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Ambient Ozone and Plant Health

To those disciplinarians, we add plant scientists who consider that surface level ozone (O_3) poses a critical threat and a challenging problem to present and future world food, fiber, and timber production and conservation of natural plant communities, including their species diversity. In 1840, the German scientist C. F. Schönbein (43) suggested the existence of an atmospheric constituent with an electrical odor freed in noticeable amounts during thunderstorms (lightning). He proposed the name ozone (O_3) for this substance. It was chemically proven to exist at the ground level by Houzeau in 1858 (19). Soon thereafter, measurements were commenced at more than 300 stations to determine relative atmospheric O_3 concentrations. One hundred years had elapsed since those first measurements before Richards and coworkers (39) showed in 1958 that O_3 was a constituent of smog causing foliar injury to grape in California. During the following year, Heggstad and Middleton (18) reported that O_3 was responsible for flecking on tobacco leaves in the eastern United States. Similarly, Daines et al. (7) identified O_3 as the predominant air pollutant affecting agriculture in New Jersey. Since those early investigations, it has become evident that O_3 is by far the most important air pollutant toxic to plants worldwide. Ozone causes foliar injury and reduces growth and yield in many agronomic and horticultural crops, deciduous trees, and conifers (26). As population, urban centers, and industries have grown, an increasing number of reports have appeared during the past 25

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“To the philosopher, the physician, the meteorologist, and the chemist, there is perhaps no subject more attractive than that of ozone” (12).

years regarding O_3 -induced foliar injury on sensitive plants in many countries. These include Australia, Austria, Belgium, Canada, France, Germany, Greece, India, Israel, Italy, Japan, Mexico, the Netherlands, Pakistan, Peoples Republic of China, Poland, Russia, Spain, Sweden, Switzerland, Taiwan, United Kingdom, and Ukraine.

Occurrence of Ground Level Ozone

Atmospheric ozone (O_3) is part and parcel of global climate change. Although ozone at the ground level is a “greenhouse gas,” it plays a minor role in regulating our air temperature, contributing only about 7% to the total warming effect (23). There is also a naturally occurring beneficial O_3 layer in the upper atmosphere (between 15 and 50 km above the surface) that strongly absorbs harmful ultraviolet radiation (about 210 to 290 nm: radiation <280 nm is UV-C and 280 to 315 nm is UV-B). In contrast, there are both natural and human-made sources of O_3 at ground level. Because of these natural sources, there is a background average O_3 concentration of roughly 20 to 30 nl/liter (ppb) everywhere (10). It is highly questionable whether there is any place on earth that has not been influenced by modern-day human activity, and therefore background values will vary with location. Natural sources consist of lightning during thunderstorms and downward intrusions of O_3 from the upper atmosphere.

The most important mechanism for the formation of O_3 at the earth surface, however, is driven by sunlight (photochemical cycle). The two key precursors (volatile hydrocarbons and the oxides of nitrogen) required in the reaction cycle are also produced by natural biological processes, although their contribution is much less important than that from fossil fuel combustion (10). In this context, because of the seasonal changes in the intensity of the sun, high concentrations of O_3 occur during the plant-growing season. Ozone is the major constituent of photochemical smog (a combination of smoke and fog), the oxidant complex that includes peroxy acyl compounds (e.g., peroxy acetyl nitrate [PAN]) and the oxides of nitrogen (10).

Figure 1 provides an idealized daily pattern of O_3 and its precursors in the photochemical reaction. Emissions of nitric oxide (NO) and hydrocarbons (HCs) are high in the early morning as a result of heavy urban traffic during the rush hour. These vehicular emissions destroy some of the O_3 present at that time, leading to the conversion of NO to NO_2 (nitrogen diox-

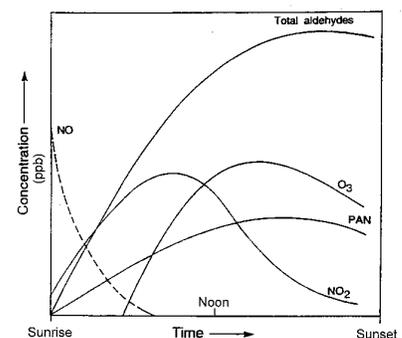


Fig. 1. Idealized diurnal profiles of the reactants (e.g., NO_2 , nitrogen dioxide, and total aldehydes or hydrocarbons) and products (e.g., O_3 and PAN, peroxy acetyl nitrate) in photochemical air pollution (Krupa [22]).

ide, one of the precursors for O₃). The NO₂ concentrations reach peak values when the atmosphere has developed sufficient capability to oxidize NO without totally consuming O₃. This process involves the sec-

ond precursor, the HCs and their oxidation products. Nitrogen dioxide is cleaved by sunlight to yield NO and atomic oxygen (O). The latter combines with molecular oxygen (O₂) to form O₃. In a typical urban

atmosphere, O₃ concentrations increase rapidly between 1200 and 1500 h, when the intensity of solar radiation is at a maximum and the NO₂:NO ratio is large. The rate of O₃ formation may then decline, reaching a steady state during the late afternoon to early evening hours. After that period, O₃ concentrations fall as NO₂ breakdown diminishes and as fresh emissions of NO deplete the O₃. This daily pattern is quite different at high elevations (in general, above approximately 1,500 m from the surface or above the so-called mixed layer of the atmosphere), where O₃ concentrations remain relatively steady through day and night. At that altitude, there is an O₃ reservoir, and destruction of that O₃ by the surface is insufficient to produce the type of daily patterns observed at lower elevations (Fig. 2).

During the growing season, stagnant air masses, varying in duration from one to several days, will result in high surface O₃ concentrations on a regional scale. Thus, crops and forests are exposed, for a few hours to days, to relatively high O₃ concentrations (e.g., >80 nl/liter), with other periods of relatively low concentrations (e.g., <40 nl/liter). The average lifetime of O₃ is about 16 h, and once produced, it can be transported long distances to rural agricultural and forested areas (Fig. 3). Thus surface level O₃ is an inter-regional (in the eastern United States, California) and even a continental (Europe) scale problem. Table 1 provides a comparison of maximum daily O₃ concentrations measured in different regions.

