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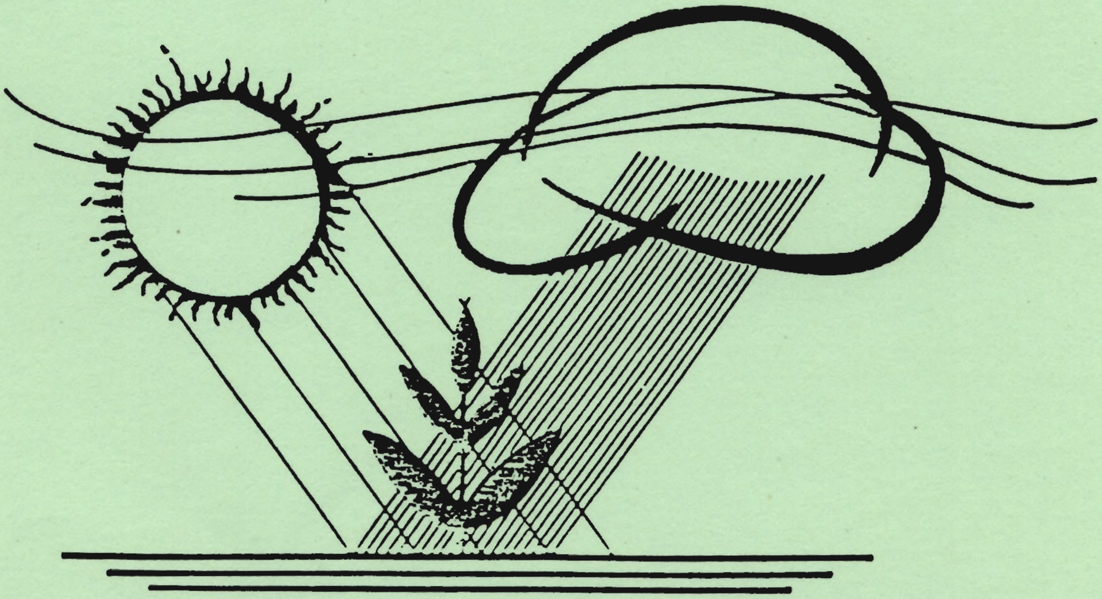

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CALIFORNIA CHAPTER

AMERICAN SOCIETY OF AGRONOMY

February 4-5, 1987

Clarion Hotel—Ontario Airport

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METHODS FOR ASSESSING CROP YIELD LOSSES FOR AIR POLLUTANTS

Robert C. Musselman
Associate Research Plant Physiologist
Statewide Air Pollution Research Center
University of California
Riverside, California 92521

Determination of crop yield reductions from air pollutants is necessary for economic assessment of effects of ambient pollutants on agricultural production. Early experiments measured yield of crops grown in ambient air compared to yield in clean air. However, data from these experiments is inadequate for predicting losses from other levels of pollutants which might occur. Most experiments now use several treatment levels of pollutants, and use regression or similar analyses to derive predictive functions of crop yield loss based upon pollutant dose to which the crop is exposed during growth.

Several methods have been available for experimental determination of yield under multiple pollutant dose levels. One of the earliest methods was to measure yield of crops in plots from several areas of a specific geographic region having different pollutant dose exposure levels, but similar environmental and cultural growing conditions. This ambient gradient method has been useful in areas where the pollutant level varies greatly in a relatively small geographic zone, such as near large urban centers or other point sources of pollutants. However, environmental differences between sites affecting growth and yield often confound the analysis.

More recent methods to determine crop yield response to pollutants involve use of field fumigation systems located at one site to remove variability in response due to environmental differences. Crops are grown in air which has been filtered to remove specific amounts of pollutant, or alternatively, to which a specific amount of pollutant has been artificially added. Several levels of pollutant are included in an experiment, with plants in each treatment exposed to a different pollutant dose throughout the experiment. Yield is therefore related to pollutant level to which the plants were exposed during growth. As with the field ambient gradient plots, a yield loss function can be generated to predict yield at any level of pollutant to which the crop might be exposed.

There are several field systems currently being used for air pollutant, crop yield loss analysis. These range from closed-top environmental chambers located in field plots, to open-top field chambers, to non-chamber fumigation grids. Each has its own advantages and disadvantages. Closed-top chambers offer excellent control and monitoring of environmental and pollutant levels. Plants growing in these chambers can be exposed to precise levels of a pollutant. However, a closed-top chamber environment is often different from ambient. Plants grown in closed-top chambers are exposed to less light and higher temperatures during their growth, and relative humidity is often higher within the chambers. Nevertheless, they have been useful for crop loss analyses where pollutant levels need to be precisely controlled and monitored.

The most commonly used chamber system for air pollutant crop yield loss assessment is the open-top chamber. Open-top chambers are placed over field plots and air containing a specific level of pollutant is delivered to the bottom of the chamber where it passes over the plant and out the top of the chamber. The open top allows for more natural environmental conditions, including light and temperature closer to ambient. However, open-top chambers do affect plant growth causing slightly higher temperatures and lower light intensity at canopy level. In addition, pollutant levels are more difficult to control and monitor, since downdrafts of ambient air often occur. Open-top chambers are relatively simple to construct and easy to move in the field. They have been successfully used for a number of years for crop yield loss assessment for pollutants.

Non-chamber fumigation systems involve a series of tubes or pipes placed throughout a field plot from which pollutants can be added to ambient air. These systems have also been used to exclude ambient air by directing filtered air over plant canopies. Their use in areas where ambient pollutants are high is limited, since the lower pollutant treatment levels are difficult to obtain or maintain in these areas. With non-chamber systems, growing conditions are near ambient, eliminating the confounding chamber effect on plant growth. Disadvantages include the extreme difficulty in controlling and monitoring pollutant levels to which the plants are exposed. Pollutant levels are highly dependent upon wind direction and intensity, and distance from the emitting source. This problem can be minimized with sophisticated computer control of emission of the pollutant from the grid of pipes or tubes. The control system is dependent upon feedback from pollutant monitoring at the plant canopy to the fumigation control system. Rapid changes in wind speed and/or direction and speed of sampling and control of fumigation affect the success of this system. These non-chamber systems have been used in natural ecosystems, and have been used to produce a gradient of pollutant to plots located at various distances from the point of emission.

Other methods have been suggested to estimate yield loss from air pollutant. One proposal has been to use chemical anti-oxidants to scavenge pollutants before they can attack plant cells. However, determining dose to which the plant is exposed is difficult using this technique. Another method which might be used to determine crop yield losses from pollutants would involve expansion of the ambient gradant technique. This would require analysis of crop production data from several geographic regions, using a factor analysis to relate yields to environmental, cultural and ambient pollutant data. Large data bases and computational facilities are necessary for this type of analysis.

Conclusions

Several methods are available for assessing yield losses from air pollutants. Multiple treatment levels of the pollutant are necessary to derive predictive crop loss functions. Systems available for delivering multiple pollutant treatment levels to experimental plots include locating plots in an ambient pollutant geographic gradient, or delivering several levels of a pollutant to different plots at one site. Pollutants can be delivered to plots at one site using closed-top chambers, open-top chambers, or non-chamber tube or pipe grids. Pollutant levels become easier to control and accuracy of monitoring increases from non-chamber to open-top chamber to closed-top chamber. However, variation from natural growth environment increases with amount of plant enclosure. Experimental objectives will determine which technique to use for individual studies, since all of these methods have been successfully used for air pollutant crop yield loss analysis.

RESPONSE OF CALIFORNIA CROPS TO ACIDIC DEPOSITION

Carl A. Fox
Senior Research Scientist
Southern California Edison Company
Rosemead, California

Deposition of wet (e.g., rain, fog, mist) and dry (e.g., gases, aerosols, particles) acidic materials is currently occurring over much of the major crop growing regions of the United States, including California. While no widespread damage to California's crops has been observed, acidities (pH) of rain and fog have been measured for single events as low as 2.9 and 1.69, respectively (South Coast Air Quality Management District, 1984). Annual mean pH values, however, are generally much greater (e.g., rain pH > 5.00) (California Air Resources Board (CARB), 1985).

Numerous studies have examined the effects of acid deposition on crops over the last decade. These studies have generally utilized simulated acid rain in controlled environment (e.g., growth chamber, greenhouse) or field exposures to determine crop response. Observed direct effects of simulated acid rain include: necrotic injury to leaves; premature leaf abscission; increased foliar leaching; erosion of leaf cuticle and epithelial waxes; decreased photosynthesis; decreased pollin viability; and increased, decreased, or no significant effect on crop yield (Council for Agricultural Science and Technology (CAST), 1984). Indirect responses include: increased susceptibility to drought and other environmental stresses; altered plant (host)-parasite interactions; and inhibition of nitrogen fixing bacteria in agricultural soils. Detrimental effects on crops via changes in soil properties are unlikely in agricultural systems due to natural soil processes and management practices such as fertilization, crop harvesting, and liming (Hidy et al., 1986).

Crop Yield Response

The average annual yield of crops is determined by the action and interaction of natural and manmade factors. Drought, insects, disease, and numerous other factors can decrease yields, while fertilization, irrigation, and genetic improvements can increase crop production (Table 1). Studies examining the specific yield response of crops to acid rain have been summarized by Irving (1983). Of the 14 crop cultivars studied in field experiments and 34 examined in controlled environments, the majority exhibited no response to simulated rain having ambient or higher levels of acidity (i.e., pH 3.9 - 4.5). Among the field studies, three crop species (garden beet, mustard greens, "Pioneer" field corn) were negatively affected, three responded positively (radish, alfalfa, fescue), and eleven cultivars showed no yield effects. In general, detrimental effects on crops from exposure to simulated acid rain do not occur unless pH values are less than 3.0 to 3.5 (CAST, 1984).

Table 1. Effect of natural and anthropogenic factors on crop yield in the U.S. for corn, wheat, and soybeans (Irving, 1985).

Source	Factor	Annual Effect On Actual Yield (%)	Annual Effect On Attainable Yield (%)
Natural			
	Drought and other physico-chemical factors		-72.0
	Disease	-16.8	
	Insects	-11.0	
	Weeds		-2.7
Anthropogenic			
	Ozone	-5.8	
	Fertilization	+50.0	
	Genetic Improvement	+40.0	

The yield response of California crops to acid rain has not been determined in any systematic manner. However, numerous studies have included crops commonly grown in California (Table 2). These studies show that most crops are not affected after exposure to simulated acid rain at pH values as low as 3.0. Carrots may be one exception as Cohen et al. (1981) and Lee et al. (1981) have reported market yield losses of 27%, 45%, and 44% after exposure to simulated rain with pH values of 4.0, 3.5, and 3.0, respectively. However, these results have not been demonstrated in field studies.

Many economically important California crops (e.g., cotton, grapes, nursery products) and specific cultivars have not been evaluated in terms of their response to acid rain. It has been established that crop cultivars can have differing sensitivities to acid deposition, thus extrapolation of data from one cultivar to predict the response of the entire species cannot entirely be justified (Irving, 1985). Nevertheless, the apparent "no effect" observed over a wide variety of crops coupled with known plant processes and structural characteristics which can exclude or neutralize acid deposition suggest that crop yield losses due to acid rain in California are small compared to losses from natural factors such as drought and disease.

Acid Fog Effects

Recent evidence suggests that acid fog may be of greater concern to crops than acid rain, particularly in southern and central California. Fog and cloud water acidities are typically greater than rain and often in the range of pH 2.2 - 4.0 (Brewer et al., 1983; Waldman et al., 1982). However, fogwater in agricultural areas of the southern San Joaquin Valley is not consistently acidic with reported pH values as high as 7.5 (CARB, 1985).

Table 2. Yield Response of California Crops to Acid Rain (pH 5.6-3.0) (USEPA, 1984)

Crop	Controlled Environments	Field Studies
Alfalfa	+	+
Barley	0	*
Beans, Dry	0	0
Broccoli	0	*
Carrots	-	*
Cauliflower	0	*
Corn, ("Pioneer 3992")	-	0
Corn, Sweet	0	*
Hay	0	0
Lettuce	0	0
Oats	0	*
Onions	0	*
Potatoes	0	*
Spinach	0	0
Strawberries	+	0
Tomatoes	0	*
Wheat	0	0

+ = Increased yield
0 = No significant change in yield
- = Decreased yield
* = Data not available

Early evidence of the potential damaging effects of acid fogs on California crops has been reported by Thomas et al. (1952), Thomas (1951), and Middleton et al. (1950). Injury to the upper surface of leaf crops was observed to be distinct from smog injury. Thomas et al. (1952) noted "spot-type lesions" on endive, alfalfa, table beet, and spinach after exposure to fog of pH 3.0 or less. More recent studies have been conducted by Musselman (1985) and Granett and Musselman (1984). These studies have exposed agricultural and horticultural crops to simulated acid fog. Exposed crops include: azalea, zinnia, radish, celery, tomato, lettuce, strawberry, spinach, and carrot. Generally, greenhouse and field experiments showed that leaf necrosis develops after short-term exposures (e.g., five biweekly one hour foggings) at approximately pH 2.5 with growth and yield effects occurring at pH 2.0 or less. Radishes (foliage) appear to be more sensitive than the other crops tested. However, foliar necrosis from acid fog (i.e., pH 2.6) on crops such as celery, lettuce, and spinach may be more economically important.

Results of these studies also showed that leaf surfaces have the ability to buffer acid fog deposition. Only when treatment pH levels were less than 3.6 after a two hour exposure did leaf surface pH begin to decrease. These results may explain why injury was not apparent at the higher pH treatments (i.e., pH > 3.6). Further research using longer duration fog events and examining gaseous pollutant interactions is necessary before definite conclusions on crop response can be drawn.

Beneficial Effects

The addition of NH_4^+, NO_3^-, and SO_4^{2-}, major constituents of acid deposition, can make an important contribution to the nutrient needs of crops (Hidy et al., 1986). Deposition of ammonium, nitrate, and sulfate in

California generally occurs in the ranges of 0.7 - 3.7, 2.0 - 11.0, and 3.0 - 8.0 kg/ha/yr, respectively (Hidy et al., 1986). While the amounts of ammonium and nitrate are probably too small to make a significant contribution to cultivated crops, they may be important to the long-term nitrogen supply of "unmanaged" lands used for grazing.

Sulfate deposition from the atmosphere may be important to crop production in California. Deficiencies of sulfur in soils have been identified in 32 states, including California (CAST, 1984). According to Ensminger and Jordan (1958) annual sulfur requirements for medium to high yielding crops range from 8 to 32 kg/ha. Thus, in regions where required sulfur needs of crops are exceeded by the sulfur-supplying capacity of the soil, acid deposition may make an important contribution to meeting crop nutrient requirements to achieve maximum yields.

Conclusion

The reliable, but limited scientific evidence to date suggests that current levels of acid precipitation are not having a major effect on crop yields in California. Results of field and controlled environment studies indicate that the net response of crops to acid precipitation is a result of the interaction between the positive effects of S and N fertilization and the negative effects of air pollution, including acid rain. While not all crops grown in California have been evaluated for their response to acid deposition, crop loss due to acid rain and fog appears to be very small compared to losses from natural factors such as drought and disease.

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INTERACTION OF AIR POLLUTANTS AND ABIOTIC STRESSES ON CROP PLANTS

Patrick J. Temple
Assistant Research Botanist
Statewide Air Pollution Research Center
University of California
Riverside, California 92521

Some General Considerations

An interaction between two (or more) stresses can occur when the presence of one stress alters the response of the plant to the other, so that responses to the combination of stresses is greater or lesser than that of the additive responses of the two acting alone. Greater than additive responses can be termed positive or synergistic; while less than additive responses can also be called negative or antagonistic. This definition also includes the possibility that additive effects of two environmental stresses could force a plant response (e.g., ratio of photosynthesis to respiration; visible foliar injury; reduction in growth) above a biochemical, physiological, or economic threshold, thus appearing to be a positive interaction.

The primary mode of entry of gaseous air pollutants into leaves is through the stomata. The intensity of plant responses to air pollutants is strongly influenced by the degree of stomatal opening. Environmental factors conducive to high rates of stomatal conductance, such as high relative humidity, moderate to high light intensity, and adequate soil moisture, are also conducive to maximum impacts of air pollutants. Conversely, environmental factors that reduce stomatal conductance, either directly or indirectly through effects on photosynthesis, render plants less susceptible to air pollution effects. Since abiotic stresses, by definition, have adverse effects on the physiological functioning of plants, interactions between air pollutants and abiotic stresses usually produce additive or less than additive (negative) responses.

Some Specific Examples

Drought Stress

Most field and laboratory studies have shown that under certain circumstances drought stress can protect plants from air pollution injury. The mechanism of this protective effect is presumed to be reductions in stomatal conductance induced by drought stress, although participation by biochemical products triggered by drought stress has not been eliminated. Field studies of crop responses to ozone (O_3) under normal or water-stressed growing conditions have shown that severely water-stressed cotton, soybeans, and alfalfa had less foliar injury and less reduction in yield induced by O_3 than adequately-watered plants. The degree of protection from O_3 was influenced by the severity of the drought stress and by environmental conditions, particularly potential evapotranspiration (ET_o). For cotton and soybeans, protection from O_3 was observed only when the drought stress was severe enough to reduce yields 25 per cent or more, relative to adequately-watered plants. For alfalfa, O_3 protection was found only during summer months, when ET_o was high. In April and October, when ET_o was relatively low, drought stress had no effect on alfalfa responses to O_3.

The protective effect of water stress against air pollution injury has been suggested as a management tool to protect sensitive stages of crop development from severe air pollution injury. For example, irrigation could be delayed a few days to keep a crop at a less sensitive stage of development or to reduce stomatal conductance during severe air pollution episodes, which typically last only a few days. Such a technique would be particularly useful for ornamental, horticultural, or leafy crops in which appearance and quality are more valuable than bulk yield. Crop models relating plant responses to air pollutants under various drought stress scenarios are now being developed which could aid in the implementation of this technique.

Other environmental stresses which either induce water stress directly or are similar to water stress in that they reduce gas exchange and photosynthesis also tend to reduce plant responses to air pollutants. These stresses include temperature extremes, high light intensity, excess salinity, and soil anoxia.

Toxic Substances

Large numbers of gaseous, liquid, and particulate pollutants have been shown to interact among themselves and with air pollutants to alter plant responses to air pollutants. Interactions among gaseous air pollutants, particularly O_3, sulfur dioxide (SO_2), nitrogen dioxide (NO_2), hydrogen fluoride (HF), and peroxyacetyl nitrate (PAN) have received the most attention, but potential interactions between air pollutants and acid precipitation, heavy metals, and volatile organic compounds (VOC) have been demonstrated in the laboratory. Studies of interactions among gaseous air pollutants have shown that, in most cases, interactions between O_3 and SO_2 are additive; that is, crop plants respond to each pollutant independently. However, NO_2, which by itself is not phytotoxic at ambient concentrations, can lower the threshold of plant response to SO_2 and/or O_3. The mechanism of this interaction has not yet been established.

