). In one experiment, sugar beet (*Beta vulgaris* cv Saxon) was sprayed with phenmedipham (Betanal E) at a rate of 10 l ha^{-1} seventeen days after sowing. Four days later, half of the treated plants were subjected to an ozone episode comprising of 7h at 125 nl l^{-1} on the first day and 7h at 100 nl l^{-1} on the second day. Fourteen days after ozone exposure, each plant was assessed for visible injury, excised at soil level and the leaf area, and total fresh and dry weight were determined (Table 3, Figure 3).

The sugar beet plants developed similar, but distinctly different symptoms to ozone and phenmedipham. Phenmedipham induced a mottled and spreading chlorosis, whereas ozone induced chlorotic flecking of the type typically associated with this pollutant. Plants treated with ozone developed 16% chlorosis on the first pair of true leaves, whilst those treated with phenmedipham were 10% chlorotic (Figure 3). When the phenmedipham treated plants were exposed to ozone, the total injury was 18%, significantly lower than the anticipated injury if the effects of the two treatments had been additive. The effects of ozone and phenmedipham on leaf area and shoot dry weight were also less than additive. Indeed, the herbicide appeared to partially protect the sugar beet by reducing the amount of ozone injury by half (Figure 3), and by preventing an additional reduction in leaf area and dry weight.

Table 3: The effect of phenmedipham and/or ozone on sugar beet (*Beta vulgaris* cv Saxon). Analyses were performed 14 days after ozone exposure, and the values are presented as mean ± SE where n=16. Those values followed by different letters were significantly different from each other at the $p<0.05$ level (Duncan's Multiple Range Test).

	Leaf Area (cm^2)	Dry Weight (g)
Control	141 ± 10 a	0.58 ± 0.05 a
Ozone*	130 ± 14 a	0.46 ± 0.05 a
Phenmedipham**	85 ± 9 c	0.24 ± 0.03 c
Ozone and Phenmedipham	78 ± 7 bc	0.22 ± 0.02 bc

* day 1, 7h @ 125 nl l^{-1}; day 2, 7h @ 100 nl l^{-1}.
** 1.14 kg ai ha^{-1}.

In this example, an interaction occurred between two agents which are known to interfere with photosynthesis. Phenmedipham is a classic Photosystem II inhibitor which induces lipid peroxidation in thylakoid and other cell membranes (Cobb, 1992). Conversely, ozone inhibits photosynthesis by reducing carboxylation and effects on the thylakoid membrane only occur later (Farage *et al.*, 1991). The underlying mechanism(s) of this and other interactions between herbicides and ozone are currently under investigation in this laboratory (Dixon *et al.*, these Proceedings).

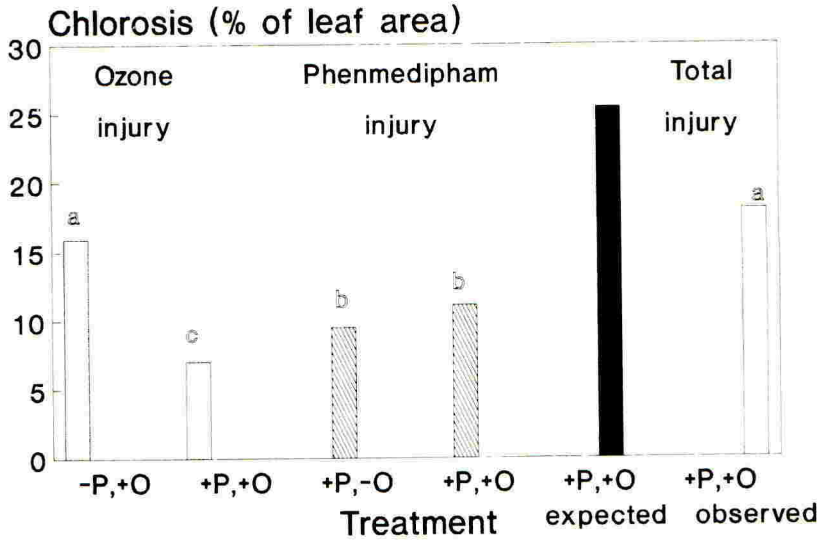

Figure 3: Chlorotic injury on the first pair of leaves of sugar beet (*Beta vulgaris* cv Saxon) treated with ozone (O) and/or phenmedipham (P). Values with different letters are significantly different from each other at the $p<0.05$ level (Duncan's Multiple Range Test).

The majority of studies on herbicide:ozone interactions have concentrated on reporting gross changes in plant weight, and have not studied the physiological consequences of interactions. However, a series of papers by Hodgson and co-workers examined the effects of ozone on diphenamid uptake, movement and metabolism in hydroponically-grown pepper and tomato exposed continuously to 240-300 nl l^{-1} of ozone (Hodgson *et al.*, 1973, 1974, Hodgson and Hoffer, 1977). These experiments showed that ozone did not affect the absorption and translocation of diphenamid in pepper (Hodgson and Hoffer, 1977). In tomato, more detailed metabolic studies revealed an increase in the rate of metabolism in ozone exposed plants, and a shift towards the production of more polar conjugates (Hodgson *et al.*, 1973, 1974).

Several hypotheses on the mechanism(s) of ozone:herbicide interactions can be proposed. Synergistic interactions imply that the herbicide in some way predisposes the plant to ozone stress. Possible mechanisms might be an overloading of the naturally occurring free radical scavenging systems following treatment with a photosynthetic inhibitor herbicide which would allow the free radicals released in ozone action to have greater effect. For other herbicides such as clopyralid, which may interact synergistically with ozone (Sanders, unpublished), stress ethylene may be involved in the interaction. Clopyralid induces stress ethylene production (Thompson, 1989), and Mehlhorn *et al.*, (1991) have suggested that stress ethylene production may be necessary for the induction of ozone injury. Antagonism might also occur between ozone and herbicides if the herbicide induces stomatal closure in the plants. This would prevent ozone uptake and thus reduce the impact of the ozone. A similar effect occurs when drought-stressed plants are exposed to ozone (Tingey *et al.*, 1982). A further potential mechanism for antagonism might be the induction of cellular antioxidants by herbicides which would reduce the impact of ozone episodes. Clearly, the exact mechanism of the interaction would depend upon the chemical nature and mode of action of the herbicide being studied, and the target species.

CONCLUSION

The predicted increase in O_3 pollution associated with global climate change will become an increasingly important concern for agriculture. The yields of commonly-grown crops (eg wheat, beans, soybean and maize) are likely to decrease in response to the effects of ozone pollution *per se* on growth and development. The evidence reviewed in this paper also indicates that the tolerance of crops to some herbicides may also be altered when herbicide application coincides with O_3 episodes. In some cases, O_3 pollution would decrease crop tolerance (eg EPTC and pebulate), whereas for others (eg phenmedipham and chlorsulfuron) O_3 pollution would increase tolerance to herbicides. These interactions are likely to be of increasing significance as the O_3 concentration rises by the predicted 17-40% over the next 30 years.

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