Toxicity to Plants

Plants are subjected to acute and chronic exposures of ground-level O₃. An acute exposure consists of relatively high O₃ concentrations (e.g., >80 nl/liter) from a few consecutive hours to days. In comparison, a chronic exposure consists of relatively low O₃ concentrations (e.g., <40 nl/liter) for the entire life of a plant, with periodic intermittent or random episodes of high concentrations. Both acute and chronic O₃ exposures can result in symptoms of foliar injury on sensitive plants (Table 2, Fig. 4A to D and 5A to D).

Plant response to O₃ varies with the genus, species, cultivar or variety, and geno-

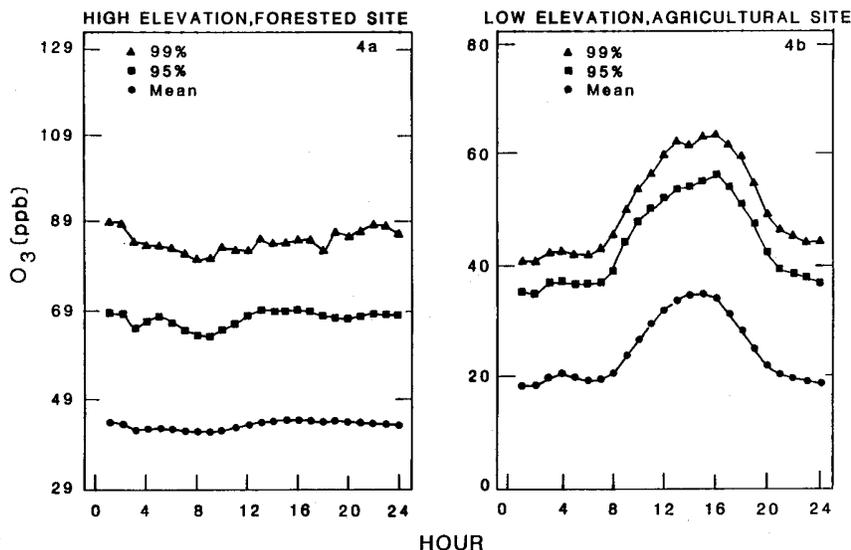


Fig. 2. Daily patterns of hourly mean, 95th and 99th percentile O₃ concentrations (10 m above the surface). Comparison of results from a higher elevation (2,103 m MSL) forested site to a lower elevation (1,098 m MSL), agricultural monitoring site. Note difference in scales of vertical axes, 1 ppb = 1 nl/liter or ~1.96 µg/m³ (Krupa and Kickert [24], with kind permission from Kluwer Academic Publishers).

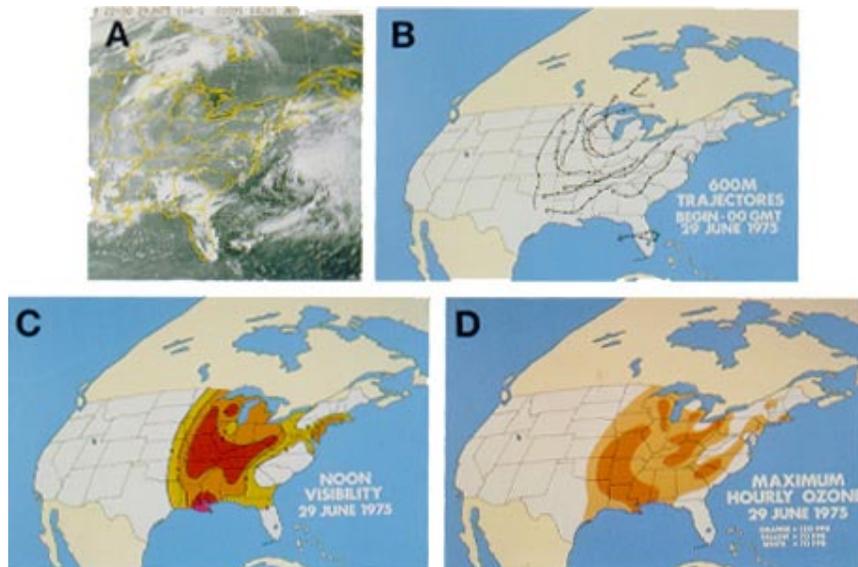


Fig. 3. A, Satellite photograph showing clockwise movement of pollutant clouds from northeast through central United States into eastern half of Minnesota on 29 June 1975. B, Meteorological model showing movement of pollution-containing air parcels across central US into Minnesota on 29 June 1975. Air parcel's path was determined with weather sounding data collected at 600 m above the ground at various locations in the US. Arrows indicate air parcel location at 12-h intervals, starting at 0.00 h Greenwich Mean Time (GMT). C, Contours showing number of miles of visibility (1 mile = 1.61 km) at various locations in eastern and central United States at noon on 29 June 1975. In the eastern half of Minnesota, visibility ranged from 4 to 8 miles (6.4 to 9.6 km). In the clear area to the northwest, visibility was >15 miles (>18 km). Microscopic sulfate particles in the air caused the reduction in visibility. D, Contours showing maximum hourly O₃ concentrations in eastern and central United States on 29 June 1975. In the eastern half of Minnesota, maximum hourly O₃ concentrations were >70 ppb (1 ppb = 1 nl/liter or ~1.96 µg/m³). During this episode, long range air pollutant transport brought O₃ and microscopic sulfate particles together into Minnesota. Two days later, sensitive plant species showed symptoms of O₃ injury (Krupa et al. [25]).

Table 1. Typical summertime daily maximum surface level O₃ concentrations^a

Region	O ₃ , nl/liter ^b
I. Urban-suburban	100-400
II. Rural	50-120
III. Remote tropical forest	20-40
IV. Remote marine	20-40

^a Source: Modified from National Research Council, US (34).

^b 1 nl/liter = 1 ppb or ~2 µg/m³.



Fig. 4. Ozone-induced symptoms of foliar injury: A, close-up view of a healthy tobacco (*Nicotiana tabacum*) leaf (right) and a leaf showing flecking (left); B, part of grape (*Vitis vinifera*) leaf showing purple stippling; C, pinto bean (*Phaseolus vulgaris*) leaf showing bifacial necrosis between larger veins; D, wheat (*Triticum aestivum*) leaf blade showing vertical, chlorotic streaks between larger veins (A, C, D, Krupa et al. [26], B, courtesy of the late F. A. Wood).