Potential interactions among gaseous air pollutants, particularly O_3, and acid precipitation have recently been the focus of numerous laboratory and field studies. Recent hypotheses concerning the causes of forest declines in Europe and high altitude forest declines in North America have centered around possible combinations of gaseous and liquid pollutants, possibly coupled with heavy metal toxicity and drought stress. Since O_3 has been shown to increase membrane permeability leading to leakage of ions, O_3 injury may predispose plants to acid precipitation effects by increasing loss of nutrient ions or buffering capacity from plant foliage. Conversely, high acidity in precipitation may impair the mechanism of stomatal closure, leading to increased flux of pollutant gases into leaves. Most field studies of acid precipitation and gaseous pollutant interactions have not found any evidence of significant positive or greater than additive responses in plants exposed to combinations of the two stresses. However, this does not exclude the possibility that combinations of pollutant stresses, while not interacting *per se*, may additively reduce plant growth and yield below an economically acceptable threshold that otherwise would not have been reached.

Interactions between air pollutants and toxic heavy metals have not been studied extensively. Laboratory experiments suggested that plants grown in the presence of elevated but sub-toxic levels of nickel, cadmium, or zinc had greater foliar injury and growth reduction induced by O_3 than plants not exposed to these metals. The mechanism for this apparent interaction or its practical significance have not been determined. Interactions between air pollutants and VOC have also not been studied in detail. Combinations of O_3 and herbicides have given greater than additive, less than additive, or no interaction, depending upon plant species and particular herbicide. Because of the extensive use of agricultural chemicals these interactions between air pollutants and toxic organic compounds should be investigated more thoroughly, to determine the potential significance of these interactions under field conditions.

Conclusions

Since environmental stresses, by definition, reduce optimal physiological functioning in plants and since plants are typically most susceptible to air pollutants when exposed under optimal growing conditions, interactions between air pollutants and abiotic stresses often produce either no interaction or less than additive responses in plants. However, potential combinations of gaseous air pollutants and abiotic stresses including drought, salinity, temperature, radiation, acid precipitation, heavy metals, toxic organic compounds, nutritional imbalances, and many others, are so numerous that only a few have been studied in detail. More research is needed to determine the significance of these interactions for crop growth and productivity.

INTERACTION OF AIR POLLUTION AND BIOTIC STRESS ON CROPS

P. M. McCool
Assistant Research Plant Pathologist
Statewide Air Pollution Research Center
University of California
Riverside, California 92521

Plant growth and productivity is determined by the external environmental factors which impact upon it. These environmental factors may enhance growth or result in plant stress. Biotic factors, such as pathogens, insects, or beneficial microbes interact with environmental factors to modify plant response.

Air pollutants are a major environmental stress factor for plants and research has demonstrated that interactions occur between pollutants and biotic organisms. Most research on biotic stress/air pollutant interactions has focused primarily on plant pathogens interacting with ozone and sulphur dioxide. More recently, however, other biotic stresses such as insects, and other pollutants, primarily acidic precipitation, have been investigated.

Ozone/Biotic Stress Interactions

Ozone has been shown to decrease the severity of disease of several obligate fungal parasites. Responses have been shown to depend on the ozone dose, and the host-parasite combination. Presumably, since the obligate parasites require healthy plant tissue, necrosis or senescence induced by ozone may reduce the potential sites of infection. However, recent studies of O_3 on *Uromyces phaseoli* of bean indicate both a negative and positive effect. Although ozone decreased the size of primary uredia, increased pustule numbers with secondary uredia were noted. Effects of ozone on facultative parasites have demonstrated increased, decreased, or no effect on levels of parasitism. For example, Botrytis spp. have demonstrated increased levels of disease incidence on potato and onion after plants are injured by O_3. Beneficial mycorrhizal fungi have also been shown to be negatively affected by exposure of the host plant to ozone.

Bacerial pathogens also respond to ozone-injured plants, but the response can be negative or positive depending on dose and timing of exposure relative to inoculation. Nodulation of Rhizobium has been shown to decrease in roots of legumes exposed to ozone, while foliar infection by Pseudomonas and Xanthomonas sp. can be reduced or unaffected depending on timing of ozone exposure.

Ozone by influence the feeding preferences of insects. Mexican bean beetle and gypsy moth both preferred O_3-exposed foliage over non-exposed foliage. Growth rate of aphids was higher on roses grown in ambient polluted air as compared to filtered air. Root-inhabiting nematodes have been shown to suffer reductions in reproduction after plants were exposed to ozone prior to inoculation.

Sulphur Dioxide/Biotic Stress Interactions

Much like ozone interactions, sulphur dioxide tends to inhibit obligate fungal pathogens while non-obligate pathogens are variable in response. Parasitism of wheat by Puccinia graminis was inhibited by SO_2 but type of wheat resistance to the disease was important in the response to SO_2.

Lesions of Scirrhia acicola on needles of Scots Pine have been shown to increase with exposure to SO_2 after inoculation. However, Alternaria solani on tomato was unaffected by sulphur dioxide exposure and tar spot of sycamore (Rhytisma acerinum) has shown excellent negative correlations between numbers of lesions and SO_2 concentrations.

The relationship of SO_2 and bacterial diseases generally seems to be inverse with reductions in lesions size and rate of lesion development frequently occurring as SO_2 increases. However, increases in viruses have been observed in bean and maize as a result of SO_2.

As observed with ozone, sulphur dioxide may enhance feeding by insects. Mexican bean beetle and black bean aphid exhibited feeding preferences or enhanced growth rates with SO_2-exposed foliage.

Acidic Precipitation/Biotic Stress Interactions

Acidic input in the form of rain, mist, or fog may also interact with biotic organisms. Most reports to date demonstrate inhibition of plant disease, beneficial mycorrhizal fungi and nodulation of Rhizobium with increasing acidity of rain. Most effects occur at acidities below pH 3.5. Plants exposed to increasing acidity of fog, however, have had increased incidence of disease and insect infestation.

Conclusions

In general, biotic stresses seem to interact with air pollutants. Most obligate fungal parasites are inhibited on plants exposured to ozone, sulphur dioxide, or acidic precipitation. The impact on non-obligate fungi varies, with negative, positive or no effect present depending on the host-parasite system under examination. Beneficial mycorrhizal fungi are negatively influenced by pollutant exposure. Bacteria may respond positively or negatively to pollutants. Beneficial Rhizobium generally seems to be inhibited by pollutant stress. The effects on nematodes also seem to be somewhat variable, depending on the host-parasite system. In contrast, viruses and insects seem to be enhanced somewhat by pollutant stress. Since insects may be viral vectors, this relationship might be potentially important.

Because of the apparent interactions between air pollutants and a wide variety of biotic organisms, these relationships should be considered carefully when evaluating and designing pest management strategies.

THE CALIFORNIA AIR RESOURCES BOARD'S CROP LOSS ASSESSMENT PROGRAM

Homero Cabrera, Sylvia Champomier
California Air Resources Board
Sacramento, California

David M. Olszyk, C. Ray Thompson
University of California
Riverside, California

In 1984, the California Air Resources Board (ARB) initiated a program to assess crop losses caused by air pollution in California on a statewide basis. In 1986, the first statewide assessment of crop losses caused by air pollution was completed. The program used ozone dose-yield response information from a variety of sources, including EPA's National Crop Loss Assessment Network (NCLAN), ARB-funded extramural research, California Department of Food and Agriculture's (CDFA) California Crop Loss Program, and others. Crop production information was furnished through the Department of Agricultural Economics, U.C. Davis.

Data Sources and Methods

ARB's ambient ozone data were used to estimate exposure on a county-by-county growing season basis for each crop in the assessment. Monitoring sites were selected that were deemed to best represent the air quality experienced by each crop in each county where it is grown, and daytime seasonal ozone exposures were estimated on the basis of each crop's growing season. The daytime average was either over seven hours or twelve hours, depending upon the specific yield loss function used for each crop. Yield loss equations for 19 crops were drawn or developed from the literature. The hierarchy for selection of equations was essentially on the basis of comprehensiveness of applicable work: 1) equations derived from EPA National Crop Loss Assessment Network research; 2) equations derived from ARB-sponsored research; and 3) equations derived from research sponsored by California Department of Food and

Agriculture or other agencies. The crops for which yield loss equations were found were: barley, grain sorghum, lettuce, spinach, sugar beets, alfalfa, field corn, sweet corn and silage corn, processing tomatoes, winter wheat, fresh tomatoes, dry beans, cotton, grapes, lemons, onions, oranges, and rice. A yield loss function was found for potatoes, but it was not used because it overestimated losses due to ozone exposure. Thirty crops, for which dose response information is not available, were considered to show no yield loss. Sixteen of those crops are thought to be at risk of damage from air pollution because they are grown in areas where they are exposed to elevated ozone levels.

Ozone-induced yield loss was estimated for each designated crop in each county. The individual county loss estimates were weighted according to the proportion of the crop produced in each county, then aggregated to provide a statewide loss estimate. This procedure was applied to seven air quality scenarios as follows:

1) ambient air quality for 1984,
2) three scenarios for attainment in 1984 of the current state oxidant standard of 0.10 ppm for one hour, and
3) three scenarios of 12-hour seasonal ozone averages not in excess of 0.06 ppm, 0.05 ppm, and 0.04 ppm.

1984 was used as a base year because it was the most recent year for which agricultural production information was available. All yield loss estimates used a seasonal daytime ozone background value of 0.025 ppm based on the background value used by the NCLAN program and values for relatively clean, mostly coastal sites in California. The question of the appropriate value for the seasonal daytime ozone background level for rural areas is currently under review by ARB staff.

Results

The Crop Loss Assessment for 1984 indicates that a number of important crops suffered substantial yield losses on a statewide basis due to exposure to ambient levels of ozone. Among these crops were cotton and grapes, California's two most economically important crops, each of which is estimated to have suffered loss of about 20% on a statewide basis. Other crops believed to have suffered losses in excess of 10% were oranges, rice, lemons, dry beans, and onions. Alfalfa, field corn, sweet corn, silage, processing tomatoes, winter wheat, and fresh tomatoes had estimated yield losses of less than 10%. Barley grain sorghum, lettuce, spinach, and sugar beets are believed to have suffered no yield losses.

The yield loss estimates for the various scenarios indicate that the twelve-hour growing season average ozone concentration of 0.06 ppm would have approximately the same impact on crop yields as ambient air quality for 1984. The scenario which examined the twelve-hour growing season ozone average of 0.05 ppm indicated that yield losses would be similar to those expected to occur with attainment of the current standard.

The changes in yield loss are not uniform with the different scenarios, in part because different crops are grown in different regions. Those regions with poor air quality would show the greatest changes in the yield loss.

Limitations of the Crop Loss Assessment

The ARB assessment of statewide crop loss due to ozone has a number of limitations that arise primarily from the lack of complete information about air quality, species and varietal sensitivity, and influence of growing conditions.

There is little ozone monitoring at sites that can be considered rural, and modeling capabilities are not well enough developed to give reliable air quality estimates for crop growing areas of interest. The approach taken in this assessment was to select the nearest monitoring site with the least urban influence for the location in each county where each crop is grown. Thus, there may be errors of either overestimation or underestimation or both, depending upon the particular conditions in each area.

The range of sensitivities to ozone displayed by different varieties of a single crop species also imposes a limitation on the ARB crop loss assessment effort. For many crops, there may be several varieties available to growers, and older varieties are displaced as new ones are developed. Yield loss studies have tended to focus on those varieties considered the most important economically, not those which are most sensitive. The selection of crops for study on the basis of economic importance rather than sensitivity should reduce the likelihood that the assessment would deliberately favor either sensitive varieties or insensitive ones.

Another limitation of the assessment effort is that, for some important crops, notably tree fruit and melons, there exist no ozone dose response equations. For this assessment it has been assumed that these crops suffer no loss. This has the probable effect of underestimating yield losses and their impact on the agricultural community.

Finally, some environmental factors are known to alter plant sensitivity to air pollutants, but for the vast majority of plants information bearing on this is not available. As a result, adjustments of the loss estimates cannot be made for local differences in growing conditions. However, most of the field studies conducted in California were conducted either in the San Joaquin Valley or in the South Coast Air Basin. These areas are generally relatively arid, and plant sensitivity to pollutants should be lower in these locations than, for example, in coastal areas. Furthermore, there are no grounds to assume that plants should be uniformly more or less sensitive on other sites in the regions where the experimental work was carried out, or in areas of similar general climate.

Economic Assessment

The partial economic benefit of reduced ozone levels on crops was assessed using the California Agricultural Resources Model (CARM), which is a statewide model of California agriculture. The CARM was updated to reflect California agriculture in 1984. The model was used to project the benefit of attaining the hourly .10 ppm standard and of achieving the alternative seasonal ozone standards of .04, .05, and .06 ppm; the estimated annual benefits for these four scenarios are: $101, $206, $119, and $50 million, respectively.

The benefits analysis, while more reliable than traditional approaches to valuation and more comprehensive than most previous assessments, provides only a partial estimate of the economic benefits of reduced ozone exposure. This is because the CARM projections are based on estimated crop losses for only nineteen of the fifty crops in the CARM. For the purposes of this analysis, thirty-five of the fifty crops were assumed not to be affected by ozone.

Conclusions

The results of this first statewide crop loss assessment and economic analysis indicate that 1984 ambient ozone levels probably contributed to yield losses in a number of California's most important crops, and that improved air quality would bring about an increase in economic benefit to producers and consumers. Additional work is needed to improve crop loss estimates, develop and apply approaches to verify the losses in the field, and extend the assessment capability to forest vegetation.

AIR POLLUTION IN CALIFORNIA: WHAT, WHERE, AND HOW MUCH?

D. M. Olszyk
Assistant Research Plant Physiologist
Statewide Air Pollution Research Center
University of California
Riverside, California 92521

Air pollution in California has been a reported phenomenon since the 1500's when early Spanish explorers described a "haze" over the Los Angeles area from Indian campfires. "Anthropomorphic" air pollution associated with human activities continues to be of concern, and especially since the 1940's has been studied in regards to its effects on the productivity and quality of crops. Anthropomorphic air pollution from point sources such as oil refineries, smelters, or other industries has caused localized problems. However transportation and industrial-related activities have resulted in the photochemical air pollution or "smog" problems that have affected both urban and nearby rual areas. Point source air pollution is still a localized problem in some areas of California, and is of renewed interest especially in terms of "toxic waste" questions. However, it is photochemical smog which continues to be of greatest concern for the productivity of California crops.

Types of Air Pollutants

Gases

The four common gaseous air pollutants regulated by California and Federal Ambient Air Quality Standards are ozone (O_3, or "total oxidants" for California standard), sulfur dioxide (SO_2), nitrogen dioxide (NO_2), and carbon monoxide (CO). Ozone is a highly reactive gas that is a product of photochemical reactions involving nitric oxide and hydrocarbon emissions -- primarily from automobiles. Ozone is of the greatest current concern in regards to adverse effects on vegetation of all air pollutants in California. Sulfur dioxide is also highly phytotoxic, and is emitted primarily from either fossil fuel burning plants or ore-smelters. Nitrogen dioxide is formed from emissions from fossil fuel plants, and is found in photochemical smog, however, it is comparatively non-toxic to vegetation. Carbon monoxide is a product of incomplete combustion and is highly toxic to people, but not vegetation.

Dry and Wet Particulates

There are a wide variety of small particles in the atmosphere occurring either in a dry state, or dissolved in moisture. Concentrations of dry particulates in general are governed by standards for suspended particulates and visibility reducing particulates. Concentrations of particulate sulfate and lead are regulated by specific standards. Dry particulates are generally not toxic to plants; they affect vegetation only by preventing gas exchange if the particulates are dense enough to form a dust layer on leaves.

Particulates dissolved in moisture are of concern usually only if they result in acidic rain or fog. Dissolved sulfate and nitrate particles are of greatest concern - forming sulfuric and nitric acid, respectively.

Toxic Materials

There is a wide array of other gaseous and particulate materials, usually occurring only in trace amounts in the air; but which can be very toxic to human health. Few of these toxic materials affect plants directly. The primary concern is only in regard to human ingestion of contaminated plant material.

State Ambient Air Quality Standards regulate hydrogen sulfide (H_2S) and vinyl chloride concentrations. Other toxic materials are regulated either in terms of workplace concentrations or are being evaluated to determine whether they should be regulated. Some toxic materials can result in direct effects to vegetation in relation to nearby industrial sources. For example, heavy metals such as mercury can affect some florist crops. These specific toxic situations can best be evaluated on a case-by-case basis.

Temporal Changes of Ozone in California

Since O_3 is the pollutant of primary concern in California, this paper will focus on changes in O_3 concentrations over time, and the geographical pattern of O_3 concentrations in the state. Temporal changes are difficult to determine because few monitoring stations have remained at the same site for more than a few years. Furthermore, O_3 data from before 1975 is difficult to compare to earlier data because the monitoring methods changed.

The South Coast Air Quality Management District has issued a report describing air quality trends between 1975 and 1984 in the South Coast Air Basin. Over this period there was a decrease in the number of days which the federal O_3 standard was exceeded, and the daily O_3 average for May-October. This improvement in air quality occurred despite a large increase in population in the basin.

However, the average O_3 concentrations are still high in the South Coast Air Basin and it is likely that the geographical area affected by O_3 increased as the population, in especially Riverside and San Bernardino counties, has increased substantially. A similar O_3 air quality pattern may be occurring in the Central Valley, with peak values associated with cities decreasing, but the average O_3 concentrations remaining high. Unfortunately, there is little data to quantitatively document these long term trends.

Geographical Patterns of Ozone in California

Daylight growing season concentrations of O_3 are of the greatest concern for agricultural crops. As part of the California Air Pollutant - Crop Loss project, isopleth lines were drawn to better understand the pattern of agriculturally important O_3 across the state. Concentrations for three months (June-August) were calculated for three parameters of O_3 exposure which normally have been used in controlled experiments: a cumulative dose of hours x pphm for hours with averages greater than 10 pphm, a seven-hour average concentration for hours between 9 a.m. and 4 p.m. (7 hr), and a twelve-hour average concentration for hours between 8 a.m. and 8 p.m. (12 hr). Data were used from all of the 130 plus O_3 monitoring sites in the state using 1984 as the sample year. Based on this analysis, California could be divided into roughly five geographical areas based on the pattern of O_3 concentrations for the three exposure parameters.

1) Coastal areas which ranged from Del Norte County in the north to San Diego County in the south. These areas were characterized by low 12 hr or 7 hr averages of less than 4.0 pphm, and no hours with O_3 concentra-

tions above 10 pphm. Some sites in the coastal areas of Los Angeles, Orange, and San Diego counties had 12 or 7 hr averages a little greater than 4 pphm, but still had no peaks above 10 pphm.

2) Mountain and high desert areas from Siskiyou County in the north, across eastern California to San Bernardino County in the south. These areas were characterized with relatively high 12 or 7 hr averages of greater than 4 or 5 pphm, respectively, but few hours with peak concentrations greater than 10 pphm. The high average concentrations in the southern Sierra Nevada mountains and western Mojave desert may be associated with transport from urban areas, but background concentrations also may be higher than other areas of the state.

3) Sacramento Valley counties from Shasta in the north to Solano in the south. This area was characterized by 12 and 7 hr averages of 3 to 5 pphm, but few hours with O_3 concentrations greater than 10 pphm. East and northeast of Sacramento, 12 and 7 hr averages were greater than 6 pphm, and a number of hours with concentrations >10 pphm O_3.

4) San Joaquin Valley counties from San Joaquin in the north to Kern in the south. This area was characterized by 12 and 7 hr averages of greater than 5 pphm O_3, and greater than a number of hours with >10 pphm O_3. There were increased O_3 averages to over 6 or 7 pphm in the vicinity of Fresno and Bakersfield.

5) Portions of southern California counties away from the coast, including parts of Ventura, Los Angeles, Orange, San Diego, San Bernardino, and Riverside counties. These areas had 12 and 7 hr averages of from 6 to 13 pphm, increasing with increasing distance from the coast and higher altitudes. The 10 pphm dose was very high for many sites in

this area, especially at the base of, and at higher altitudes in the Mountains.

Conclusions

Significant reductions in concentrations of some air pollutants have occurred due to Air Quality Standards, especially SO_2 from point sources, and CO and lead from automobile emissions. However, photochemical oxidants -- primarily O_3 -- still exist in high concentrations in many areas of the State. Pollution controls likely play a role in the apparent reduction in peak concentrations and episodes exceeding the standards. However, the geographical area affected by ozone concentrations may be increasing with increased urbanization of agricultural areas. Toxic air pollutants are an increasingly important issue in relation to human health, but are generally not of concern in regards to plant effects.

PLANT MOLECULAR BIOLOGY AND ITS APPLICATION TO AGRICULTURE

John R. Bedbrook. Advanced Genetic Sciences,
6701 San Pablo Avenue
Oakland, CA 94608
Phone (415) 549-2395

Technological advances in the past five years have made possible modification of plant genotypes by genetic engineering. Such developments have already made available new germ plasm for plant variety development as well as new methods for plant breeding and seed production. Specific examples of such developments will be discussed and problems associated with the commercialization of such developments will be introduced.

Molecular Approaches to Disease Resistance

Linda L. Walling
Department of Botany and Plant Sciences
University of California
Riverside, California

Each year agriculturally important crops suffer losses due to disease. Plant breeders have made significant advances in the control on plant disease by the identification and introduction of genes that confer disease resistance into plant crops. Breeding is an economical, environmentally safe, and effective means of controlling disease.

The application of biotechnology to the field of plant-disease resistance may have a significant impact on agriculture. By utilizing the information about disease resistance established by plant breeders, molecular biologists may be able to overcome several of the limitations that exist in the process of breeding plants for disease resistance. The identification, isolation, and reintroduction of genes important in protecting plants from disease by molecular techniques can potentially shorten a 15-20 year breeding program. Once resistance genes are isolated and characterized, they can be moved into a large number of commericially important plant species. In addition, this technology will allow the separation of resistance genes from tightly linked genes that confer undesirable phenotypes and allow the introduction of genes into species that are currently not interbreeding.

Disease Resistance and Gene Expression

Little is known about the relationship between host genes and disease resistance at the molecular level. One approach to understanding the biochemical mechanisms that are important in protecting plants from disease is to isolate and study genes which are induced during pathogen infection. Four distinct research avenues have been pursued in order to gain insight into plant pathogen interactions. These research stategies were designed to ultimately identify genes that code for products that confer resistance to a specific plant pathogen or genes that code for products important in general plant biochemical defense mechanisms(1).

First, numerous investigators have isolated genes from bacterial pathogens that may be important in plant-pathogen recognition (5). These genes, called avirulence genes, code for proteins that may interact with plant receptor molecules. By isolating and characterizing the avirulence genes and their products, it is hoped that the reciprocal plant molecules will be identified and studied; this will ultimately lead to an understanding of the mechanisms of disease resistance.

The second approach is to isolate genes that code for abundant proteins that accumulate in response to fungal infection or in response to wounding. Lamb and colleagues have isolated a number of genes (i.e., phenylalanine ammonium lyase, chalcone synthetase) that code for the enzymes that synthesize the phytoalexins of soybean (2). His laboratory has very convincingly demonstrated that these genes are expressed in both disease-resistant and disease-sensitive plants. During an incompatible interaction (disease-resistant plant and pathogen) these genes are expressed more rapidly than during a compatible interaction (disease-sensitive plant and pathogen). Ryan and colleagues have isolated genes that are systemically induced during wounding (3). These genes code for proteins that act as proteinase inhibitors. It is hoped that by the study of these gene systems and the elements that regulate these genes that insights into the general defense mechanisms of plants will be gained.

A third approach is to isolate and identify genes that are induced during pathogen infection. In this approach, genes which are expressed in response to pathogen invasion are identified and studied. One limitation of this approach is that the functions for the protein products will not be known at the outset of such experiments. The advantage of this experimental approach is that researchers will be able to identify and characterize genes that have important roles in protecting plants from disease but do not code for extremely abundant protein products. Hadwiger and colleagues have isolated a number of genes which are induced during fungal infection of pea (4). My laboratory has taken a similar, but distinct, approach to the study of the interaction of a bacterial pathogen with tomatoes (see section below).

The fourth approach is to try to directly isolate the genes that confer specific disease-resistance. This is in a more direct although time-consuming strategy. This approach is being used in cases where single dominant genes confer resistance to a particular pathogen. For example, members of Williamson's laboratory at Arco are attempting to isolate the gene, Mi, that codes for resistance to the nematode, <u>Meloidogyne incognita</u>. These investigators have isolated a gene, acid phosphatase -1, that maps very close to the Mi gene on the tomato chromosome 6. It is hoped that the use of the acid phosphatase gene probes will allow these reseachers to walk down chromosome 6 into the Mi gene region. Isolation of this resistance gene will open many avenues of research which will elucidate the potential of specific resistance genes.

Pseudomonas tomato

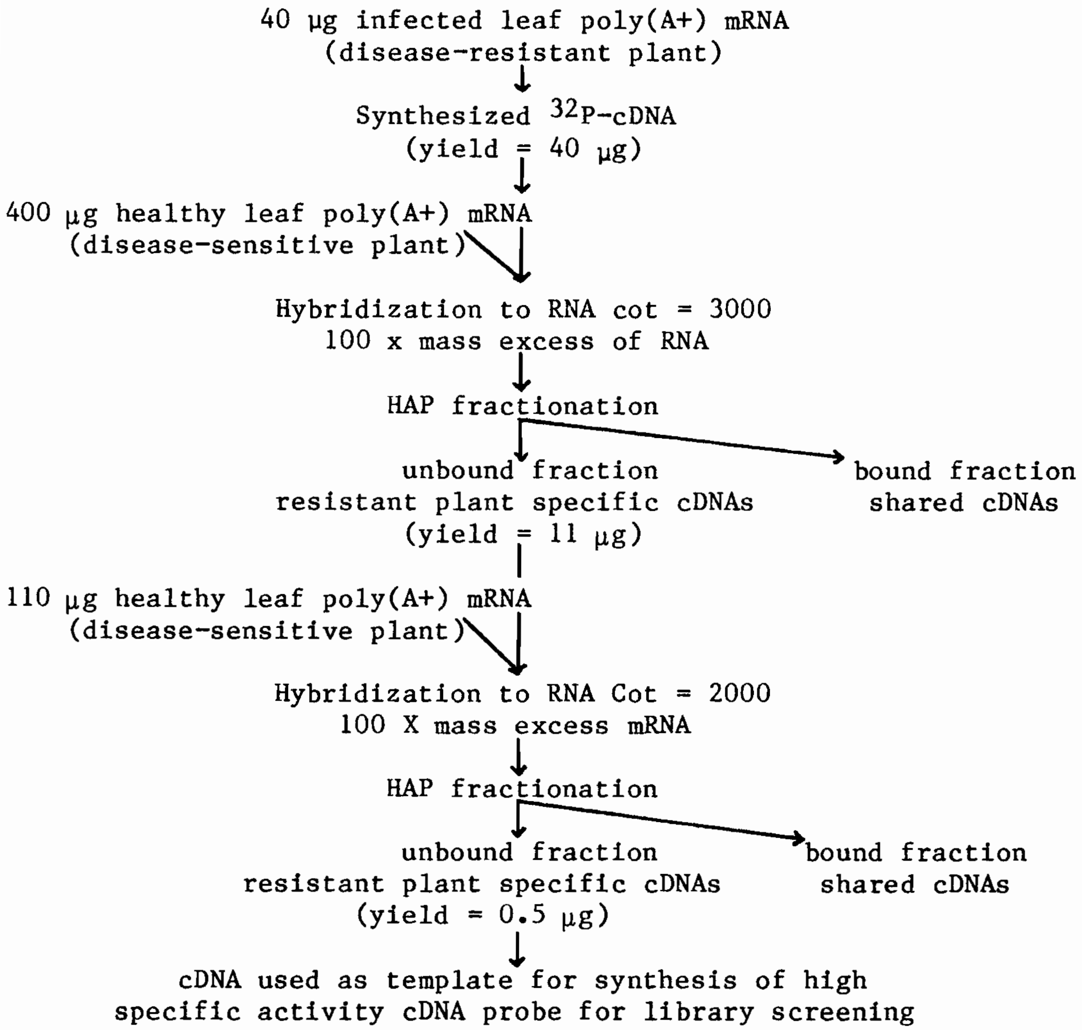
The interaction of the bacterial pathogen, *Pseudomonas syringe* pv. *tomato* (*P. tomato*), with its host the tomato, *Lycopersicon esculentum*, provide an excellent system for the study of the mechanisms of disease resistance at the molecular level. First, *P. tomato* causes the disease entitled bacterial speck which causes losses annually in California's fresh market tomato industry. Second, the study of gene expression during bacterial pathogen infection is less complex than similar investigations into fungal pathogen infections, because plant mRNAs can be readily isolated from the non-adenylated bacterial mRNA. Finally, resistance to bacterial speck disease is conferred by a single dominant gene that maps to chromosome 5 and nearly isogenic tomato lines that are sensitive and resistant to *P. tomato* infection exist.

We are interested in identifying, isolating, and characterizing tomato genes which are induced during *P. tomato* infection. In principle, genes that are induced during infection and wounding can be isolated by the cloning of complementary DNA (cDNA) copies. We have invoked a cloning strategy that will enable us to isolate genes that are induced during wounding or pathogen infection or are constitutively expressed in a disease-resistant plant. One possible and intriguing possibility is that through this study the dominant resistance gene, *Pto*, may be isolated.

To this end, we have constructed a lambda gt10 cDNA library from resistant tomato plant leaf poly(A+) mRNA was isolated 24 hours after *P. tomato* infection. This library was screened with a ^{32}-P DNA probe that is enriched for sequences expressed in a resistant plant during infection. The enriched cDNA probe was isolated using the technique of cascade hybridizations (Figure 1). In principle, this technique will allow for the isolation of cDNA molecules which are expressed in a disease-resistant plant leaf during infection and are not present in disease-sensitive plant healthy leaves. These cDNAs could represent genes which are induced during infection or genes which are constitutively expressed in a disease-resistant plant. The cascade hybridization enriched the initial cDNA population 80X for genes specifically expressed in a resistant plant during infection.

The enriched cDNA was used as a template to synthesize ^{32}P-cDNA. The tomato leaf cDNA library was screened with this ^{32}P enriched cDNA probe and positive clones were picked for further study. Currently, replica filters of the positive isolates are being hybridized to 6 different cDNA probes. These cDNA probes were synthesized from mRNA isolated from healthy, wounded, and infected leaves from disease-sensitive and -resistant plants. These probes will categorize the isolates and determine their basic expression program. Further characterization of the positive isolates is currently underway.

FIGURE 1. Synthesis of an cDNA enriched for sequences expressed during _P. tomato_ infection.



Acknowledgments

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PROSPECTS FOR THE USE OF BIOTECHNOLOGY TO CONTROL VIRUS DISEASES OF PLANTS

William O. Dawson
Department of Plant Pathology
University of California
Riverside, California 92521

Plus-strand RNA viruses are responsible for the majority of virus diseases of plants. These diseases cause chronic losses that significantly reduce production of food and fiber throughout the world. In contrast to crop losses due to other pests, there are no chemical or effective biological controls of virus diseases. There are no effective treatments for plants after they become infected, and susceptible plants have no defense mechanism to overcome the virus after they become infected. At this point the only measures that control these diseases are breeding plants for resistance to infection and cultural practices that prevent infection. Unfortunately, these measures too often are ineffective. Although breeding for resistance is effective, environmentally safe, economical, and does not quickly lose effectiveness against the virus, sources of resistance often are not available that can be bred into desirable plants or are linked to undesirable genes. Cultural methods are effective only in specific ecosystems which make up a minority of virus disease situations. New methods to produce virus-resistant plants would have significant value to agriculture.

One method being examined to control virus diseases is to utilize recombinant DNA technology to use genes for virus resistance that are not useable with conventional breeding technology. However, recent progress in the field of virology suggest the possibility of other novel methods to control virus diseases of plants. Some of these strategies involve expression of parts of viral genomes such that virus replication is prevented. The ability to clone the cDNA of RNA viruses and to artificially extend the host range of these viruses through a DNA phase now allows the genomes of these pathogens to easily be manipulated. Several different strategies are being examined in different laboratories around the world. I will describe some of these approaches.

Hybrid-arrest or antisense RNA has been shown in several biological systems to prevent gene expression. This phenomenon involves the production of an antisense RNA that is complementary to and thus hybridizes to the 5′ end of messenger RNA preventing it from being translated into protein. To make plants resistant to viral infection, the antisense of the 5′ end of the virus genome is being integrated into plants to prevent the infecting virus from initiating an infection.

Recently it has been shown that integration of the coat protein gene of tobacco mosaic virus into plants gives them a degree of resistance against this virus. With more understanding this approach can be refined and amplified.

Satellite RNAs of several viruses have been shown to reduce the amount of disease caused by the helper virus. Thus several laboratories are attempting to integrate the satellite genome into plants in a manner to protect plants against the helper virus.

A phenomenon of "cross protection" in which a mild strain of a virus can prevent the infection of a plant by another strain of the virus which causes severe crop losses has been known for some time. However, the genetic mechanism of this process is not understood. An understanding of this process should allow the design of transgenic plants that would be virus resistant.

The virus replicase appears to be a multifunctional enzyme that replicates the viral RNA which is an obligatory step of the virus lifecycle. One function of the enzyme appears to be a specific recognition of the virus RNA and another function is RNA polymerization. It should be possible to destroy the polymerization function without altering the recognition function and create a replicase that would interfere with the normal replicase. The expression of the defective interfering replicase in transgenic plants could make the plants resistant to viral infection.

Recently, by being able to manipulate the virus genome, we have found that virus resistance can be induced in plants which were thought not to have resistance to this virus. Progress is rapidly being achieved in our understanding of the virus-host interaction due to the revolution of recombinant DNA technology. I am confident that a number of novel strategies for controlling virus diseases will evolve in the next few years.

APPLICATION OF RESTRICTION FRAGMENT LENGTH POLYMORPHISMS TO PROBLEMS IN PLANT IMPROVEMENT

Tim Helentjaris
Senior Molecular Biologist
NPI
Salt Lake City, Utah

Improvement of commercially-useful cultivars and hybrids is often a difficult and costly process due to many factors such as the complex inheritance of the specific traits to be improved, the confusion due to the environment in the evaluation of the expression of those traits, and the individual intricacies involved with each species such as self-incompatibility factors or ploidy number. Molecular markers such as isozymes were previously proposed for use in addressing many different aspects of plant improvement such as analysis of relatedness among germplasm isolates (1), analysis of traits, both simple and quantitative in nature (2,3), and analysis of hybrid seed purity (4). Results gathered to date have been impressive with some problems and disappointing with others due to some of the inherent limitations of isozymes, primarily the overall lack of informative marker loci in commercially useful germplasm.

Restriction fragment length polymorphisms or RFLPs represent another type of molecular marker which can be used analogously to isozymes, but possess advantages which avoid some of the limitations of isozymes. Their number for all practical purposes is unlimited and they have demonstrated informativeness far and above what can be identified with isozymes. For instance in 37 maize inbreds the number of alleles detected with isozymes was 1.9 per locus (5), while examination of the same population with 33 RFLPs located on each of the ten maize chromosomes yielded more than 5 alleles per locus on average. This increased informativeness translates directly into more utility to plant breeding programs.

We have generated large sets of informative RFLP markers for maize and tomato (greater than 100 each, 6) and are now also beginning work in the Brassicas and Cucurbits. We have identified several direct applications of RFLPs to plant improvement programs and are now evaluating their practicality using these RFLP sets as our tools. The first is the use of single or bracketing RFLP loci to follow the inheritance of single gene traits within populations. This has already been demonstrated for different loci, such as dwarf mutations, but has not been pursued actively as these traits are usually already quite amenable to conventional plant breeding practices. Exceptions where this approach may still be desireable are those traits with phenotypes that are difficult to evaluate, such as some disease resistance loci, or those with recessive genotypes, such as some nuclear male steriles. The second application is the use of many scattered RFLP loci to evaluate the overall genetic makeup of an isolate. This could be useful in organizing germplasm into related groups or as part of a varietal protection application. Depending upon the level of variability detected by RFLPs in different species, this will be more or less practical. With maize the level of variability is very high and would allow exact classification of any line, while within domesticated tomato lines, this may prove tedious due to the much lower level of polymorphism (7). The third use is the analysis and dissection of quantitative traits into individual components and the subsequent use of several linked markers to evaluate progeny in a breeding program. We see this as probably having the most potential application to plant improvement programs as these are precisely the types of traits that are most important to the breeders and yet the most difficult to manipulate. While certainly more difficult to analyze, the ability of RFLPs to remove the environmental component from the selection process with concurrent increase in heritability makes this an attractive goal. We have already demonstrated the ability of RFLPs to detect major loci involved in several quantitative traits such as insect resistance and soluble solids in tomato and plant height and grain yield in corn. The fourth application we are investigating is the ability of RFLPs to predict combining ability in hybrid crops. If one could understand better what is involved in two inbred lines that makes their hybrid superior both to the parents as well as to other lines or varieties and learn to predict this trait, then what is now a very involved component of many breeding programs could be significantly simplified. We are now preparing a database on most commercially used maize inbreds and comparing those results with field data towards achieving this goal.

It is felt that the RFLP technology can impact several important steps in plant improvement and in the future will be an integral part of many breeding programs. Data supporting their use in several of these applications has already been obtained and improvements in technology should facilitate increased use in various types of plant improvement programs.