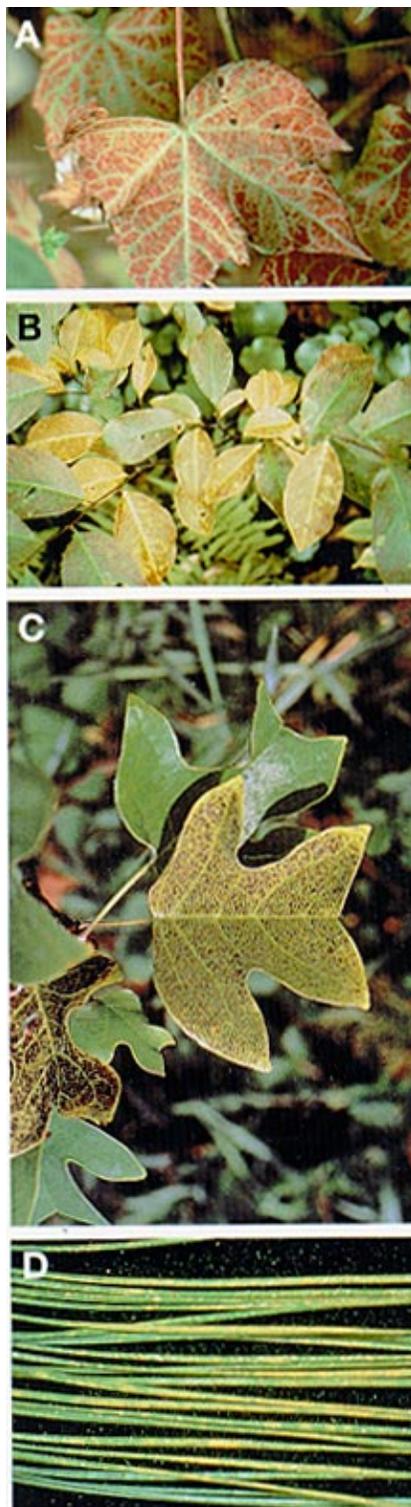


Fig. 5. Ozone-induced symptoms of foliar injury: A, close-up view of cotton (*Gossypium hirsutum*) leaf showing chronic response, premature senescence, and purple pigmentation in the interveinal areas; B, upper surface purple stippling on black cherry (*Prunus serotina*); C, yellow poplar (*Liriodendron tulipifera*) showing upper surface purple stippling; D, close-up view of chlorotic mottle on needles of Ponderosa pine (*Pinus ponderosa*) (Krupa et al. [26]).

type. Nevertheless, several plant species have been characterized for their development of O_3 injury symptoms and are used as biological indicators (Table 3) to assess relative O_3 exposures at various geographic locations. Among all indicator plants, tobacco (*Nicotiana tabacum* L.) cv. Bel-W3 (O_3 -sensitive) and cv. Bel-B (O_3 -tolerant) are the best described and the most commonly used worldwide. For example, some 20,000 residents voluntarily grew the two tobacco cultivars in pots, as part of a bio-monitoring network in the British Isles during the 1990s, to assist in mapping the geographic distribution of the occurrences of phytotoxic concentrations of O_3 in the United Kingdom (17). However, foliar injury cannot always be directly related to yield losses. Therefore, more recently, a white clover (*Trifolium repens* L.) indicator system (A. S. Heagle and J. E. Miller, USDA, ARS, North Carolina State University, Raleigh, <http://www.ncl.ac.uk/airweb/ozone/clover0.htm>), consisting of two specific clones that differ in their biomass responses (yield) to ambient levels of O_3 , is being used in the United States and Europe (Centre for Ecology and Hydrology, <http://icpvegetation.ceh.ac.uk>) to measure the relative effects. This is to indicate geographic areas where plants may suffer adverse growth and yield effects due to ambient O_3 stress, with or without foliar injury symptoms. These studies have shown, for example, that the biomass of the O_3 -sensitive clover clone can be reduced by 25% on Long Island, NY, and by at least 50% in California (16,29).

How Ozone Affects Plants

Ozone is deposited from the atmosphere onto plant canopies by diffusion and enters the leaf through stomata. Environmental, biological, and cultural (e.g., irrigation) factors that promote stomatal opening increase the risk of O_3 injury to plants (26). Ozone causes negative effects on a number of plant processes, including photosynthesis, water use efficiency, rate of senescence, dry matter production, flowering, pollen tube extension, and yield (22).

While many physiological functions necessary for growth and reproduction are impaired by O_3 , specific cellular sites that undergo damage are not completely known. For example, photosynthetic activity can be reduced by decreasing stomatal opening and restricting CO_2 uptake, by diminishing energy production in the photosystems, or by decreasing CO_2 assimilation. Although O_3 is known to cause stomatal closure, that effect appears to be secondary, since electrophysiological studies have demonstrated that O_3 affects stomatal opening only indirectly (46). The amount of active carboxylating (acceptance of CO_2) enzyme, RUBISCO (ribulose biphosphate carboxylase), can be reduced by O_3 (8). The effects on RUBISCO can be either by direct oxidation (36) or through

suppression of messenger RNA (ribonucleic acid) production (38). Chlorophyll fluorescence measurements have shown that O₃ can damage various components of the light-harvesting complex in the chloroplasts. In sensitive plants, foliar injury reduces the number of functional complexes, diminishing the plant's capability to utilize sunlight for photosynthesis (14). At lower concentrations, or in tolerant plants, O₃ can interfere with the production of electrons, limiting the energy available to "fix" CO₂ (15). Although O₃ can affect these various functions in the photosynthetic pathway, the sensitivities of the different components, the sequence of events occurring, and the specific sites of injury are yet to be identified. In addition to its effect on shoots, O₃ is known to adversely affect carbon flow to the roots and consequently their biology and biomass (Fig. 6).