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Bacillus thuringiensis Protein Toxins for Insect Control

Brian A. Federici
Professor of Entomology
Department of Entomology
University of California
Riverside, California 92521

The bacterium, *Bacillus thuringiensis*, consists of a complex of more than twenty subspecies of spore-forming bacteria, all of which produce an insecticidal proteinaceous parasporal body during sporulation. In most subspecies, the parasporal body, which makes up approximately 30% of the sporulated cell's dry weight, is a bipyramidal crystal composed of a 135-kDa protein protoxin that is highly toxic to lepidopterous larvae once activated. Upon ingestion, the parasporal body dissolves under the alkaline conditions within the insect's stomach, and is cleaved by alkaline proteases yielding an active toxin of about 65-kDa. This active toxin, referred to as the δ-endotoxin, binds to receptors on the plasma membrane of midgut epithelial cells, disrupting membrane permeability and causing widespread lysis of midgut cells. Insects typically are paralyzed within a few hours, could die within 24-48 hours.

Studies in several laboratories over the past few years have demonstrated that the genes encoding the 135-kDa protein in four different subspecies, e.g., *kurstacki*, are located on plasmids. The genes for three of these proteins have been cloned and sequenced, and are currently undergoing analysis to determine the regions essential to insecticidal activity. Researchers at Plant Genetics Systems in Belgium have used the Ti plasmid to transform tobacco plants with the gene for the toxin, and have obtained plants which express the protein at levels high enough to protect the plants from damage by lepidopterous plants.

In addition to subspecies, toxic lepidopterous larvae, subspecies of *B. thuringiensis*, have been isolated over the past several years which are active against larvae of, respectively, mosquitoes and beetles. The subspecies active against mosquitoes, i.e., *israelensis*, produces a parasporal body that is spherical rather than bipyramidal, and contains four proteins with molecular weights of 28, 65, 128, and 135 kDa. Presumably, these have different modes-of-action, and already, there is substantial evidence the 28-kDa protein is a broadly cytolytic protein capable of lysing a wide range of vertebrate and invertebrate cells *in vitro*, but, importantly, non-toxic to vertebrates upon ingestion. The 28-kDa protein has also been reported to act as a synergist, potentiating the toxicity of the high molecular weight proteins.

The most recently discovered subspecies of interest is an isolate of subspecies *morrisoni* which is toxic to beetle larvae. This isolate produces a polyhedral parasporal body that consists exclusively of a 65-kDa protein.

The variety of insecticidal proteins that occur among different subspecies of *B. thuringiensis*, combined with the potential of recombinant DNA technology, after numerous possibilities for producing protein toxins specific for insects. These toxins can be either used as conventional insecticides or expressed directly in plants.

The Engineering of Bacteria for Turnover of Toxic Metabolites

J. V. Leary
Dept. of Plant Pathology
University of California
Riverside, CA 92521

and

D. D. Focht
Dept. of Soil and Environmental Sciences
University of California
Riverside, CA 92521

The biodegradation of organic compounds, many of which can be classified as toxic wastes, is considered to be technologically possible. The persistence of many toxic chemicals in soil after a spill is not due primarily to some inherent refractile nature of the chemical but rather to the low indigenous catabolic capacity of the soil microflora. Recent work has shown that two factors are very important in determining whether or not toxic organic compounds are biodegraded: (1) the soil microflora required for biodegradation may not exist in that soil; and (2) the substrate necessary to sustain the specific biodegrading microflora population is lacking. Our present research is therefore based upon the observation that soil does not contain the necessary indigenous microflora to decompose many organic wastes. We have isolated about 50 strains of bacteria from sewage and other sources which, when inoculated into soil containing toxic chlorinated organic substrates, have greatly increased in number and been maintained at those numbers by utilizing the toxic compound as their carbon source.

The aerobic metabolism of all aromatic hydrocarbons involves the direct dioxygenation of the aromatic nucleus followed by a series of proton transfers and rearrangements to catechol. The ring is then split by the action of another dioxygenase. Utilization of the substrate as a carbon and energy source does not occur until the ring fission products are further metabolized and enter the TCA cycle.

The biodegradation of the aromatic hydrocarbons occurs by two pathways. The <u>ortho</u> fission pathway occurs when benzoates or phenols are substrates and involves enzymes of high specificity while the <u>meta</u> fission pathway occurs when aromatic hydrocarbons are substrates and involves enzymes of low substrate specificity. The significance of the <u>meta</u> fission pathway is that bacteria which utilize aromatic hydrocarbons can also oxidize chlorinated aromatic hydrocarbons yet cannot dehalogenate the ring fission products and are thus unable to grow on the chlorinated aromatic analog.

Isolation and Maintenance of Strains

It is important for the understanding of much of the experimental data we will present to be familiar with the procedures for initial isolation of the bacteria which degrade the aromatic hydrocarbons and with the procedure for maintenance of that phenotype.

The bacteria are isolated from sewage and other sources by enrichment in a liquid basal medium containing the aromatic hydrocarbon as the sole carbon source. The bacteria are then recovered as single colonies from similar agar medium. The selective pressure is constant by always growing the bacteria on media containing an aromatic hydrocarbon as the sole carbon source. However, the aromatic hydrocarbon used may not remain constant. For example, a strain which is capable of utilizing benzoate, 3- and 4- methylbenzoate, toluene and xylene may be selected on toluene but maintained and grown subsequently on 4-methylbenzoate simply because it is much easier to do it that way.

This practice is standard but has led up to the first very interesting information concerning the molecular genetics of biodegradation.

Plasmid Biology

Previous studies in other laboratories have shown that much of the genetic information for the enzymes involved in the biodegradation of "exotic" substrates including toxic substances are contained on extrachromosomal plasmids. Therefore, we first examined the total DNA of several Pseudomonas putida isolates and selected strain R5 for further study. Strain R5 utilized the substrates toluene, 3-xylene, 4-xylene, benzoate, 3-methylbenzoate, and 4-methylbenzoate in addition to the the ability to cometabolize 2-,3-,4-chlorotoluenes, and trifluorotoluene. Total DNA analysis of R5 isolates maintained on toluene as the selective sole-carbon source consistently contained three plasmids of 30-100 kb in size. When R5 was grown on different selective carbon sources the plasmid profile and copy-number of the plasmids changed to specifically reflect the substrate upon which the isolate was grown. In every instance the change in plasmid profile resulted from the loss of the 80 kb plasmid and the appearance of plasmids of intermediate size. For example, when R5 previously maintained on media containing

4-methylbenzoate as the sole carbon source to media containing only toluene or 3-xylene, the plasmid profile changed with the loss of the 80 kb plasmid and the appearance of two "new" plasmids ~50 kb and ~60 kb in size. The "new" plasmids appeared to be present in very high copy number. Restriction enzyme digest analysis revealed that, even though it appeared that the total RNA had been reduced, restriction fragments not seen previously from the 80 kb and 100 kb plasmids were present. The most likely explanation for the appearance of these new fragments is that internal rearrangements of the plasmid DNA had occurred. The restriction enzyme digests also revealed that particular fragments, present in the larger plasmids and also present in the 50-60 kb plasmids were amplified in copy number. These rearrangements and amplications were always specifically associated with the particular aromatic hydrocarbon supplied as the sole carbon source.

The relationship of one piece of DNA to another can be ascertained by determining the degree of homology of base pairs by DNA/DNA hybridizations. If the DNAs are closely related, they will share a high degree of homology and will therefore readily hybridize. We have used DNA/DNA hybridization studies to demonstrate that the intermediate size plasmids present in strain R-5 after growth on different substrates all hybridize strongly to the 100 kb plasmid and the 80 kb plasmid present in the original R-5 isolate maintained on 4-methylbenzoate. These results would indicate that all of the intermediate-sized plasmids are derived from the same "parent" plasmid and that the 100 kb plasmid is that "parent".

Subsequent hybridization studies in which specific restriction enzyme fragments were tested for hybridization to the total restriction enzyme fragment digest of the total plasmid DNA from R5 and its derivatives have revealed the following: (1) fragments which are common to all of the digests from R5 derivatives with different plasmid profiles are homologous, regardless of the aromatic hydrocarbon substrate on which the R5 derivative was grown; (2) there was considerable homology between restriction enzyme digest fragments from one R5 derivative and smaller fragments from other R5 derivatives. Such homology strongly suggests the existence of common repeating units of DNA; (3) there is no homology between plasmid DNA from _P. putida_ strain R5 and _P. alcaligenes_ which utilizes the _ortho_-fission pathway.

Genetic Engineering

The ultimate goal of the research is to engineer recombinant strains which can carry out the complete degradation of any single compound regardless of whether it is halogenated or not. Since each of the bacteria has the full capacity to complete one pathway the potential for engineering such recombinant strains appeared high.

The first step in the construction of recombinant strains is to attempt to identify the genes involved in the degradative pathway. This can be done by mutagenesis, cloning, plasmid curing, etc. However, each technique has its drawbacks. However, sometimes nature gives the researcher an unexpected assist.

A variant of *P. putida* strain R5 lost its ability to utilize xylene as sole carbon source. Subsequently, it also lost the ability to grow on the methybenzoates. Analysis of the total plasmid DNA revealed the spontaneous loss or "curing" of the 80 kb plasmid and no intermediate sized plasmids derived from it were present as had been observed previously. This information strongly implicates the 80 kb plasmid as the location of genes involved in the *meta* fission pathway. The restriction enzyme fragments which are derived from the 80 kb plasmid have been identified, inserted into a vector and cloned into *E. coli*. We are continuing our efforts to achieve expression of these genes in *E. coli* and to transfer the appropriate DNA segments into *Pseudomonas alcaligenes*. Such recombinants will greatly enhance the biodegradation of a vast array of toxic organics in soil.

POSSIBILITIES FOR IMPROVEMENTS IN NITROGEN UPTAKE AND UTILIZATION BY CROPS

F.E. Broadbent
University of California, Davis

There are several ways of expressing efficiency of uptake and utilization of nitrogen by plants. For the purposes of this discussion, uptake efficiency will be defined by the ratio (fertilizer N in crop)/(fertilizer N applied). Utilization efficiency will be defined by the ratio yield/(total N in plant). The two measures of efficiency are closely interrelated and difficult to separate; accordingly I will not attempt to do so in discussing measures for improvement.

The most common method for estimating N uptake efficiency is the so-called difference method, based on measurement of N uptake by fertilized and unfertilized plants. By this method, N uptake efficiency is given by [(N in fertilized crop) - (N in unfertilized crop)]/ (N applied). In using this method it is assumed that uptake of soil N is the same in the fertilized and unfertilized plants. However, a fertilized plant will often have a larger root system, enabling it to extract N from a larger volume of soil than an unfertilized plant. Numerous investigators have published evidence of the "priming effect", or stimulating effect of N fertilizers on uptake of soil N. In some cases, uptake of soil N is increased two- or threefold by addition of fertilizer. In other words, the assumptions underlying use of the difference method are often invalid.

A second method of measuring N uptake efficiency is the regression method. Where three or more rates of N are applied to a given crop and soil, a plot of N uptake by the crop against level of N applied may yield a straight line, the slope of which is a measure of uptake efficiency. A third method, and in my opinion the best, is the isotope method, in which the isotopic composition of a crop fertilized with isotopically labeled fertilizer makes possible a calculation of the fraction of N derived from the fertilizer. The accuracy of this estimate in the absolute sense is influenced by the degree of interchange between fertilizer and soil organic N, but it does provide an unequivocal measure of the actual amount of fertilizer N which is present in the current crop.

By any method of estimation, N uptake efficiency is relatively low. An enormous amount of research effort has been devoted to development of management practices which might improve it. These investigations have been directed toward placement, method of application, timing, modification of fertilizers to provide slow release into the soil solution, use of nitrification inhibitors, improved water management, and so on. Many of these practices, alone or in combination, have resulted in substantial improvements in N uptake efficiency, but even with the best management practices, half or more of total crop N is usually derived from soil sources. Table 1 presents some data for a variety of crops in California, where all the measurements were made by the isotope technique.

Table 1. N uptake efficiency of various crops as measured by the isotope method.

Crop	N-rate, kg/ha	% fertilizer N in crop
corn (5 yr. mean)	90	61.8
"	180	55.8
sugarbeet	56	42.3
tomato	131	32.6
lettuce	134	15.3
broccoli	134	33.5
potato	202	57.2
rice	134	52.8

Even in the best case, 38% of the applied N was not assimilated by the crop, and in the worst case 85% of the applied N failed to reach its intended destination. At least in theory, there is ample room for improvement.

Possible fates of fertilizer N include the following:

1. Plant uptake
2. Immobilized (microbial uptake)
3. Fixed by clay minerals
4. Volatilized as ammonia
5. Leached beyond the root zone
6. Denitrified

In order to maximize plant uptake we devise management practices which will provide adequate levels of N in the root zone to meet plant requirements while minimizing the other possible fates. In the long term, immobilization and fixation by clay minerals do not permanently divert N away from plant uptake, and in any case are not readily susceptible to change by management practices. The principal diversions are gaseous and leaching losses. Management approaches to improve N availability to plants include the following:

1. Placement in the root zone to improve root access to fertilizer and to avoid volatilization losses.
2. Timing of applications to provide adequate N during periods of peak demand while avoiding excess.
3. Formulation of fertilizer materials to avoid losses, including use of slow-release materials.
4. Use of special chemicals such as nitrification inhibitors and urease inhibitors to avoid leaching and/or gaseous losses.
5. Adjustment of N rate to optimum levels.
6. Good water management.

Detailed consideration of each of these is beyond the scope of this discussion. All of them have produced improvements in N uptake efficiency in specific applications, and best management practices will usually include a combination of these approaches tailored to the individual site and crop. Speaking in general terms, the point of diminishing returns has been reached for improvements in placement and timing for many economic crops. Further improvements may be attainable, but are ruled out for reasons of economics or grower convenience. Improvements through refinement of optimum N rates can still be made if there is an economic incentive to do so. We need more research on the relationships between N uptake efficiency and water management practices in order to exploit the potential advantages beyond the obvious one of avoiding excess leaching.

What else can be done? Possibly by turning our attention to the plant, further improvements can be made. Here it is appropriate to consider N utilization efficiency, since the objective is maximum yield per unit of fertilizer N applied. Genetic manipulation of plants for other purposes has produced dramatic improvements in yield potential, disease and insect resistance and better quality of plant product. There is good evidence to support the view that plants exhibit genetic differences in nutrient uptake efficiency. Some work which has been done with corn indicates considerable potential for exploiting genotypic differences in N utilization by that crop.

Breeding programs for other crops are looking toward development of cultivars which are tolerant to deficiencies of nutrients such as iron and zinc. Our work with rice over the past 6 years has shown that not only are there differencies among varieties/lines with respect to N utilization efficiency, but also that these differences are quite consistent from season to season, and under different soil conditions. A few illustrative data are presented in Table 2.

Table 2. Values of the ratio of panicle weight to fertilizer N in crop for 24 varieties/lines of rice grown at two locations in the Philippines.

Season	Location	N rate, kg/ha	Minimum	Maximum	F-value
1984 dry season	IRRI	60	265	474	2.79**
1984 wet season	IRRI	30	557	1530	2.54**
1985 dry season	Maligaya	80	164	236	3.13**
1985 wet season	Maligaya	60	167	291	3.67**

These differences are associated with plant characteristics which have not yet been clearly identified. Growth duration is important, and associative N_2-fixation also appears to be. Tillering and root weight have not been found to be consistently related to N utilization efficiency. The superior performance of certain lines over several seasons on both good and poor soils suggests that a breeding program to maximize N utilization efficiency is both feasible and practical.

Optimal Distribution of Nitrogen in Foliage Canopies

R. S. Loomis and C. I. Badilla[1]
Department of Agronomy and Range Science
University of California, Davis

Nitrogen is the nutrient most often found limiting to plant productivity and it is by far the most-used fertilizer in agriculture. Since maximum rates of leaf photosynthesis increase as a diminishing returns function with leaf N content (1,3), we may question how much N a canopy should have or how a finite but limited supply of N is best distributed within a canopy. Should we have a large leaf area (complete interception) of low capability per unit area or a smaller area with greater capability? Should N content vary vertically with the upper, exposed leaves having the greatest content?

The answers to such questions can be obtained through the integration of several factors in an appropriate, quantitative model. The nature of the relationship between leaf photosynthesis rate and N content is critical. In addition, the optimal distribution of N will vary with the amount of light received and its distribution within a canopy so canopy architecture, solar altitude and the relative contributions of direct and diffuse sources are important. Also important are the costs for acquisition of N from the soil, its reduction, and the synthesis and maintenance of leaf proteins. Acquisition costs, for example, would include costs for root growth as well as those for ion uptake.

We took the econometric concepts outlined by Mooney and Gulmon (2) as the starting point for layered-canopy model. Those authors defined the net marginal gain of carbon by a plant, G, in the following form:

$$G = \frac{dP}{dN} - \frac{dC}{dN}$$

In this equation, dP/dN is the slope of photosynthesis vs N content and dC/dN is the slope of carbon costs vs N content. Since dP/dN declines with increasing N while dC/dN can be assumed to remain constant, G is a large positive number with

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[1] Permanent address: Facultad de Agronomia, Universidad de Chile, Santiago

low N and declines as N content increases. The greatest N-use efficiency (defined here as net photosynthesis achieved per unit N) occurs with low N but the problem then is that P is very small. Mooney and Gulmon concluded that carbon gain is maximized when the N content of the plant's leaves is adjusted so that G = 0. The amount of N in the canopy at G = 0 is strongly dependent on acquisition costs.

We are not sure how to estimate acquisition costs since some of the cost of root growth should be assigned to uptake of other nutrients and water. By confining our attention to the optimal distribution of a finite, limiting amount of N (G > 0), acquisition costs can be assumed to be constant. We have included in the model only the costs associated with synthesis and maintenance of proteins. The distribution of those costs over the life of a leaf is illustrated in Fig. 1 for two N levels.

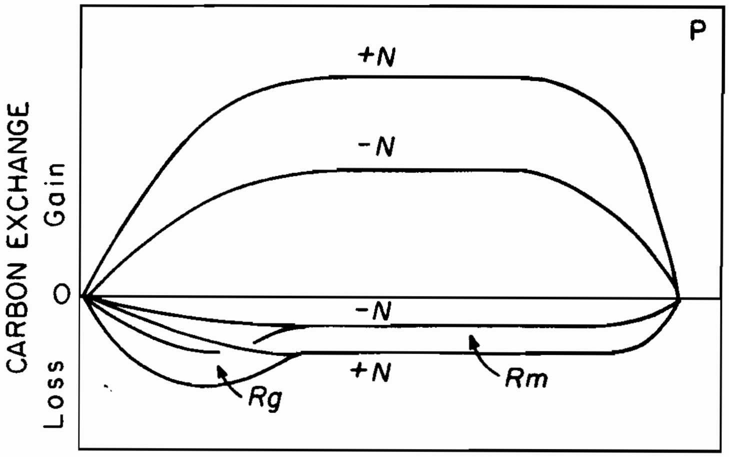
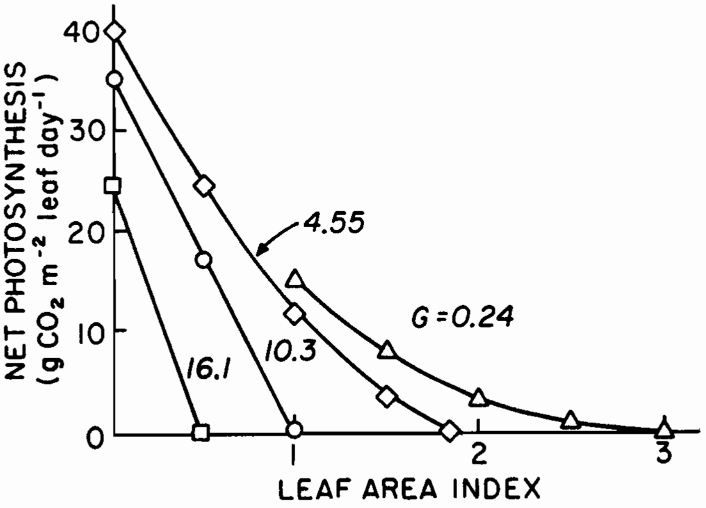


Fig. 1. Stylized daily rates of photosynthesis (P) and respiration associated with growth (Rg) and maintenance (Rm) over the course of a leaf's lifespan. Adapted from Mooney and Gulmon (3).

The problem for the model is defined as follows: given some amount of leaf area and N, how should a new unit of N be distributed between growing more leaves or increasing the N content of existing leaves. From here on, the model takes a rather complicated form in a series of integral equations which were programmed in CSMP and FORTRAN for computer simulations. Special problems arise with the time element. Maximum performance of a canopy would be obtained by changing the N distribution every time the light environment changed, i.e., every minute, but the time constants for leaf growth, N mobilization and protein synthesis are days or hours. In addition, synthesis costs must be prorated over the life of the leaves. Our conclusion (and the model's prediction) is that optimization occurs to the daily and weekly patterns of radiation.

Here, we will present some of our findings with the model since the details of its construction are better left to another time. The simulations were run with photosynthetic functions characteristic of C_3 plants using canopies of horizontal leaves with solar data for June 23 and 33°N latitude (Imperial Valley). Photosynthesis is maximized when all leaf layers have the same marginal gain for the daily period so we set the model to calculate equal G for each layer for the solar track of June 23.

In Fig. 2, the daily photosynthesis of each canopy layer is plotted against its depth in the canopy for different values of G. Photosynthesis increases throughout these optimal canopies as G declines. The N contents of the layers are plotted in Fig. 3. In this graph, the area under each G



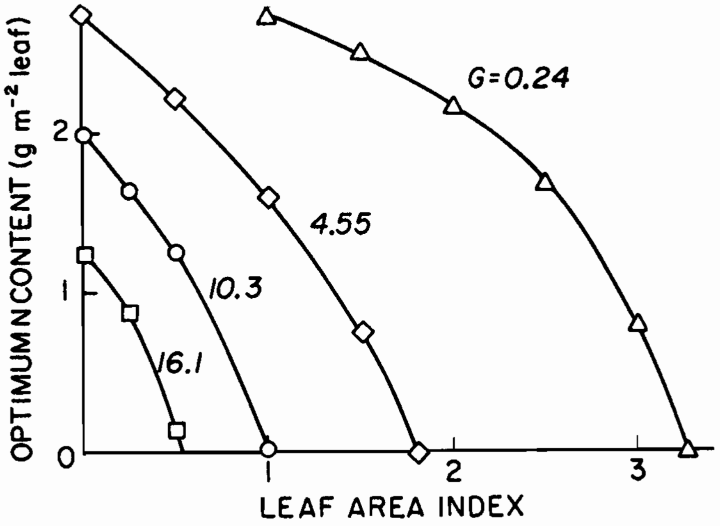


Fig. 2 (left). Simulated daily net photosynthesis of a canopy layer at various values of G as a function of the leaf area index above it (i.e., its depth in the canopy). The isolines follow common G values.

Fig. 3 (right). N content of canopy layers for various values of G as a function of their depth in optimal canopies. The area under each G isoline corresponds to the total amount of N in the canopy.

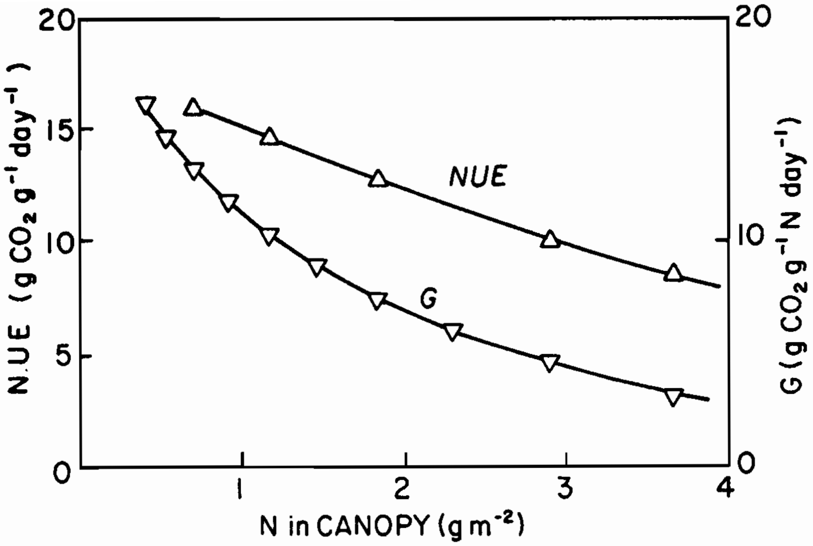


Fig. 4. Net marginal gain (G) and N-use efficiency (NUE) as functions of total N in the canopy.

isoline is proportional to the total N content of the canopy. We used those totals to illustrate the declines in G and N-use efficiency with increasing N content of the canopy (Fig. 4). A key point here is that the decline in G is exponential -- each additional bit of N in an optimal canopy gives a smaller further decrease in G (and smaller increase in photosynthesis).

These results follow intuitively from the assumptions underlying the model but detailed quantitative predictions are possible only with a model. In other simulations, we found that the optimal distribution of N in foliage canopies was strongly dependent upon leaf life span, leaf angle, solar track, photosynthesis function and other factors.

Thus far we have said nothing about what becomes of the new assimilate achieved by optimal canopies. Photosynthesis cannot proceed very long without some use of the products or some place to store them. Since the growth of new sink tissues is also dependent upon N supply, the canopy model must be linked with a model which partitions N and carbon within the crop. Our preliminary work in that area offers some very interesting insights into how natural and artificial selection for factors influencing N-use efficiency would affect the system. In natural systems where competition is mainly with unlike plants, there is a strong advantage from maximizing, early in the life cycle, both the amount of leaf area and the amount of N which it contains. This allows heavy shading of neighbors while at the same time reserving large amounts of N which can be mobilized later to reproductive growth.

It appears to us that crop plants retain many traits from wild types which may not be the most efficient for production in monocultures. Chief among these is a tendency for excessive production of leaves. It is important to remember that natural selection for traits favoring individuals rather than community performance continue in most domestic populations. We are optimistic that further work with models such as that presented here will provide a robust theory of N-use efficiency which can serve as a basis for improvements in crops and management practices.

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VERTICILLIUM WILT OF PISTACHIO: THE INFLUENCES OF INOCULUM DENSITY AND NUTRITION UPON INFECTION

Lee J. Ashworth, Jr.
Plant Pathologist, Emeritus, University of California, Berkeley
Currently, President, The Smith-Baggett Company, Kingsburg, CA

The incidence of Verticillium wilt of pistachio, grown in soils with low inoculum densities of Verticillium dahliae (0.02-0.2 microsclerotia per gram of soil), increased dramatically during 1981-1983. Increased incidence of disease was associated with unthrifty trees. Potassium (K) and phosphorus (P) deficiencies accounted for unthriftiness and poor yields of pistachio trees. K-deficient trees had sparse foliage canopies, small leaves without chlorosis, and pronounced shoot dieback. P-deficient trees had normal dense foliage canopies, normal-sized leaves, and leaf chlorosis. Chlorosis of P-deficient trees appeared first on leaves terminal to nut clusters but later involved part or all foliage of the affected trees. Chlorosis was first interveinal, but leaves later became bright yellow, desiccated, and dehisced prematurely. The threshold K level of leaves for deficiency appeared to be 0.7-0.9% in midsummer. The threshold P level for deficiency appeared to be 0.09-0.1%. Vegetative growth resumed within 2 mos. following treatment with P and within 1 yr following treatment with K.

Increased infection was first associated with trees having low levels of leaf K (0.5-0.8%) during midsummer. During 1982, 39.6% of K-deficient trees surveyed were infected. Infection was rare (0.37%) among thrifty trees having 1% or more leaf K. Infection percentages were 18.8, 13.8, and 9.7 in 1983 for, respectively, 1.0, 1.5, and 3.0 kg of K per tree treatments. Tree survival, compared with the 1.0 kg of K per tree treatments, was improved 8% and 13% in, respectively, the 1.5 and 3.0 kg of K per tree treatments during 1983. Results were similar in 1984 except the 1.5 and 3.0 kg of K per tree treatments were equally effective. Annual increases of infection observed during 1981-1983 were reversed in 1984 following treatments of 1,620 ha with 1.5 kg of K per tree in 1983. No infection of thrifty trees was observed in 1984. Incidence of disease in 1984 was 35% less than that during 1983. Phosphorus deficiency, discovered in 1984 and corrected in 1985 and 1986, resulted in further reduction in disease incidence.

DRIS AS A DIAGNOSTIC TOOL FOR ASSESSING CROP NUTRIENT STATUS--CALIFORNIA STUDIES

Milton B. Jones

Procedures for diagnosing the nutrient status of crops by tissue analysis generally require the sampling of particular plant parts at particular stages of development under given environmental and management conditions. The percent N, P, K and S decrease with age. Concentrations are usually higher in younger tissue, and defoliation such as grazing often increases nutrient percentages compared to ungrazed plants. Nutrients ratios such as N/P or N/S do not change as much through time or with plant part sampled as do the individual nutrient concentrations. The Diagnosis and Recommendation Integrated System (DRIS) takes advantage of this characteristic, making sampling time and plant part less critical.

DRIS assesses nutrient status of crop and forage plants by comparing ratios of important nutrient element concentrations in the tissue being assayed with the same ratios (called reference norms) from high producing crops. These comparisons are made simultaneously on several ratios by computing a DRIS index for each nutrient being studied. From these indices nutrients are ranked in order of those most limiting to yield, the most negative index indicating the most deficient nutrient, and the highest positive index value indicating the most abundant nutrient. The total of these indices, added together whether they are positive or negative numbers, indicates the degree of nutrient balance; the lower the sum, the more nearly the nutrients are in balance.

Reference norms to enable use of DRIS are available for corn, wheat, soy beans, alfalfa, sugar cane, potatoes, pineapples, sorgum and rubber trees. Subclover has recently been added to the list. Large responses to N, P, S and sometimes to K and Ca have been observed on the annual grasslands of California, and subclover has supplied the N needs of many pastures. Assaying the nutrient status of subclover growing in these nutrient deficient pastures can be very helpful in efficiently utilizing fertilizers to increase production.

Deriving Reference Norms

Reference norms for subclover were derived from the top yielding 100 observations out of 396 in the data base. The designation "top yielding" was for plots having a yield of at least 90% of maximum for that site. These observations were made on five fertilizer trials, taken during March and April from several soils with site rainfall varying from 20 to 60 inches in northern California.

Computing DRIS indices

N INDEX = [-f(P/N) + f(N/S) - f(K/N)]/X, where X = 3 and f(P/N) = (P/N - p/n)10/sd, where P/N = actual ratio and p/n is the value of the norm ratio; sd = standard deviation in the high yielding clover. The following nutrient percentages illustrate the computation of DRIS indices; N 2.80%, P 0.23%, S 0.13% and K 3.13%.

	(RATIO	- NORM RATIO)=	DIFF.	X 10	/ sd	= f(P/N)
P/N	0.082	0.091	-0.009	-0.089	0.022	-4.026
N/S	21.538	14.438	7.100	71.005	3.910	18.160
K/N	1.118	0.751	0.367	3.669	0.184	19.938
P/S	1.769	1.291	0.478	4.782	0.386	12.389
K/P	13.609	8.507	5.102	51.017	2.244	22.735
K/S	24.077	10.510	13.567	135.669	2.615	51.881

N INDEX = [-f(P/N) + f(N/S) - f(K/N)]/X = -1.935

P INDEX = [f(P/N) + f(P/S) - f(K/P)]/X = -2.106

S INDEX = [-f(N/S) - f(P/S) - f(K/S)]/X = -27.477

K INDEX = [f(K/N) + f(K/P) + f(K/S)]/X = 31.518

The above indices indicate S to be the most limiting element with N and P in balance and K in luxury supply. A similar conclusion may be drawn in this case from the concentration data.

DRIS vs Critial level approach

Several studies have evaluated the effectiveness of DRIS as compared to other plant analysis interpretive systems for several crops. In almost all cases DRIS has produced more accurate diagnosis than conventional approaches (Kelling and Schulte 1986). Escano et al. (1981) found that use of published DRIS norms was not as accurate as locally calibrated critical values, but when DRIS norms were established with local data it was 2 to 8% more accurate than locally calibrated critical values. We have found in our California studies on subclover that the norms we have developed are not accurate on some soil types, indicating local calibration is important.

There is no reason to choose between the two approaches since the same chemical analyses can be used for both. The two should be used to compliment each other. Since DRIS is based on ratios and nutrient balance, it is possible to have all low nutrient levels in a plant, and still have the nutrient ratios within the optimal range. In which case it is important to look at concentrations directly. On the other hand we have seen cases where DRIS indicated a nutrient imbalance when concentrations were above critical levels, and a responce to fertilization was observed. Soil tests are also useful in pinpointing some problems not apparent with the plant assay methods.

Kelling and Schulte (1986) indicate many workers tend to over-interpret DRIS data. The system is designed to examine all nutrients simultaneously. Since it is almost impossible to have all ratios exactly at the norm value, some indices may be slightly positive or negative even if the plant is in near perfect balance. Since the sum of the indices must be zero, any positive values must be off set by negatives. Workers interpreting the data must have sufficient experience to know what optimal DRIS ranges are.

Conclusions

DRIS is a useful tool in diagnosing the P and S status of subclover under field conditions. The K DRIS index also apprears to be accurate, but more K responsive sites growing subclover are being tested. The percentage of Ca and Mg did not decrease with subclover maturity in our studies. Assaying the status of these two elements on subclover with DRIS does not seem promising.

Like any tool DRIS must be used with skill and judgement in conjuction with traditional methods of evaluating plant analysis and soil test data.

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BERSEEM CLOVER N_2 FIXATION AND FORAGE PRODUCTION

W. L. Graves and W. A. Williams
Farm Advisor, UC Cooperative Extension, San Diego, California
and Professor, Agronomy & Range Science Dep., UC Davis, California

Legumes played an important role supplying nitrogen to crop rotations in many parts of the United States prior to World War II. Since that time, the use of the Haber process to produce NH_3 has resulted in a false sense of security in having relatively cheap and abundant artificial fertilizer nitrogen from natural gas. For the past 40 years commercial agriculture has relied on artificially produced nitrogen fertilizers, putting aside the use of legumes as soil builders and biological nitrogen fixers. Thus, the importance of legumes in crop rotations diminished and the associated research and development of these crops was laid aside for more rewarding career endeavors. However, the 1973 Oil Embargo reminded us that oil, and oil-associated products such as natural gas, are nonrenewable resources and are limited in quantity. The price of these products rose manyfold during the 1973 to 1981 period, and the cost of producing artificial nitrogen fertilizer increased accordingly. We recently have experienced a temporary reduction in fossil fuel costs, but experts expect fossil fuel energy costs to begin to rise again in the near future when demand begins to equal worldwide production levels.

Manufacturing nitrogen from fossil fuels is very inefficient and places a dangerously high demand on available energy. It takes the energy equivalent of 2.3 liters of diesel fuel to produce 1 kg of nitrogen in a urea fertilizer (Green 1978). Considering these factors, the renewed interest in legumes and their role in cropping systems is understandable. Natural biological nitrogen fixation has been shown to be highly effective, yet only a small percentage of the legume family's 20,000 species has been studied (FAO 1984).

Because of this renewed interest in nitrogen fixing legumes, a program was started at UC Davis to reevaluate berseem clover (*Trifolium alexandrinum*) introductions for their use in California as a winter forage (green chop and/or silage) and as a green manure plant. Berseem is a winter and spring annual and is thought to have its region of origin somewhere in the Middle East. Today the country of greatest use is Egypt, where up to 1.5 million hectares have berseem grown as a winter forage or a green manure crop, usually preceding cotton or summer vegetables. Most of our modern day berseem varieties' origin can be traced to one of the Egyptian cultivated landraces: Miscawi, Saidi and Fahl. These three landraces represent the general types of berseem, based on stem branching. Miscawi is a basal, or crown, branching type that can be cut five or six times during its growing season. Fahl is a stem branching type and can be cut only once, while the Saidi is both a basal and stem branching type and can be cut only two to three times during the growing season.

Berseem was introduced into California as early as 1896, into Texas by 1916, and into Florida by 1950. Yet, in spite of its early expectations, it performed sporadically in all of these areas. Most researchers blamed its poor showing on its lack of winter cold tolerance, yet proper inoculation with its Rhizobium was mentioned as a factor but never indicated as the primary cause of its sporadic behavior. Having had the opportunity to work with this plant growing under cultivation in its native habitat, we concluded that berseem, because of its large forage and nitrogen-fixing ability, was worthy of a second chance.

Exploration for Rhizobium strains was conducted in Tunisia by Burton in 1981, and this resulted in the development of significantly superior Rhizobium strains that are available commercially (Graves et al., 1986). Berseem strain and variety introductions were made during the fall of 1981, and since the fall of 1982 field trials have been established yearly at UC Davis to assess the forage and nitrogen fixing potential of some of the multiple cut varieties under supplemental irrigation.

1982-83 Trial Results. This initial variety trial was established October 1, 1982, consisting of the entries "Burton" (French multicut), Sacromonte (Italy), Tunisian common and annual ryegrass. The berseem was inoculated, and annual ryegrass was used as a check on nitrogen fixation. Harvests were made when the berseems were 40 to 50 cm. high. No fertilizer was used, and moisture was maintained nonlimiting by supplemental irrigation. Dry matter production and nitrogen concentration in the forage was measured. Nitrogen fixation was estimated by the difference method, i.e., the nitrogen uptake into the harvested tops of the ryegrass was subtracted from that in each berseem entry to give nitrogen fixed.

Five harvests were made beginning January 2, 1983, and extending to July 13 (Table 1). "Burton" significantly outyielded the other entries with 11,000 lb/a of dry matter and an average protein content of 21%. Ryegrass produced 6,900 lb/a with average protein content of 10%. The nitrogen fixation by the top yielding variety "Burton" was estimated to be 236 lb/a. These results encouraged us to continue the following years' evaluation trials with the "Burton" variety being used as a standard for comparison.