How Some Plants Tolerate Ozone

It is generally accepted that O₃ does not persist in the intercellular spaces of the leaf. Rather, organic radicals and various reactive forms of oxygen are generated through O₃ decomposition and interactions with cell components (37). These oxidizing compounds damage proteins and membranes, leading to impaired physiological function and cell death. Acute O₃ injury in sensitive genotypes, usually seen as development of foliar lesions, resembles the hypersensitive response (HR) of plants to pathogen attack. An oxidative burst occurs as the initial reaction to both O₃ exposure and pathogen assault, and similar signal molecules have been implicated in induction of the HR and O₃ injury (40,44). In O₃-tolerant genotypes, either the oxidative burst is suppressed (45) or oxidative damage is highly localized (21), thereby restricting the extent of foliar lesions.

Since the signal pathways that mediate the response of plants to O₃ and pathogens appear to have some common features, our understanding of mechanisms that limit O₃ injury could benefit from studies on the molecular aspects of plant-pathogen inter-

actions (41). The plant antioxidant system, which scavenges naturally occurring reactive oxygen compounds, could function as a primary mechanism to alleviate the oxidative burden resulting from O₃ exposure. The ascorbate (vitamin C)-glutathione (an amino acid tripeptide) cycle has been the most intensively studied, and generally there is a positive correlation of O₃ tolerance with levels of antioxidants and antioxidant enzyme activities (5). For example, mutants that are deficient in vitamin C are very sensitive to O₃ (6), as are plants that have been genetically manipulated to produce less ascorbate peroxidase, an enzyme that uses vitamin C to detoxify hydrogen peroxide (35). Transgenic plants that have been engineered to overproduce ascorbate-glutathione antioxidant enzymes have pro-

vided mixed results regarding O₃ tolerance, depending upon the species, the cellular compartment in which the enzyme is expressed, and the isozyme (distinct forms of an enzyme, but with identical function) chosen (32).

Numerous other antioxidant compounds are found in plants, and their role in O₃ tolerance needs to be further explored. In addition to signaling processes and biochemical protective mechanisms, plants may express differential tolerance depending upon the rate of influx of O₃ into the leaf interior. Uptake may be affected by stomatal density or by guard cell response to oxidative conditions. It appears that O₃ does not directly affect stomatal closure (46), but acts indirectly, perhaps by influencing CO₂ "fixation" or altering hormone



Fig. 6. Left: radish (*Raphanus sativus*) plant grown in filtered air; right: plant grown in unfiltered air containing O₃ and other photochemical oxidants (Courtesy of H. E. Heggestad).

Table 2. Common symptoms of O₃-induced foliar injury^a

Acute injury	Chronic injury
Broad-leaved plants	
Bleaching (unifacial/upper surface or bifacial): small unpigmented necrotic spots or more general upper surface bleaching. Palisade cells and, when injury is more severe, upper epidermal cells collapse and become bleached.	Pigmentation (bronzing): leaves turn red-brown to brown as phenolic pigments accumulate.
Flecking: small necrotic areas due to death of palisade cells, metallic or brown, fading to tan, gray, or white.	Chlorosis: may result from nongreen pigmentation or may occur alone as chlorophyll breakdown.
Stippling: tiny punctate spots where a few palisade cells are dead or injured, may be white, black, red, or red-purple.	Premature senescence: early loss of leaves, flowers or fruit.
Bifacial necrosis: when the entire tissue through the leaf is killed, bifacial, dead areas develop ranging in color from white to dark orange-red. While small veins are usually killed along with the other tissue, larger veins frequently survive.	
Conifers	
Banding: clear bands of chlorotic tissue on semimature needle tissue following O ₃ episodes.	Flecking and mottling: flecking is the earliest symptom on the older needles of conifers. Mottling is generally associated with diffuse chlorotic areas interspersed with green tissue on first-year needles.
Tipburn: characterized by dying tips of young elongating needles. At first reddish brown in color, later turning brown, injury spreading from the tip downward.	Premature senescence: early loss of needles.

^a Source: Krupa et al. (26).

Table 3. List of some plant species that are sensitive to O₃ and used as indicator plants

Common name	Latin name	Common name	Latin name
Annual blue grass	<i>Poa annua</i>	Morning glory	<i>Pharbitis nil = Ipomoea</i> spp.
Bean	<i>Phaseolus vulgaris</i>		<i>Ipomoea purpurea</i>
Blackberry	<i>Rubus</i> spp.	Quaking aspen	<i>Populus tremuloides</i>
Black cherry	<i>Prunus serotina</i>	Sassafras	<i>Sassafras albidum</i>
Clover	<i>Trifolium repens</i>	Spinach	<i>Spinacea oleracea</i>
Green ash	<i>Fraxinus pennsylvanica</i>	Tobacco	<i>Nicotiana tabacum</i>
Milkweed	<i>Asclepias syriaca</i>	Yellow poplar	<i>Liriodendron tulipifera</i>
		White ash	<i>Fraxinus americana</i>

^a Source: Modified from Krupa et al. (26).

levels. The role of stomates in influencing O₃ tolerance is not fully characterized at present.

Ozone and Plant Disease

Although ozone can influence the development of plant diseases (28), the underlying processes are not well understood. In general, O₃ can decrease the incidence of diseases caused by obligate parasites, while increasing the problems associated with facultative parasites (Table 4). Ozone is unlikely to have direct effects on fungal pathogens; rather, the effects are host plant mediated. Conversely, occurrence of diseases can alter the foliar responses to O₃. Much of this knowledge is based on empirical field observations and experimental studies conducted in controlled environment, greenhouse, or field exposure

chambers. There is no information on the joint effects of O₃ and disease on plant growth and yield.

Effect on Crops and Trees

Numerous investigators have shown that chronic, whole growth season or whole life cycle exposures to O₃ can result in losses of marketable yield in crops (Fig. 7A and B) and reductions in growth and productivity of tree species (47). The chronic effects of O₃ on tree populations have consequences to aesthetic and recreational aspects of our national forests and national parks and are of much concern to governmental agencies. In this context, the forests of the San Bernardino Mountains of Southern California serve as a classic illustration of the long-term impact of ambient O₃ on a forest ecosystem (31) (Fig. 8).