<u>1983-84 Trial Results</u>. A series of plantings were established to assess the effect of time of planting on forage production and nitrogen fixation under supplemental irrigation. Planting dates were September 12, October 7 and October 28. Annual ryegrass was used again to assess nitrogen fixation. A mixture of annual ryegrass and "Burton" (half and half) was used at all planting dates. The berseem seeds were pellet inoculated with the Tunisian <u>Rhizobium</u> strains at the rate of 5 lb/100 lb of raw seed. Again, no fertilizer was used, and moisture was maintained nonlimiting.

The first two plantings produced six cuttings with the first harvest on December 21 and the last on July 20. The latest planting only produced five harvests with the first on March 2. The "Burton" variety again was a high yielder with 7 tons/a, and the total yield was not affected much by planting date. However, the earliest planting date produced one ton/a by December 21, whereas this yield was not attained until March 2 by the latest planting date. Ryegrass alone yielded slightly more than half of clover and was nitrogen limited after the March 2 harvest. The mixture treatments yielded about the same as berseem clover alone, with ryegrass dominating in the early cuts and berseem dominant in the last three harvests. Total nitrogen fixed by the "Burton" variety varied from 303 to 325 lb/a.

<u>1984-85 Trial Results</u>. This trial was established to test dry matter production and nitrogen fixation by various proportions of "Burton" berseem

and annual ryegrass in seed mixtures and to continue the testing of variety adaptation to the Sacramento Valley under supplemental irrigation.

"Burton" berseem and annual ryegrass were planted in monoculture and mixtures (based on viable seed numbers) of 75, 50 and 25 percent berseem clover on October 9, 1984. The other cultivars of berseem included were Fahl (single cut), Bigbee (multicut) and a Miscawi multicut type from Egypt. Inoculation and irrigation were similar to the previous trial, and no fertilizer was used.

Five harvests were made beginning February 26 and ending July 2, 1985. In pure stands the "Burton" variety was the most productive with 13,800 lb/a dry matter and 16% (significant at the 5 percent level) more than the next highest yielder, Bigbee. Of the multicut entries, Egyptian Miscawi was a poor third with 11,500 lb/a dry matter. Fahl, the single cut variety, only produced one cutting at 3,500 lb/a dry matter. Annual ryegrass alone yielded about half as much forage as "Burton" alone. The mixtures of 75 percent and 50 percent "Burton" with annual ryegrass yielded as much forage as "Burton" alone. The amount of nitrogen fixed by "Burton" alone totaled 277 lb/a and the 75 and 50 percent "Burton" with ryegrass mixture did not reduce the amount of nitrogen fixed significantly. The "Burton" berseem became dominant in all ryegrass mixtures by the third harvest.

1985-86 Trial Results. The "Burton" variety was tried with five other multicut berseems; Bigbee, Gigande de Lage, Khadraovi, Egyptian Miscawi and Belem. Annual ryegrass and a half-and-half mixture of "Burton" and Bigbee berseem were also included. Planting occurred on October 7, 1985 following sudangrass for hay. Inoculation and irrigation was similar to previous trials and no fertilizer was used. Six harvests were made from February to July, 1986.

The two top yielders were the Burton-Bigbee mixture and the Burton alone, with both fixing in excess of 300 lb/a nitrogen. Since Bigbee was developed for frost tolerance (Knight, 1985), it was included in a mixture with "Burton" as insurance against a freeze-out, although in our four years of testing no frost damage has occurred on "Burton." Because of its shorter stature, Bigbee appeared to contribute progressively less to the mixture after the first harvest. Annual ryegrass did poorly throughout and was clearly N stressed because of the prior crop of sudangrass and the absence of N fertilizer. Nitrogen uptake by the top treatment (Burton-Bigbee) mixture was about 10 times the 45 lb/a total uptake by the ryegrass, showing the tremendous N_2-fixing power of berseem clover. The poorest producer of the berseems was the Egyptian Miscawi. These two years of poor showing by the Egyptian "Miscawi" has led us to believe that the "Burton" germplasm is markedly different from Egyptian source.

During the 1985-86 season, our consistently good results with the "Burton" berseem encouraged us to apply for and obtain release of this strain as a variety through the California Foundation Seed program. Breeders seed has been increased, and a limited amount of certified seed should be available in 1987.

Summary

These past four years of field testing of multicut berseem germplasm has proven its ability to be a high nitrogen fixer under supplemental irrigation in the Central Valley of California. It has significantly out-performed annual ryegrass, a commonly used winter and spring forage grass, during this period, while fixing 300 lb/a of atmospheric nitrogen. This represents a substantial savings in fossil fuel energy costs for commercial fertilizer nitrogen. These encouraging results have resulted in a varietal release named "Burton" which has consistently outperformed other strains and varieties during these four years of trials.

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Table 1. Dry matter produced and nitrogen fixed by various treatments with berseem clover varieties compared to annual ryegrass at Davis under supplemental irrigation.

Treatment	Planting date	Harvest 1	2	3	4	5	6	Total	N Fixed
		--------------	Dry	Matter	lb/a	-------------			lb/a
					1983				
		1/21	4/1	5/3	6/2	7/13			
Burton	10/1/82	1540	2370	2410	2060	2610		11000	236
Sacromonte		1590	2840	2100	1440	1360		9300	186
Tunisia		0	2240	1960	1920	2430		8600	150
Ryegrass		2980	2240	260	520	860		6900	(169)*
LSD 5%		490	590	760	260	700		1700	
					1984				
		12/21	3/2	3/28	4/24	5/22	7/20		
Burton	9/12/83	2010	1940	2460	2320	2500	2850	14100	311
50:50 mix		2940	1970	1300	1480	2030	2660	12300	196
Ryegrass		2860	1630	620	480	1050	1150	7800	(163)*
		1/13							
Burton	10/7/83	1360	1690	2210	2290	2640	3680	13900	303
50:50 mix		2340	1540	1850	1960	2620	3670	14000	268
Ryegrass		2840	1400	1120	1000	1030	1240	8700	(182)*
Burton	10/28/83	0	1860	2600	2400	2680	3800	13300	325
50:50 mix		0	3440	2260	2120	2630	4170	14600	326
Ryegrass		0	2160	1110	1200	1210	1250	6900	(120)*
LSD 5%		650	890	420	310	350	690	1900	
					1985				
		2/26	3/24	4/23	5/21	7/2			
Burton	10/9/84	3100	1940	2480	2360	3940		13800	277
75:25 mix		3350	2000	2220	2250	4030		13800	270
50:50 mix		3640	1980	2270	2120	4030		14000	268
25:75 mix		3900	1510	1840	1870	3550		12700	206
Ryegrass		3980	800	930	620	1060		7400	(174)*
Fahl		3530	0	0	0	0		3500	0
Miscawi-E		3150	2080	2050	1760	2500		11500	210
Bigbee		3840	480	2780	1360	3430		11900	235
LSD 5%		470	380	290	210	860		1500	
					1986				
		2/11	3/20	4/18	5/16	6/17	7/15		
Burton	10/7/85	1420	1690	1820	1860	3710	3390	13900	358
B&B mix		1530	1720	1920	1970	4070	3590	14800	396
Bigbee		1500	2060	1150	1400	4440	530	11100	311
Gigande		830	1480	1860	1840	3630	2170	11800	314
Khadraovi		1050	1310	1540	1510	3630	1100	10100	233
Belem		1420	1310	2250	2010	3890	2220	13100	353
Miscawi-E		1320	1110	1700	1360	2410	540	8400	236
Ryegrass		1160	730	50	170	590	370	3100	(45)*
LSD 5%		260	200	590	530	870	760	1800	

* Soil N uptake by annual ryegrass (not fixed). This value has been subtracted from each clover N uptake to estimate the corresponding N-fixed value.

PESTICIDE RESISTANCE AND ITS MANAGEMENT

T. A. Miller
Department of Entomology
University of California
Riverside, California

Insecticides have been used intensively in agriculture to control pests since the introduction of DDT, lindane (γ-BHC) and cyclodienes (chlordane, dieldrin, myrex) - just after World War II. For a number of years these chlorinated hydrocarbon insecticides plus later the carbamate and organophosphorus insecticides, represented the three major categories of insecticides used for pest control, and were hailed as near miracles as crops were kept nearly pest free (Hutson and Roberts, 1985).

Resistance to pesticides in pest insects had been a curiosity, but fairly soon after the introduction of synthetic organic insecticides, resistance became an accepted phenomenon and insecticides showed signs of having a limited life against some of the more serious pests. The development of new insecticides began to follow a predictable pattern as one product after another was used, discarded and replacements marketed by the agro-chemical industry.

This trend of new compounds introduced and tolerance gradually developed was documented recently for the leafminer (Parrella and Keil, 1984) (Table 1). Note the tendency for short active life when fewer materials were used at one time. With three products used between 1962 and 1974, tolerance took 12 years to develop. With one compound used between 1975 and 1977, tolerance was seen in two years.

By 1964 a public ground swell of opinion began to gain momentum. Called the environmental movement, and based in highly regarded and established societies such as the Sierra Club, this expression of public concern over abuse of the environment finally gained the legislative ear. The results were, among other things, unleaded gasoline, environmental impact reports, monitoring of air cleanliness and water quality to a degree never before attempted, and establishment of the Environmental Protection Agency, ending in the Super Fund - an expensive Congressional attempt to correct years of poor waste disposal practices.

Table 1. History of insecticide use to control Liriomyza trifolii in Florida. Modified from Parrella and Keil (1984).

Compound	Dates Used	Field Life (Years)
nicotine sulfate	?-1947	
chlordane	1947-1958	11
toxaphene	1947-1952	7
parathion	1948-1960	12
diazinon (Spectracide)	1958-1962	4
azinphos-methyl (Guthion)	1961-1974	13
dimethoate (Cygon)	1962-1974	12
naled (Dibrom)	1962-1974	12
oxamyl (Vydate)	1975-1977	2
permethrin (Pounce/Ambush)	1978-1980	1-
methamidophos (Monitor)	1976-?	

Pesticides were and still are used more-or-less as a rallying cry by environmentalists. The ban on DDT for most uses, an awareness that persistence in pesticides is undesirable, and attempts to regulate uses of pesticides have all made the development of pesticides an expensive and risky business, particularly given the finite life of some compounds due to resistance.

The search for acceptable products to fit the new criteria has made the discovery of new insecticides difficult, as Figure 1 shows.

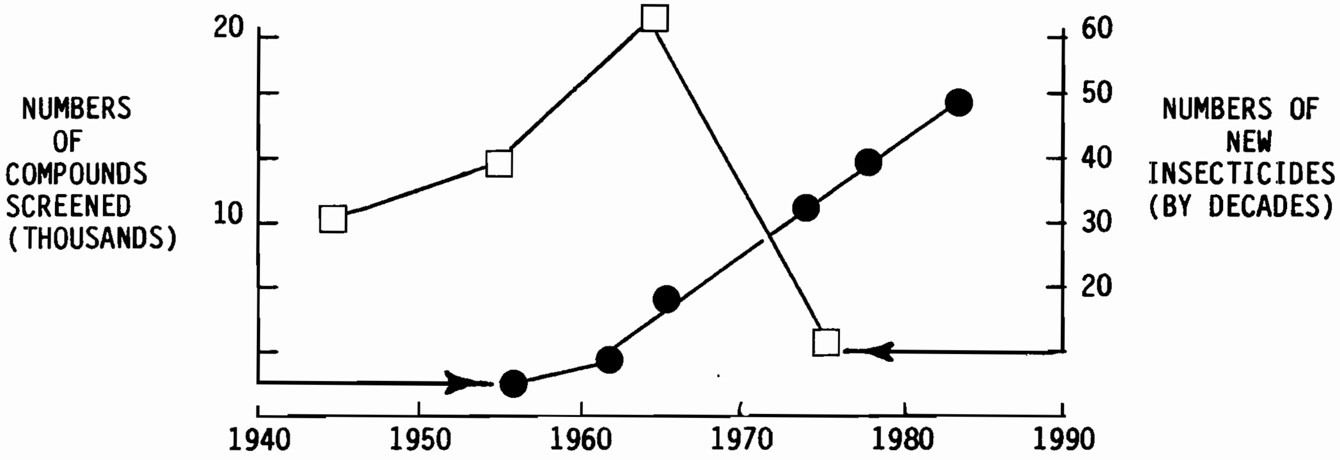

Fig. 1. The numbers of compounds screened to produce one commercial product (filled circles) over the past 45 years and the number of new insecticides registered every ten years since 1940. Taken from data supplied by Kurt Gubler (1983) of Ciba-Geigy and Julius Menn (1983) (then at Zoecon).

Since the three main categories of insecticides were developed about thirty years ago, only the pyrethroid insecticides as a large class of new compounds have been introduced, and that occurred about 1975.

Since introducing insecticides has become risky at best, the introduction of pyrethroids was done tentatively because some industry scientists were concerned that resistance would intercede before the development costs were paid from profits. There was a warning issued in 1978 (Elliott et al., 1978) that pyrethroids were known to induce resistance if used improperly. And, indeed, a great deal was known about resistance to pyrethroids since the toxicology of natural pyrethrins had been studied for over fifty years.

There has been some debate concerning the types of insecticide use strategies to use in preventing resistance. For example, the Australians restricted the use of pyrethroid insecticides to a 40 day period in the middle of the growing season in New South Wales and Queensland. The need for such drastic measures was called for by the occurrence of a particularly high tolerance to pyrethroids induced in Heliothis armigera in Emerald, Queensland in February 1983.

Others have recommended alternating insecticides or using several for control of a particular pest so that tolerance would have to be developed against several different chemicals (in effect repeating what is shown on Table 1 for leafminer control in the years 1962-1974). To see the reasons behind these solutions, one needs to understand the cause of resistance.

We now suspect that resistant insects can tolerate insecticides by developing one or more of several inheritable traits (Table 2). The first of these phenotypes, behavioral resistance, is difficult to measure. If this phenotype is tested by standard topical assay methods in the laboratory, insects do not show a difference in toxicity from susceptible strains, and incidentally, most resistance measurements are comparisons with standard susceptible strains.

Table 2. Factors responsible for developing a phenotype that is measured as tolerance to applied insecticides.

1. Behavioral resistance
2. Penetration resistance
3. Site-insensitive resistance
4. Metabolic (enzymatic) resistance
   (esterase, carboxyesterase,
   oxidase, transferase, etc., factors)
5. Others

Penetration phenotypes are somewhat easier to measure. Applying a given dose topically or by injection shows obvious differences in toxicity in the former and lack of difference with susceptible strains in the later. The remaining types are also measureable to one degree or another.

It has been stated many times that monitoring for the resistance phenotypes in crops is a critical element in devising resistance management strategies (Roush and Miller, 1986). It is clear that unique monitoring schemes must be designed for each pest individually, and that even then the type of treatment (by topical application, by feeding, by contact) may not show the resistance (Tabashnik and Cushing, 1986) to explain growing tolerance to field treatments.

In the end we are left largely to our instincts in designing strategies. Sparks et al. (1985) devised a list of factors affecting resistance (Table 3). We know, for example, that an insecticide when first applied in the field deposits a toxic residue on a crop; however, as the residue weathers, it eventually becomes a selecting dose, either driving resistant individuals away (behavioral) or killing only the susceptible members of the population. This selecting dose then leaves individuals with the ability to survive to produce the next generation.

In at least one case (Denholm et al., 1983), the residual property of permethrin was shown to induce resistance dramatically in a few weeks in houseflies in a pig barn. The nonresidual pyrethroid, bioresmethrin, did not induce resistance in two years of use under the same circumstances. Therefore, the residual property is important in developing resistance by providing the selecting mechanism.

Instincts are really the main element behind the Australian strategy which basically removes the pest from constant selective pressure. The Australian strategy was followed voluntarily because of widespread fear of the consequences of the loss of pyrethroids as tools in controlling *Heliothis armigera*.

Resistance management strategies can be thought of as methods of fine-tuning IPM or Insect Pest Management. For example, there are now successful examples of predators that have developed tolerance to field rates of given pesticides. Ironically, selective insecticides that would control pests and not harm predators and would therefore fit most compatibly into IPM are, by their nature, less valuable to companies since the greater the selectivity the more narrow the insecticide market becomes. As one company representative put it a few years ago: there is no market for an insecticide to kill mosquitoes because those areas with endemic malaria cannot afford the cost of insecticides.

Table 3. Conditions contributing to the rapid development of resistance. After Sparks et al. 1986.

<u>Operational</u>

1. Insect has a prolonged exposure to a single insecticide, or the insecticide is used in a slow-release form.
2. Every generation of the insect is selected.
3. Insecticide selection pressure is high.
4. No functional refugia, i.e. coverage by the insecticide is effectively complete so that no part of the population remains unselected.
5. A large geographic area is covered (i.e. all populations in a given area are likely to have been treated).
6. Selection occurs prior to mating.
7. Insecticide is closely related to one used earlier.
8. Low population threshold for the application of the insecticide.
9. Insecticide is inherently irritating and/or repellent.
10. A portion of the habitat is left untreated and is accessible to the insect.

<u>Biological</u>

1. No migration between populations.
2. Monophagous.
3. Short generation time.
4. Numerous offspring per generation.
5. Highly mobile.

Until now, registration procedures have been aimed almost exclusively at restricting materials, and allowing alternative materials to be used as an emergency when resistance occurs. We know now that such well-meaning procedures in many cases increase pesticide use, produce side effects such as predator mortality, force resistance to occur faster and do not allow for use of the most optional strategy in resistance management.

Attempts to deal with the resistance problem almost immediately forces one to consider pesticide use patterns. Restrictions on pesticide use have been imposed before, such as during the re-introduction of chlordimeform for cotton pest control in California in 1982, but heretofore it has been difficult to get growers and PCAs to voluntarily adopt sound resistance management practices. There is little doubt that each pest complex on each crop in each of California's diverse growing areas are unique and would have to be addressed or handled separately. No one scheme for resistance management would fit all cases.

State government can do a few things through creative legislation. It can provide funding through Agriculture Commissioners specifically for resistance monitoring of major pest complexes such as leafminer, Liriomyza trifolii; pink bollworm, Pectinophora gossypiella; tobacco budworm, Heliothis virescens; beet armyworm, Spodoptera exigua and citrus thrips, Thrips citrusi. There is every indication that the resistance problem is finally being dealt with rationally this year. What action will occur as a result is yet to be seen.

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Pesticide Resistance and Resistance Management in Plant Parasitic Nematodes

Michael V. McKenry
Associate Nematologist
Department of Nematology
University of California
Riverside, CA 92521

After forty years of soil fumigation experience there are no reports of nematode resistance to the halogenated hydrocarbon containing soil fumigants. There have been indications, however, of a loss of usefulness of several non-fumigant nematicides following repeated applications (1,4). There are also reports of soils with aggressiveness toward carbamate insecticide/nematicides applied to soil (3). Since biodegradation is a major event in the fate of certain soil applied nematicides (eg. fenamiphos), one can expect that some soils will be more aggressive to nematicides than others. Most nematicides are also degraded much faster in the presence of soil reactions above pH 7.