Reductions in growth and yield can occur with or without symptoms of O₃-induced foliar injury. Nevertheless, based on numerous experimental studies, it has been estimated that ambient O₃ exposure causes \$0.8 billion annually in crop loss in the United States (47). A similar estimate is not available for tree species, but the adverse effects of O₃ on our national parks and forests are not in doubt (4,30).

Alteration of Plant Community Structure and Biodiversity

Several plant species in U.S. national parks are known to exhibit foliar injury to ambient O₃ exposure. For example, in Acadia National Park in Maine, black cherry, quaking aspen, white ash (*Fraxinus americana* L.), jack pine (*Pinus banksiana* Lamb.), big-leaf aster (*Aster macrophyllus* L.), and spreading dogbane (*Apocynum androsaemifolium* L.) are sensitive. Similarly, widespread O₃-induced foliar injury has been observed on the native black cherry and tall milkweed (*Asclepias exaltata* L.) in the Great Smoky Mountains National Park. How these foliar responses translate to other issues such as competitive sustainability of a given species within the native plant communities are not fully understood at the present time.

Nevertheless, tests conducted to determine the effects of O₃ on the competitive relationships between native plants showed, for example, that the rate of blackberry:broom-sedge (*Andropogon virginicus* L.) litter decomposition was reduced with increasing O₃ concentrations, with implications for altering nutrient cycling and biological diversity (2).

One aspect of assessing biological diversity is species fitness or reproductive capacity. For example, experimental O₃ exposures reduced growth and flowering of

Table 4. Some examples of the effects of O₃ on plant diseases^a

Pathogen	Host plant	Effect on disease ^b
Fungi - Obligate biotrophs		
<i>Puccinia coronata</i>	Oats	Reduced growth of uredia
<i>Puccinia graminis</i> f. sp. <i>tritici</i>	Wheat	Decreased hyphal growth and uredio-spore production on O ₃ -injured leaves
<i>Erysiphe graminis</i> f. sp. <i>hordei</i>	Barley	Reduced rate of infection if exposed to sufficient O ₃ during incubation; enhanced colony size when infection is established
Fungi - Necrotrophs		
<i>Drechslera sorokiniana</i>	Wheat	Increased percentage of diseased leaf area
<i>Botrytis cinerea</i>	Potato	Infection only on O ₃ injured leaves
<i>Lophodermium</i> sp.	Pine	Increased severity of needle blight
Bacteria		
<i>Pseudomonas glycinea</i>	Soybean	Reduced number of lesions
<i>Xanthomonas alfalfae</i>	Alfalfa	Reduced severity of bacterial infection
<i>Xanthomonas fragariae</i>	Wild strawberry	Reduced number of lesions

^a Source: Modified from Manning and von Tiedemann (28).

^b Observed effect on disease was dependent upon whether plants were exposed to sufficient dose of O₃ before or after inoculation with the pathogen.

Characterization and Mechanisms of Plant Responses to O₃

The agricultural experiment stations of the land grant universities have supported O₃ exposure-plant effects research through NE (northeast)-176 and its predecessors, representing the longest multistate or institutional research and outreach project in the United States. Although the U.S. Environmental Protection Agency (EPA) has maintained large-scale, in-house and cooperative research programs with other governmental agencies, NE-176 represents the best-sustained effort through continued commitment of the agricultural experiment stations.

This multistate project began as Northeast Regional Project 56 (NE-56) in 1966, with nine states (extending from Maine to North Carolina and the Mid-Atlantic region) participating (20). The principal aim of the project was to measure atmospheric oxidant (O₃) concentrations at different locations within the region during the growing season and observe foliar injury to indicator plants (tobacco sensitive cv. Bel-W3 and tolerant cv. Bel-B). This was to determine the frequency of occurrence and distribution of oxidants and phytotoxicity of the atmosphere. By the early 1970s, it was well accepted that ambient O₃ was an inter-regional problem of concern for crop and forest productivity. Thus in 1972, after reaching its initial goal, NE-56 ceased to function. Subsequently project scientists expanded their objectives beyond the use of indicator plants to include the effects of O₃ on plant physiological processes, growth, and yield. After a series of 5-year cycles of study, the project was designated NE-176 in 1990.

Current research activities in NE-176 include studies on the biochemical and molecular mechanisms of O₃ injury and sensitivity or tolerance in plants; impact of O₃ on carbon allocation, carbon/nitrogen relationships, and secondary metabolism as they affect pathogen and pest relations, decomposition rates, and nutrient cycling; joint effects of O₃ with other factors influencing growth and yield, such as increasing CO₂ concentrations, and changes in temperature and water availability; yield responses of crops and growth responses of trees, and effects of O₃ on plant community structure and species diversity. Additional research activities relate to modeling cause-effect relationships, including ecosystem processes. Overall, these efforts are continuing to serve target audiences ranging from individual homeowners to large industrial corporations and state and federal governmental agencies (Table 5).

the butterfly bush (*Buddleia* spp.) (9). Similarly, exposures to O₃ reduced flowers and fruits in spreading dogbane. A very important finding here was that foliar injury was not necessarily required to elicit negative effects on sexual reproduction and thus changes in biological diversity (3).

A Look to the Future

Ozone is a "criteria air pollutant." This designation is based on the fact that there are documented adverse effects of O₃ on people, plants, or materials at concentrations, or approaching those, found in polluted air. There are air quality standards (U.S.), air quality objectives (Canada), and critical levels (Commission of the European Communities) for O₃ to protect vegetation against adverse effects (27). In the United States, Congressional mandate requires the Environmental Protection Agency (EPA) to review all available scientific literature and revise air quality standards where required, for approval by the Congress, once every 5 years. At present, the tentative O₃ standard in the United States is an 8-h running average of 80 nl/liter, not to be exceeded more than twice in three consecutive years. This standard is tentative because of a pending decision regarding court challenges by industry against the EPA's legislative authority within the Clean Air Act of the U.S. Congress (American Trucking Associations, ATA Inc. versus EPA). This case is now in the Supreme Court.

Leaving aside the judicial controversy, much of our knowledge of the effects of O₃

on plants is derived from controlled environment or field chamber exposure studies. Plants in the ambient environment do not grow in chambers. Although chamber studies have the value of providing us with a basic understanding of cause and effect, results from such studies cannot be directly extrapolated to the chamberless ambient environment, where cost-benefit policies are involved. There is a significant need to conduct chamberless field studies to determine the effects of ambient levels of O₃ on plant growth, productivity, and species fitness, in the context of biological diversity. The emphasis has been on univariate studies (O₃ only as the cause of an effect), but there is a need to address the O₃ issue in the context of the presence of other air pollutants (47) and the incidence of pathogens (28,47) and insect pests (11,47). The resulting joint effects can be additive, more than additive, or less than additive (47). Because of its complexity, no studies have directly addressed this overall question. The subject becomes much more complex when we try to integrate O₃ and climate change (increasing CO₂ concentrations, changes in temperature and precipitation patterns, etc.). This holistic research is a prime target for plant disease epidemiologists. In all of these cases, the limiting factor is the integration of the science spanning multiple disciplines.

During the last decades, as urban centers and consequent highway traffic have continued to grow in the United States and elsewhere, so have the ground level O₃ concentrations. The National Research

Council published an authoritative document in 1992 called "Rethinking the ozone problem in urban and regional air pollution" (34). Most recently, a number of state-of-the-science papers have been published (33). Alternative transportation strategies such as mass transit and the use of fuel other than gasoline (a major source of O₃ precursor pollutants) in automobiles may provide some relief in the United

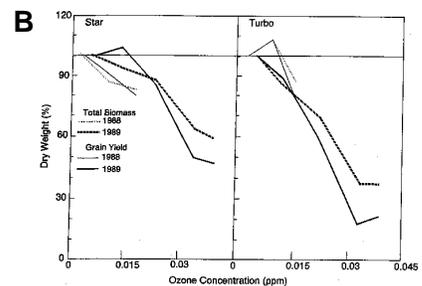


Fig. 7. A, Differential biomass responses of tolerant (left) and sensitive (right) white clover (*Trifolium repens*) clones to ambient O₃ exposures in Massachusetts (Courtesy of W. J. Manning); B, yield responses of two different cultivars of wheat subjected to chronic O₃ exposure (Adaros et al. [1]).

Table 5. NE-176: Characterization and mechanisms of plant responses to ozone (O₃) in the northeastern United States

Participating institution ^a	Target audience	Type of outcome
1. NE agricultural experiment stations: MA, University of Massachusetts; MD, University of Maryland; NJ, Rutgers University; NY, Cornell University/Long Island; PA, Pennsylvania State University.	(a) EPA; (b) NASA; (c) managers of national parks, class I wilderness areas and forested land; (d) managers of urban public park and recreational lands; (e) county extension & outreach educators; (f) home and master gardeners; (g) arborists, landscape, and nursery managers; (h) private plant disease and pest consultants;	(a) Application of the results in developing federal air quality standards to protect crops, forests and native plants; (b) breeding tolerant crop cultivars for seed production and use; (c) identification of O ₃ tolerant materials for use in gardens, urban landscapes and parks; (d) increased environmental awareness through outreach; (e) improved management of national parks and protected ecosystems; (f) early detection of ecosystems under O ₃ stress for developing mitigation policies; (g) sharing knowledge through international education and cooperative research.
2. Other agricultural experiment stations: AL, Auburn University; CA, University of California-Riverside; MN, University of Minnesota; VA, Virginia Polytechnic Institute & State University.	(i) regional energy and chemical industry; (j) provincial environmental regulators; (k) scientists and environmental regulators in developing countries (e.g., Mexico, Eastern Block countries), including undergraduate and graduate students and faculty members in academe; (l) environmental conservation groups; (m) crop breeders and (n) seed companies.	
3. Other institutions: CA, USFS, Riverside; MD, USDA/ARS-BARC, Beltsville; NC, USDA/ARS-North Carolina State University; NY, Boyce Thompson Institute at Cornell University; OR, US EPA.		

^a Additional institutions joining are: IA - Iowa State University, Ames; NB, Canada - CFS, Fredericton; and NC - Appalachian State University, Boone.



Fig. 8. Stressed conifer ecosystem of the San Bernardino National Forest, CA, exposed to smog (O₃) from Los Angeles (Courtesy of P. R. Miller).

States and other developed countries, perhaps some 20 years hence (42). Nevertheless, O₃ is and will continue to be a growing problem in developing countries. Already Mexico City, New Delhi, and Beijing are among urban centers generating significant ground level O₃ concentrations. Hemispheric O₃ problems are being created by forest fires and burning clear cut for land reclamation in Central and South America and Southeast Asia. Forest fires and biomass burning result in emissions of the precursor compounds for O₃ formation.

Our current understanding of the effects of O₃ on plants is almost entirely based on studies conducted with temperate species and environmental conditions. We have very little knowledge of crop and forest responses in the Southern Hemisphere. This is also the part of the world where the rate of population growth has been the most between 1960 and 1990 (13). Thus, given the instability of sociopolitical and economic issues, sustaining global populations will require a global perspective of

ambient O₃, air quality in general, including climate change (O₃ is a part of that process), and food, fiber, and timber production.

Acknowledgments

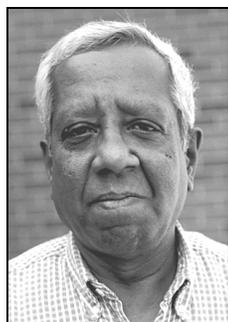
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Literature Cited

1. Adaros, G., Weigel, H. J., and Jäger, H.-J. 1991. Die Wirkung von ozon auf wachstums- und Ertragsparameter zweier Sommerweizensorten (*Triticum aestivum* L.). Z. Pflanzenkrankh. Pflanzenschutz 98:113-124.
2. Barbo, D. N., Chappelka, A. H., Somers, G. L., Miller-Goodman, M. S., and Stolte, K. 1998. Diversity of an early successional plant community as influenced by ozone. New Phytol. 138:653-662.
3. Bergweiler, C. J., and Manning, W. J. 1999. Inhibition of flowering and reproductive suc-

cess in spreading dogbane (*Apocynum androsaemifolium*) by exposure to ambient ozone. Environ. Pollut. 105:333-339.