During the last four years we have studied the use of nematicides applied through dripper irrigation systems (2). This method of pesticide delivery provides management possibilities such as repetitive treatments and nematicide timing which have not been commercially possible among perennial crops. This system also provides a unique tool for the experimenter wishing to specifically study repetitive treatments with precision control *in vivo*.

In our studies in California vineyards a loss of the effectiveness of fenamiphos occurred where emitters were closely spaced (less than 60 cm between emitters), but did not occur where they were widely spaced (120 to 180 cm distances). Given the differences between both the absolute rates of toxicant in the sampling area and toxicant distribution patterns for closely versus widely spaced emitters, the following explanation is proposed. In the blanketed area where emitters are closely spaced, virtually all the nematode and flora population in the top 45 cm of soil are impacted and nematode control in this zone is initially good. However, the level of control decreases after about 2 years or about 10 kg/ha of active ingredient has been applied. This effect could be due to: 1) the selection for flora capable of rapidly detoxifying the toxicant, 2) the selection for plant parasitic nematodes capable of detoxifying or avoiding the toxicant effects, or 3) the combination of 1 and 2 above.

At the rates these nematicides are being applied to soil their major impact is expected to be in the disorientation, movement inhibition and hatching stimuli they promote in the nematode population.

The nematicides are promoting behavior modification to the detriment of the nematode. Direct lethal effects are expected in only a small portion of the treated soil profile. It is possible that nematode populations (egg stage through adult) could develop mechanisms for counteracting the effects of the nematicides. However, it is more probable that the diverse microorganisms of soil have developed mechanisms for metabolism of the active toxicant rendering them unavailable to the nematodes. Along this line of reasoning we have observed a slight loss of effectiveness among five of six nematicides tested within 2 years after first treatment. The loss of effectiveness comes faster when high treatment rates or close emitter spacing are utilized.

The new methods of nematicide delivery through a dripper irrigation system provides one of the best tools of nematode control ever available for established permanent crops. They also provide a unique tool allowing the experimenter to influence soil microflora and fauna and measure the eventual response in terms of microbe changes from site to site. Future research will be into the area of avoiding detoxification of nematicidal agents by the nematode or associated microorganisms. These studies will need to be complemented with studies of toxicant movement into the Vadose zone.

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HERBICIDE RESISTANCE

David W. Cudney

Extension Weed Scientist

University of California, Riverside - Cooperative Extension

Jodie S. Holt

Assistant Professor, Plant Physiology

University of California, Riverside

Background

Since the early 1900's, workers have been concerned about the resistance of pests to various pesticides. The problem was first noted with insects and has since expanded to plant pathogens and now to weeds. Harper, a well-known plant population biologist, theorized in 1956 that herbicide resistance in weeds might become a problem, but he had no evidence at that time. The first known case of herbicide resistance was found in 1968, when Senecio vulgaris, common groundsel, was shown to have a triazine-resistant biotype. These plants were found in areas that had received simazine applications over a period of approximately ten years. Since then, approximately 48 species have been found to have triazine resistant biotypes, including species of Brassica, Kochia, Chenopodium, Amaranthus, Setaria, Panicum, Digitaria, Poa, Bromus, and Echinochloa. Limited occurrence of resistance has also been noted with some other herbicides, including trifluralin, diuron, diclofop methyl, pyrazon, and paraquat.

The lack of herbicide response in plants of the same species can be divided into two groups. These groups are 1) those biotypes that are indistinguishable morphologically from their native populations and are not affected by herbicides even at elevated rates, and 2) those that exhibit a greater herbicide response as rates are elevated.

The first type of response has been noted in certain weed biotypes exposed to such herbicides as simazine, linuron, terbacil and trifluralin. These biotypes do not differ morphologically from their native populations. There appears to be true resistance in that the biotypes retain their resistance even when elevated rates of herbicides are used.

The second type of response is noted in individuals which show herbicidal effects at normal rates of application, but show reduced effects as rates of application are raised. This second type of response in plants is considered herbicide tolerance. Tolerance may be based on morphological differences between biotypes, including pubescence, cuticle thickness, growth habit, time of emergence, position of roots in the soil, etc. The tolerance to herbicides in these plants is because of reduced herbicide exposure through these morphological differences. Tolerance may also be based on physiological or biochemical differences between biotypes, including metabolism, absorption, translocation, uptake, etc.

True resistance is usually inherited on one or at most two major genes. In the case of triazine and phenyl-urea herbicide resistant weeds, the resistance is presumed to be inherited from the maternal chloroplast DNA. The resistance occurs due to a slight alteration of a 32 kilodalton protein which is in the light reaction center of the chloroplast membrane. This slight change, thought to be the substitution of just one amino acid, prevents the herbicide from binding to the protein and blocking photosynthesis. The slight change also reduces the photosynthetic efficiency of these biotypes making them less fit in native populations. The resistant biotypes are thought to be present at very low levels in native populations. When the selection pressure of continual herbicide use is applied, the frequency of the less fit herbicide-resistant biotypes increases. However, when herbicide use is discontinued, the resistant types no longer have a competitive advantage and the weed population will gradually shift back to its native, susceptible state. Thus, herbicide resistance, at least in the case of the triazine and phenyl-urea herbicides, poses a threat to agricultural production only in the case of continual use of a trazine or phenyl-urea herbicide. This illustrates the need for the rotation of herbicides and weed control measures.

A similar case of resistance has been noted where <u>Elucine indica</u> has been reported to be resistant to trifluralin where the herbicide had been used continually over a period of several years.

Where is Resistance Likely to Develop?

Resistance is likely to develop under conditions where a selection pressure is applied continually over a period of three or more years. Resistance is also favored with herbicides which have one mechanism of action (affect only one enzyme or protein). Thus, just one gene change can alter the enzyme or reactive protein and render the plant resistant. If an herbicide affects several plant processes simultaneously, like 2,4-D, true resistance would be much more difficult, if not impossible, to develop. Other factors favoring the development of resistance would include a large weed population with considerable variation or the presence of resistant mutants.

The current trend in the newer herbicides is towards those that affect specific plant metabolic pathways. This includes such herbicides as the imidazolinones, the sulfonylureas, and glyphosate. The advantages of the more specific herbicides is that they can be used at lower rates, thus reducing potential environmental impacts, and that they can target pathways that are plant specific and not found in animal systems. The disadvantage is that these herbicides are more vulnerable to the development of resistance.

Herbicide Resistance Could be an Advantage

The development of crop cultivars resistant to broad spectrum herbicides like the triazines or glyphosate has been the goal of several enterprising seed and chemical firms. The advantage of crop resistance would be that a triazine-resistant crop, such as soybeans, could be planted into an area with soil residual activity of an herbicide such as a triazine. In addition, glyphosate-resistant crops would render that herbicide selective, such that improved weed control and reduced production costs could be achieved. Two of the methods of incorporating herbicide resistance into crop cultivars involve recombinant DNA techniques and tissue culture in media enriched with elevated herbicide concentrations to take advantage of somatic cell variants. There is some optimism for the development of glyphosate resistant biotypes utilizing both methods.

Conclusions

Herbicide resistance at present is a problem in specific areas where one herbicide or group of herbicide analogs have been used continually over an extended period of time. More examples of weeds resistant to previously noted herbicides as well as weeds resistant to new herbicides are being reported each year. So far resistance has not posed a serious economic threat to agriculture. In the case of triazine and phenyl-urea resistance, the resistant weeds have been less fit and the weed populations have reverted back to susceptible biotypes when the herbicide selection pressure was removed. Herbicide resistance can be delayed or averted through the practice of rotating herbicides and weed control measures so as to avoid applying a continual selection pressure for a specific herbicide or herbicide group. Herbicide resistance in the case of crop plants could be a decided advantage.

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Strategies for the Utilization and Preservation of Resistant and Tolerant Plant Germplasm in IPM

R. K. Webster
Department of Plant Pathology
University of California
Davis, California

Introduction:

Control of plant diseases by genetic means has undergone extensive modification during the past 90 years. Early efforts were characterized by mixed cropping and use of land race cultivars with a fair degree of intra-specific diversity most likely resulting from subconscious selection toward polygenic resistance. This was followed by a period of emphasis on selection for monogenic resistance and the application of Mendel's laws to plant breeding. In some systems plant scientists have been very successful in obtaining long-term protection against disease by means of monogenic resistance. In others, results have been variable. In these cases where pathogens are highly variable and capable of adapting to intense cropping of cultivars with relatively narrow genetic backgrounds we see examples of the so-called vicious "Boom-Bust" cycle of pest control. The recent period was stimulated by recognition of and attempts to avoid the problems of short term control resulting in epiphytotics due to shifts in pest populations. It is characterized by (1) interest and attempts to develop cultivars with polygenic control of host-resistance and (2) systems of gene-management or deployment for extending and preserving the use of single resistance genes.

Meaning of disease resistance:

Numerous definitions for disease resistance have been offered. The commonality of them is that there are two kinds of resistance and that it is easier to characterize the kinds than to define them. The first is that resistance exhibited when the host resists the establishment of a successful parasitic relationship by restricting the pathogen at the infection site. Common terms describing resistance to infection include verticle, major gene and race-specific resistance (RSR). The second kind of resistance occurs when the host resists subsequent colonization and growth of the pathogen following successful infection, i.e. field, generalized, minor gene, horizontal and non-race specific resistance (NRSR).

Resistance of a host is characterized by the amount of disease that it sustains. Disease however, is not a character of the host, but rather is a product of the interrelationship of host and parasite under a given environment. A relative level of incompatibility between host and parasite usually results in a low level of disease. In fact, a low level of disease can be accounted for by two different phenomena: (a) either the host has a sufficient capacity to defend, or (2) the pathogen has an insufficient capacity to attack.

The notion that resistance should be discussed with its counterpart seems valid since resistance and susceptibility speak to the amount of disease sustained. Because they are relative terms, they are often used ambiguously. When for example is a plant resistant and when is it susceptible. The extreme perhaps being the classification of plants as moderately susceptible as compared to moderately resistant.

Origin and Population Dynamics of Pathogenic Races:

Analysis of population shifts of plant pathogens and the role that presence or absence of host resistance genes play in this phenomenon has received considerable attention in recent years.

Increases in frequency of virulent races and the concurrent "loss" of resistance of a host cultivar is usually associated with cases in which cultivars have been developed with single gene or race specific resistance. Based on current knowledge of the genetics of such cases, it is probable that a single host resistance gene can and often is overcome by a single gene change in the pathogen. Since virulence against a single gene is usually recessive in the pathogen, mutation to recessiveness renders a race virulent against a cultivar with a particular gene. This being the case, it appears that breeding for race-specific resistance has made variation worthwhile to plant pathogens. New races of plant pathogens arise by chance events, i.e. mutations, genetic recombination through conventional sexual processes, genetic reassortment via mitotic recombination or heterocaryosis etc. Host genotypes and fitness attributes influence the ultimate frequency or the sustained presence of new races, but not their origin.

Resistance Gene Management Strategies:

Control of diseases by using host resistance is successful to the degree that changes in the pathogen population towards increased pathogenicity are prevented or delayed. The various gene management strategies, or attempts to combat variable pathogens are or will be successful in attaining effective, lasting (or enduring) resistance to the extent that they successfully meet the challenge of continuous pathogen variation and selection for variants with increased virulence.

Use of Non-Race Specific Resistance Genes:

More often than not, polygenic resistance has been effective for long periods of time. The classical concept of NRSR is that resistance is effective against all pathogen genotypes. The stability of NRSR appears to rest on genetic probabilities, i.e. a new race would have to acquire several new genetic abilities to overcome resistance conditioned by several genes. As such, NRSR should be enduring and not prone to being overcome, at least as rapidly as RSR has been where only a single genetic improvement is theorized as needing to occur.

It has been argued, and rightly so, that we have not used NRSR to the extent we should have. Reasons for this include: (1) difficulties in breeding for polygenically controlled characters and the accompanying need for large population sizes, (2) difficulties in scoring or detecting increases in NRSR and (3) failure to incorporate epidemiological principles into disease screening programs.

Use of Race Specific Resistance Genes:

Use of single genes (RSR) for resistance has been the most popular approach for disease control. Single genes of large effects (restricting the infection site) are relatively easy to breed into already agronomically suitable cultivars, their effects are easy to detect and efficacy of disease control in the absence of virulent pathogen races is very impressive both production wise and aesthetically. These same single genes of large effects exert strong directional selection favoring new or low frequency pathogen genotypes with the ability to overcome the resistance bestowed by them.

Fears regarding continued use and reliance on RSR and dramatic examples of where it has failed due to occurrence and increase of new pathogen races are the basis for the current emphasis on genetic vulnerability of many of our major crop species. Nevertheless, since when RSR is operative it is so dynamic and effective there has been considerable research and theorizing to develop strategies whereby their use can be continued and still delay or prevent changes in pathogen populations toward increased pathogenicity and disease loss. Various gene management strategies based on epidemiological and genetic principles have been devised in attempts to meet the challenge of "dynamic" pathogens and preserve our option of using RSR genes. Experience thus far indicates that at least in some cases this will be possible.

Four primary strategies for deploying host genes for RSR have been suggested as potential means of curbing or avoiding population shifts in plant pathogens. These are multilines, geographical gene deployment, pyramiding RSR genes and composite breeding. Stabilizing selection sense Vanderplank would be another method and is by far the most controversial of gene management systems proposed.

Conclusion:

Fortunately, many species of cultivated plants posses adequate resistance to most of their parasites. If they do not, their wild relatives do, and in many cases we can take advantage of this. Some of the main questions we are faced with today are the identification and preservation of useful germplasm and how we can best use the resistance we find. It is inevitable that new races of plant pathogens will continue to appear from time to time and they will do so irrespective of the relative resistance or susceptibility of currently grown cultivars. The challenge is to find and practice the best use of our resistance genes that will negate the selection and consequences of newly appearing races.

EXPERT SYSTEMS AND ARTIFICIAL INTELLIGENCE IN INTEGRATED PEST MANAGEMENT

R. E. Plant
Professor
Departments of Mathematics and Entomology
University of California
Davis, CA 95616

The possibility of using expert systems technology in agricultural applications, and in particular, in integrated pest management, has attracted widespread (one might say explosive) interest in the last year or two. This report will review the basics of expert systems and how they are being applied to IPM problems in California. It will also discuss some possible future directions for expert systems research in agriculture.

An expert system is, fundamentally, a computer program which is used to provide advice or information in a manner similar to that which would be provided by a human expert on the subject. Not all computer programs that do this are expert systems, however. Indeed, almost any computer program provides information in the manner of an expert. A linear regression program, for example, may be said to give the same answer to a data fitting problem that a statistical consultant would.

There are two features that distinguish expert systems from other computer programs that perform a superficially similar function. These are:

1) An expert system can make use of "symbolic" as well as numerical information in formulating its solution to a problem. This

information, which is called "domain specific knowledge," is kept in a part of the system called the "knowledge base."

2) The part of the program that manipulates the domain specific knowledge is kept separate from, and independent of, the knowldege base. The manipulative part of the program is called the "inference engine."

The type of expert system in most common use is called a "rule based" expert system. In this the knowledge base, also called the "rule base," consists of a collection of rules called "production rules." In the simplest case these rules have the form

IF <antecedents> THEN <consequents>.

The antecedents and consequents are each collections of statements. An example of a production rule for automobile fault diagnosis might be: IF <car won't start> <headlights dim when ignition turned on> THEN <battery is low>. This example also illustrates what is meant by "symbolic" as opposed to "numerical" information. Symbolic information is simply such things a English sentences.

The inference engine in a rule based expert system begins with a collection of "hypotheses", or "goals". Each goal must be that consequent of at least one rule. The inference engine attempts to determine which goal or goals are true by forming chains of rules in which the antecedents of one rule in the chain are known to be true, and one or more consequents of that rule are the antecedents of the next rule in the chain. There are two commonly used methods of inference, called "forward chaining" and "backward chaining."

An expert system intended for use as an aid in a task such as managing a crop is called an "expert decision support system" or sometimes an "integrated expert system." Such systems must be more complex and sophisticated than the inference engine and knowldege base described above since they must keep track of in season crop, pest, and weather information and must integrate several knowledge bases together to make a recommendation. An expert decision support system called CALEX is being developed for the University of California Statewide Integrated Pest Management Program. This report will briefly describe some of the features of the CALEX program.

CALEX is a shell program that is combined with modules developed for a specific crop to create an expert decision support system for that particular crop. Development is currently under way for two crops, cotton and peaches, and other packages will be developed in the future for other crops. In its present version, CALEX is set up to provide information in one or more of the following categories: pest management, agronomic management, economics, and symptom diagnosis. Presently, CALEX/Cotton provides pest and agronomic management information and CALEX/Peach provides symptom diagnosis. This report will briefly describe the software architecture of the CALEX package and how it works.

Prototype versions of CALEX/Cotton and CALEX/Peach will be field tested during the 1986 crop season. At the same time, development will be under way of a version that will provide integrated, in season management recommendations for the whole crop, incorporating each of the specific categories as well as projections of the crop development.

Using Expert Systems for Irrigation Management in Cotton

Lowell J. Zelinski
Farm Advisor
University of California
Cooperative Extension
Fresno, California

Introduction

Incorporating the knowledge of experts into machines has been practiced for the last 15 years. The science of artificial intelligence has tried to develop machines and algorithms which mimic intelligent action. Recently, this technology has moved from research into practical applications, and the University of California is developing a cotton crop/pest management expert system. This paper is a discussion of the irrigation component of that system.

Expert Systems

Expert systems are computer programs which emulate the decision making process of human experts. They can make decisions which rely on judgment and the decisions can be tested by heuristics, or trial and error procedures.

There are many examples of expert systems. Some of these include: TAXADVISOR, a program which gives advice on estate planning (Michaelsen, 1984); MYCIM, diagnoses infections in humans (Shortliffe, 1976); and PROSPECTOR,which aids geologists in mineral site evaluation (Duda et al, 1979).