4. Chappelka, A. H., and Samuelson, L. J. 1998. Ambient ozone effects on forest trees of the eastern United States: A review. New Phytol. 139: 91-108.
5. Conklin, P. L., and Last, R. L. 1995. Differential accumulation of antioxidant mRNAs in *Arabidopsis thaliana* exposed to ozone. Plant Physiol. 109:203-212.
6. Conklin, P. L., Williams, E. H., and Last, R. L. 1996. Environmental stress sensitivity of an ascorbic acid-deficient *Arabidopsis* mutant. Proc. Natl. Acad. Sci. USA 93:9970-9974.
7. Daines, R. H., Leone, I. A., and Brennan, E. G. 1960. Air pollution as it affects agriculture in New Jersey. Bull. 794. New Jersey Agricultural Experiment Station, 1960. Rutgers, NJ, p. 14.
8. Enyedi, A. J., Eckardt, N. A., and Pell, E. J. 1992. Activity of ribulose biphosphate carboxylase/oxygenase from potato cultivars with differential response to ozone stress. New Phytol. 122:493-500.
9. Findley, D. L., Keever, G. J., Chappelka, A. H., Gilliam, C. H., and Eakes, D. J. 1997. Differential response of buddleia (*Buddleia davidii* Franch.) to ozone. Environ. Pollut.



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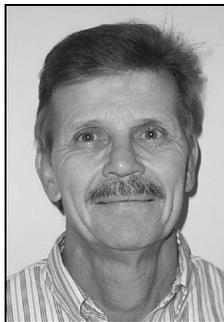
- 98:105-111.
10. Finlayson-Pitts, B. J., and Pitts, J. N. 1999. Chemistry of the Upper and Lower Atmosphere. Academic Press, New York.
 11. Flückiger, W., Braun, S., and Bolsinger, M. 1988. Air pollution: Effect on host plant-insect relationships. Pages 366-380 in: Air Pollution and Plant Metabolism. S. Schulte-Hostede, N. M. Darrall, L. W. Blank, and A. R. Wellburn, eds. Elsevier Applied Science, London.
 12. Fox, C. B. 1873. Ozone and Antozone. J. & A. Churchill, London.
 13. Gommers, R. 1993. Current climate and population constraints on world agriculture. Pages 67-86 in: Agricultural Dimensions of Global Climate Change. H. M. Kaiser and T. E. Drennen, eds. St. Lucie Press, Delray Beach, FL.
 14. Guidi, L., Di Cagno, R., and Soldatini, G. F. 2000. Screening of bean cultivars for their response to ozone as evaluated by visible symptoms and leaf chlorophyll fluorescence. Environ. Pollut. 107:349-355.
 15. Guidi, L., Nali, C., Ciompi, S., Lorenzini, G., and Soldatini, G. F. 1997. The use of chlorophyll fluorescence and leaf gas exchange as methods for studying the different responses to ozone of two bean cultivars. J. Exp. Bot. 48:173-179.
 16. Heagle, A. S., Miller, J. E., Chevone, B. I., Dreschel, T. W., Manning, W. J., McCool, P. M., Morrison, C. L., Neely, G. E., and Rebeck, J. 1995. Response of a white clover indicator system to tropospheric ozone at eight locations in the United States. Water Air Soil Pollut. 85:1373-1378.
 17. Heggestad, H. E. 1991. Origin of Bel-W3, Bel-C and Bel-B tobacco varieties and their use as indicators of ozone. Environ. Pollut. 74:264-291.
 18. Heggestad, H. E., and Middleton, J. T. 1959. Ozone in high concentrations as a cause of tobacco leaf injury. Science 129:208-210.
 19. Houzeau, A. 1858. Preuve de la présence dans l'atmosphère d'un nouveau principe gazeux, l'oxygène naissant. C.R. Acad. Sci., Paris 46: 89-91.
 20. Jacobson, J. S., and Feder, W. A. 1974. A Regional Network for Environmental Monitoring: Atmospheric Oxidant Concentrations and Foliar Injury to Tobacco Indicator Plants in the Eastern United States. Bull. 604. Massachusetts Agricultural Experiment Station, Amherst.
 21. Koch, J. R., Creelman, R. A., Eshita, S. M., Seskar, M., Mullet, J. E., and Davis, K. R. 2000. Ozone sensitivity in hybrid poplar correlates with insensitivity to both salicylic acid and jasmonic acid. The role of programmed cell death in lesion formation. Plant Physiol. 123:487-496.
 22. Krupa, S. V. 1997. Air Pollution, People and Plants. American Phytopathological Society, St. Paul, MN.
 23. Krupa, S. V. 1997. Global climate change: Processes and products - An overview. Environ. Monitor. Assess. 46:73-88.
 24. Krupa, S. V., and Kickert, R. N. The greenhouse effect: The impacts of carbon dioxide (CO₂), ultraviolet-B (UV-B) radiation and ozone (O₃) on vegetation (crops). Vegetatio 104/105:223-238.
 25. Krupa, S. V., Pratt, G. C., and Teng, P. S. 1982. Air pollution: An important issue in plant health. Plant Dis. 66:429-434.
 26. Krupa, S. V., Tonnejck, A. E. G., and Manning, W. J. 1998. Ozone. Pages 2-1 to 2-28 in: Recognition of Air Pollution Injury to Vegetation: A Pictorial Atlas. R. B. Flagler, ed. Air & Waste Management Association, Pittsburgh, PA.
 27. Legge, A. H., Grünhage, L., Nosal, M., Jäger, H.-J., and Krupa, S. V. 1995. Ambient ozone and adverse crop response: An evaluation of North American and European data as they relate to exposure indices and critical levels. J. Appl. Bot. (Angew. Bot.) 69:192-205.
 28. Manning, W. J., and von Tiedemann, A. 1995. Climate change: Potential effects of increased



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Dr. Chappelka is a professor in the School of Forestry & Wildlife Sciences at Auburn University. He received his Ph.D. in plant pathology from Virginia Tech in 1986. Since 1987, Dr. Chappelka has been on the staff within the school investigating the responses of forest trees and associated plant species to air pollutants, primarily O₃ and acid rain. His primary interests are in studying the effects of these pollutants on the structure and function of terrestrial ecosystems.

Dr. Chevone is an associate professor in the Department of Plant Pathology, Physiology, and Weed Science at Virginia Tech. He received his Ph.D. from the University of Minnesota in 1974. For the past 25 years, he has been studying air pollutant effects on terrestrial plants. His primary interest is characterizing physiological and biochemical mechanisms involved in O₃ tolerance and sensitivity of crop species and forest trees.

Dr. Pell was appointed vice president for research and dean of the graduate school at Penn State University in January 2000. Prior to this, she had a joint appointment in the Environmental Resources Research Institute and Department of Plant Pathology and still serves as the John and Nancy Steimer professor of agricultural sciences. Dr. Pell earned a B.S. in biology from City College of the City of New York, and a Ph.D. in plant biology from Rutgers University. She was appointed as an assistant professor of plant pathology at Penn State in 1973. She was promoted to associate professor in 1978 and professor of plant pathology in 1983. In 1991, she was named distinguished professor of plant pathology, and in 1995 was named the Steimer professor of agricultural sciences. Dr. Pell's research has focused on physiological, biochemical, and molecular aspects of O₃ effects on vegetation. In particular, she has developed a framework for understanding the mechanism by which O₃ induces accelerated leaf senescence and the implications of this response to the whole plant.

Dr. Zilinskas is a professor at Rutgers University in the Department of Plant Science. She received her Ph.D. in plant physiology in 1975 from the University of Illinois in Champaign/Urbana. She was a postdoctoral fellow at the Smithsonian Institution's Radiation Biology Laboratory for 1 year before joining the faculty at Rutgers in the Department of Biochemistry and Microbiology. She received tenure in 1980 and was promoted to professor in 1987. She joined the Department of Plant Science in 1994. Dr. Zilinskas' research program has been focused in the last 10 years on antioxidant enzymes and their role in oxidative stress tolerance in plants.

- atmospheric carbon dioxide (CO₂), ozone (O₃) and ultraviolet-B (UV-B) radiation on plant diseases. *Environ. Pollut.* 88:219-246.
29. McGrath, M. T. 2000. Impact of ambient ozone on clover at Long Island, New York. (Abstr.) *Phytopathology* 90:S50.
 30. McLaughlin, S., and Percy, K. 1999. Forest health in North America: Some perspectives on actual and potential roles of climate and air pollution. *Water Air Soil Pollut.* 116:151-197.
 31. Miller, P. R., and McBride, J. R., eds. 1998. *Oxidant Air Pollution Impacts in the Montane Forests of Southern California*. Springer-Verlag, New York.
 32. Mullineaux, P., and Creissen, G. 1999. Manipulating oxidative stress responses using transgenic plants: Successes and dangers. Pages 525-532 in: *Plant Biotechnology and In Vitro Biology in the 21st Century*. A. Altman, M. Ziv, and S. Izhar, eds. Kluwer Academic Publishers, Amsterdam.
 33. NARSTO (North American Research Strategy for Tropospheric Ozone). 2000. *The Narsto Ozone Assessment - Critical Reviews*. *Atmos. Environ.* 34:1853-2332.
 34. National Research Council, US. 1992. *Rethinking the Ozone Problem in Urban and Regional Air Pollution*. National Academy Press, Washington, DC.
 35. Orvar, B. L., and Ellis, B. E. 1997. Transgenic tobacco plants expressing antisense RNA for cytosolic ascorbate peroxidase show increased susceptibility to ozone injury. *Plant J.* 11:1297-1306.
 36. Pell, E. J., Eckardt, N., and Glick, R. E. 1994. Biochemical and molecular basis for the impairment of photosynthetic potential. *Photosyn. Res.* 39:453-462.
 37. Pell, E. J., Schlagnhauser, C. D., and Arteca, R. N. 1997. Ozone induced oxidant stress: Mechanisms of action and reaction. *Physiol. Plant.* 100:264-273.
 38. Reddy, G. N., Aeteca, R. N., Dai, Y. R., Flores, H. E., Negram, F. B., and Pell, E. J. 1993. Changes in ethylene and polyamines in relation to mRNA levels of the large and small subunits of ribululose biphosphate/oxygenase in ozone-stressed potato foliage. *Plant Cell Environ.* 120:819-826.
 39. Richards, B. L., Middleton, J. T., and Hewitt, W. B. 1958. Air pollution with relation to agronomic crops. V. Oxidant stipple of grape. *Agron. J.* 50:559-561.
 40. Sandermann, H. 1998. Ozone: An air pollutant acting as a plant signalling molecule. *Naturwissenschaften* 85:369-375.
 41. Sandermann, H., Ernst, D., Heller, W., and Langebartels, C. 1998. Ozone: An abiotic elicitor of plant defense reactions. *Trends Plant Sci.* 3:47-50.
 42. Sawyer, R. F., Harley, R. A., Cadle, S. H., Norbeck, J. M., Slott, R., and Bravo, H. A. 2000. Mobile sources critical review: 1998 NARSTO assessment. *Atmos. Environ.* 34:2161-2181.
 43. Schönbein, C. F. 1840. Recherches sur la nature d'l odeur qui se manifeste dans certaines actions chimique. *C.R. Acad. Sci. Paris* 10:706-710.
 44. Schraudner, M., Langebartels, C., and Sandermann, H. 1997. Changes in the biochemical status of plant cells induced by the environmental pollutant ozone. *Physiol. Plant.* 100:274-280.
 45. Schraudner, M., Moeder, W., Wiese, C., Van Camp, W., Inze, D., Langebartels, C., and Sandermann, H. 1998. Ozone-induced oxidative burst in the ozone biomonitor plant, tobacco Bel W3. *Plant J.* 16:235-245.
 46. Torsethaugen, G., Pell, E. J., and Assmann, S. M. 1999. Ozone inhibits guard cell K⁺ channels implicated in stomatal opening. *Proc. Natl. Acad. Sci. USA* 96:13577-13582.
 47. U.S. Environmental Protection Agency. 1996. *Air Quality Criteria for Ozone and Related Photochemical Oxidants, Vol II*. Office of Research and Development, Washington, DC.