There are many forms of expert systems, some of which are very general, and some more specific. The structure of the cotton management systems uses a 'rule based system', and is generally set up as follows:

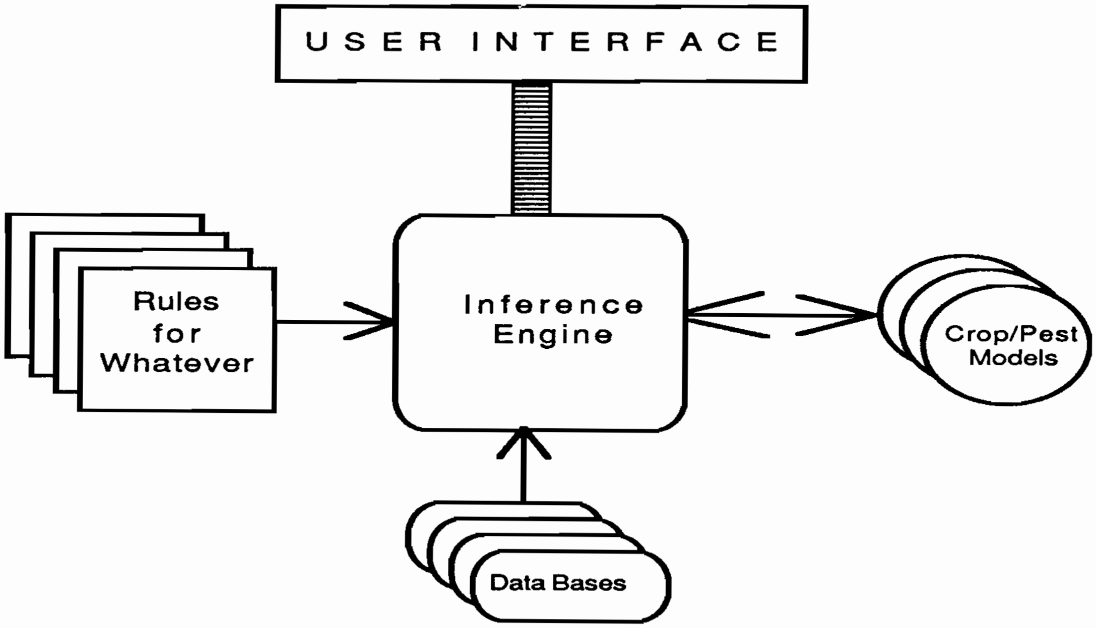

Figure 1. Generalized structure of rule based expert systems.

Expert systems exist in agriculture, but few have been implemented at the farm level. Cotton has received considerable attention in the expert system area with four models currently under development and testing. These include: GOSSYM/COMAX, (Baker, 1986) under development at the USDA ARS Crop Simulation Unit at Mississippi State; COTFLEX, (Lieth, 1986) under development at Texas A&M for cotton production in Texas; SIRATAC, (Wilson, 1986) developed and implemented in Australia with strengths in pest management and record keeping; and CALEX, under development at the University of California. All of the expert systems in cotton are interdisciplinary, and each has particular strengths and weaknesses.

Cotton expert systems must acknowledge that many components of cotton production are interrelated. A decision in irrigation, insect control, etc., can have effect on other areas of cotton production. Many of the effects are not known quantitatively, but are significant nonetheless. Growers make decisions daily in the absence of quantitative information, and expert systems can provide expert advice on these types of decisions. If the expert system is linked to realistic simulation models, decisions can be hypothesized, tested, modified, and suggested, all within a short period of time.

Cotton Irrigation Management

Irrigation management of cotton seeks to direct growth into reproductive tissues (lint and seed). This is a complex task requiring optimum timing and amounts of irrigation (Stromberg, 1978). The criteria which affect irrigation timing are modified continuously throughout the season. For irrigation management, cotton growth can be broken down into three different periods; early, mid, and late season, (Grimes and El-Zik 1982).

Early Season

Many factors interact early in the season to modify timing decisions. Some of these include: temperature of the air and soil, disease inoculum levels, variety, leaf water potential, soil texture, and soil fertility status. For 'normal' conditions, the following dates for first irrigation are suggested:

Texture	First Irrigation Date
Sandy loam	June 2
Loam	June 6
Clay loam	June 10
Clay	June 15

The pressure bomb, and soil water depletion are also methods of determining the timing of the first irrigation.

Mid-Season

Irrigation management during the mid-season seeks to promote just enough vegetative growth to provide sufficient fruiting positions for optimum yields. Several methods are used to schedule irrigations. Historical, weather based, soil based and plant based measurements are all currently used. These approaches are myopic however. They are concerned only with irrigation, and related factors, such as economics, fertility, insect pressure, etc., are ignored.

Late Season

Timing the final irrigation is also an important management decision. If all is going 'normally' the following dates (based on historical conditions, and soil type) are recommended:

Texture	Final Irrigation Date
Sandy loam	Aug. 23
Loam	Aug. 17
Clay loam	Aug. 10
Clay	Aug. 1

'Normal' seasons rarely exist. Modification to these final irrigation dates are often required. How decisions are made to modify these dates, and the magnitude of the modification, require the synthesis of large amounts of interrelated data. This synthesis is one thing we hope to model in CALEX.

The Marriage

How can expert system technology be applied to cotton irrigation management? Initially the irrigation component of CALEX will be an enhanced version of the California Irrigation Management Information System's irrigation scheduling program (CIMIS), (Synder 1983) . The CIMIS program uses the water budget approach to irrigation scheduling, which relies on weather parameters and a modified Penman equation to estimate water use.

A major improvement which will be made by CALEX this first year is changing the basis for determining the crop coefficient (Kc). The crop coefficient is a major component of crop water use estimation and is multiplied by the reference evapotranspiration (ETo) to get the crop ET (ETc), or:

$$ETc = Kc \times ETo$$

Currently, in the CIMIS model, Kc is a function of days after planting, and it assumes a 'normal' year. In years that are warmer or cooler than 'normal' the Kc value will be in error. The CALEX irrigation scheduling package will estimate Kc values from degree day information collected from the same weather stations used to generate the ETo information.

Eventually the following information will be included:

1) The Kc values will be more closely related to plant growth. Though degree days help to estimate stage of growth, nothing can substitute for field observation. These field observations of plant height and stage of growth will be used to modify Kc values predicted from degree days.

2) The incidence and severity of Verticillium wilt, the major cotton disease in the San Joaquin Valley, is affected by irrigation and temperature. Devay, Grimes, and Husiman (Devay, 1982; Grimes and Husiman, 1984) have done considerable work to estimate the interactions, and this information will be incorporated into CALEX.

3) Recommendation for the timing of the final irrigation is based on a 'normal' season, and a 'normal' fall. These recommendations can be improved by the field observations of growth stage and maturity, information on soil type and salinity levels, and coordination with the harvest timing of various fields.

4) Cotton yields are influenced by soil fertility, irrigation management and an interaction between the two. With the aid of an expert system, the interaction can be exploited to provide the most cost effective strategy for cotton production.

There are many other interrelationships which modify cotton irrigation decisions. As information about these relationships is developed they too can be incorporated into CALEX.

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ABSTRACT

Toward an Understanding of the Environmental Fate of Pesticides

Until just the past 10-15 years, what we knew about the environmental distribution and disposition of pesticides was learned in retrospect, by analyzing samples of the the affected soil, water, and biota in the general vicinity of pesticide use. A more prospective approach, including modelling and prediction, is now possible based on the accumulated information from environmental analyses, combined with physical and chemical principles. If one knew such basic properties as water solubility, partition coefficient, vapor pressure, and hydrolytic half-life, a reasonable prediction of fate may be made. This notion is embodied in the EPA environmental fate testing guidelines for registration of pesticides, and in the AB 2021 legislation recently passed in California. An evaluation of our ability to predict environmental fate, including some applications and shortcomings will be presented.

THE RAPID DEPLETION OF NITROGEN APPLIED TO TURFGRASS

D.C. Bowman, J.L. Paul, and W.B. Davis
Department of Environmental Horticulture, University of California,
Davis, CA 95616

S.H. Nelson
Department of Horticulture Science, University of Saskatchewan,
Saskatoon, SK S7N 0W0, Canada

INTRODUCTION

Nitrogen is the fertilizer nutrient most heavily-used by managers to control both appearance and growth of turfgrasses. Inorganic sources of N such as ammonium and nitrate salts are commonly used because they are relatively inexpensive and provide a rapid greening. Turf response to a typical application of 1# inorganic N/1000 sq ft. may be evident in 3-5 days but last less than 4 weeks.

The fate of N fertilizer during this short period is complicated and not completely understood. Obviously the plant is the target of applied N, via uptake by the root system. However, prior to absorption by the plant, applied nitrogen will be subject to a number of different processes which compete for the nutrient. Leaching, gaseous loss (volatilization and denitrification), and microbial immobilization all may, in effect, rob the turf plant of nitrogen. These processes depend on the presence of inorganic nitrogen; consequently that period when fertilizer N remains free in the system is crucial to its fate. The faster a root system removes N from the soil, the less likely is its loss to competing mechanisms. This study was undertaken to examine the fate of inorganic N, particularly the kinetics of its depletion from the soil, following application to a turf system.

MATERIALS AND METHODS

SITE. Field studies were performed in July and August, 1985, on experimental turf plots at the Univ. of Calif., Davis. The three species used in this investigation were Kentucky bluegrass 'Columbia' (Poa pratensis L.), perennial ryegrass 'Pennant' (Lolium perenne L.), and tall fescue 'Mustang' (Festuca arundinaceae Schreb.). All were mowed at 5 cm and irrigated to meet evapotranspirational demand. Kentucky bluegrass had a thatch layer approximately 1.5 cm in depth; the other species had little or no thatch. The soil was a well-drained Yolo loam (Typic Xerorthents) with a pH of 7.3.

EXPERIMENTAL PROCEDURE. The following methodology was developed to follow the rate of depletion of inorganic N from the turf-soil system following application: nitrogen fertilizer was applied in solution to 1 m^2 turf plots using a hand-held pressurized sprayer to provide a uniform distribution. Either $Ca(NO_3)_2$ or $(NH_4)_2SO_4$ was spray-applied at a rate of 5 g N m^{-2} (equivalent to 1# N/1000 sq. ft.) dissolved in a volume of 2 l m^{-2}. This was followed immediately by spray-applying 3 l m^{-2} deionized water to remove the N solution from the foliage and position it in the thatch and soil. Control plots received 5 l m^{-2} deionized water. A completely randomized design was used with 3 replicate plots of each of the 3 treatments.

Depletion of the applied inorganic N was followed by a soil sampling procedure. Sampling commenced 30 minutes after application and consisted of 3 cores per plot obtained with a standard golf green cup cutter to a depth of 10 cm. Cores were extracted with 2 M KCl and the NO_3-N and NH_4-N content of the extract determined. Sampling was discontinued when the extractable inorganic N content reached control values.

NITROGEN POSITION. The vertical position of applied N in the turf-soil profile was determined for both NO_3 and NH_4 30 minutes after application. Cores were sectioned by depth into foliage, thatch, and soil from 0-1, 1-2, 2-3, 3-5, and 5-10 cm. Inorganic N was then determined in a KCl extract for each section. Root density with depth in the soil was estimated by the line intersect method.

PLANT N ABSORPTION. ^{15}N-labelled $(NH_4)_2SO_4$ was used to quantify the N incorporated by the plant. Cores samples were taken 4 days after N application. Total biomass, Kjeldahl N, and % ^{15}N enrichment were determined on both shoot (above the soil line) and root material after washing away the soil.

VOLATILIZATION. Volatilization was measured by a chamber trapping method. Two liter plastic containers were placed upside down on the turf plots, covering an area of 167 cm^2, for 15 minutes every 2-4 hours. Air movement inside the chambers was provided by a battery-powered fan mounted through the top. Ammonia gas was captured in filter paper traps clipped to the inside of the chambers and wetted with 0.1 N H_2SO_4. After a sampling period the traps were extracted with 2 M KCl and analyzed for NH_4-N.

RESULTS AND DISCUSSION

Both NH_4-N and NO_3-N were positioned principly in the thatch and 0-1 cm of the soil in the profile following application to Kentucky bluegrass (Table 1). Very little N from either source remained on the foliage or leached below the first cm of soil. It should be noted that this distribution was the result of N applied and irrigated with a total depth of 0.5 cm water. Distribution in other turfs would likely vary depending on turf condition and amount of water applied.

DEPLETION. The depletion of inorganic nitrogen from the Kentucky bluegrass turf-soil system following application was found to be very rapid with a loss of over 60% of the applied N in the first 8 hr (Fig. 1a). There was a trace of N remaining at 24 hr but by 48 hr all of the applied N had been depleted. Inorganic N content of cores taken below the 10 cm sampling depth was the same as from control plots and confirmed that this depletion was not the result of leaching.

The very rapid initial loss was followed by a gradually decreasing rate. Both forms of N were nearly identical in the pattern of their depletion, with the data fitting an exponential decay curve, although ammonium disappeared at a slightly faster rate than NO_3. These rates of depletion, in which a typical application of 5 g N m^{-2} is essentially gone in 24 hr, are considerably faster than values commonly encountered with field-grown crops.

Depletion of N applied to both turf type tall fescue (Fig. 1b) and perennial ryegrass (Fig. 1c) followed patterns very similar to the bluegrass with the rate being slightly slower in the ryegrass. However, nearly 90% of the fertilizer N was lost from the ryegrass system after 48 hr with essentially complete depletion by 72 hr. The rates of disappearance were nearly identical for NH_4-N and NO_3-N in the perennial ryegrass, while NH_4 was depleted from the tall fescue plots at a somewhat faster rate, as observed with Kentucky bluegrass.

VOLATILIZATION. Volatile loss was measured over the first 24 hr following N application. Less than 3% of the applied N volatilized from plots fertilized with $(NH_4)_2SO_4$ with most of the loss occurring in the first 12 hours. Denitrification was not measured from $Ca(NO_3)_2$-treated plots. However, the extremely rapid loss of N from the system combined with the similarity in rate of depletion of the two N forms and the well-drained nature of the soil argue against denitrification contributing significantly to the observed depletion.

RECOVERY. The initial recovery of applied N in the extract at 30 minutes was always considerably less than 100%, averaging 80% over the 2 nitrogen forms and 3 species examined. It is believed that this discrepency is at least partly due to biological immobilization; the very rapid loss measured between 30 min. and 4 hr supports this. It is also possible the N may have been rapidly absorbed directly by the turfgrass foliage and shoot tissue immediately following the initial N solution spray but before complete removal by the deionized water spray.

High variability in tissue biomass (data not shown) precluded estimating plant uptake by Kjeldahl tissue analysis. It was found, using ^{15}N-labelled $(NH_4)_2SO_4$, that shoot and root tissue contained 75% and 3% of the fertilizer N, respectively, 4 days after application. This indicates that the rapid depletion of a NH_4-N applied to Kentucky bluegrass is principly the result of uptake by the turf. Less than 5% is lost as gaseous NH_3 or N_2O with the remainder probably immobilized by soil microorganisms. Leaching does not contribute to the loss with proper irrigation management.

Two factors are thought to be involved in the rapid absorption of fertilizer N by turfgrass. First, fertilizer N is positioned in layers with extremely high root length densities, RLD, (Table 1). Thatch RLD was found to be in excess of 700 cm cm^{-3}. This high density in both the soil and thatch represents a large absorbing surface and minimizes the mean distance between roots. Consequently the distance an ion would have to diffuse to reach a root is extremely small; ignoring the presence of root hairs it is estimated that in the surface 0-1 cm of soil this distance is approximately 0.02 cm (200 microns). This may explain why the relatively immobile NH_4 ion is depleted at rates similar to the much more mobile NO_3 ion. Second, the turf used in this study was, to a degree, N deficient and might therefore have developed an increased capability to absorb N. Increased absorption capacity in response to nutrient deficiencies has been reported for a number of species.

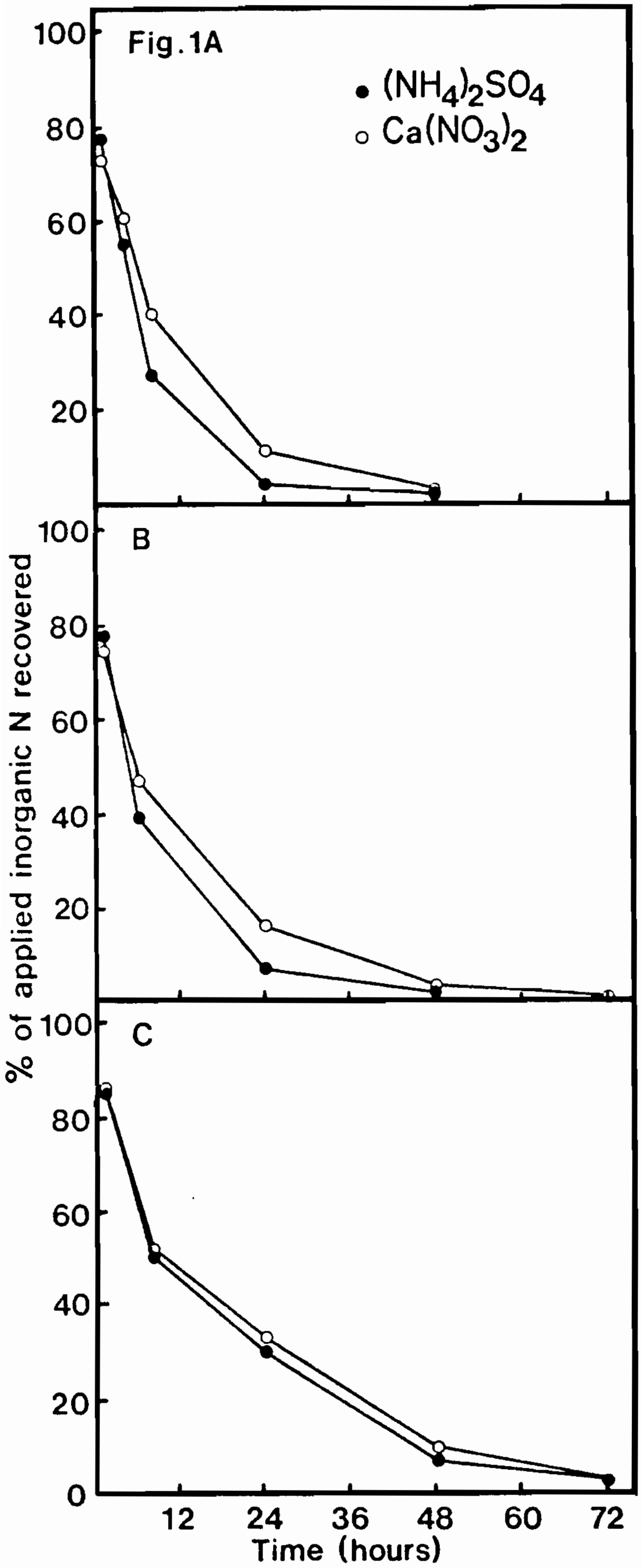
To test the possibility that rapid N uptake is related to N deficiency in a turf system, Kentucky bluegrass was heavily fertilized with a total of 25 g NH_4NO_3 applied in 5 equal portions over 3 weeks. Control plots were unfertilized. Total N in fresh leaf material from fertilized and control plots was 4.8 % and 3.15 %, respectively prior to the experiment. Depletion of both NH_4-N and NO_3-N by N-deficient turf (Fig. 2) was similar to previous experiments. Nitrogen-fertilized turf absorbed NH_4-N somewhat more slowly although the N was still depleted within 4 days. Nitrate depletion however followed very different kinetics. After a brief initial rapid loss, it disappeared much more slowly over a 9 day period at a more or less constant rate. Nitrogen uptake thus appears, at least in the case of NO_3-N, to be regulated by the N status of the tissue.

CONCLUSIONS

Both NH_4-N and NO_3-N are very rapidly depleted from a turfgrass system following application. It is speculated that this is primarily the result of plant uptake, facilitated by the high density of roots and deficient N status in the turf. Because of this very rapid uptake, very little fertilizer N should be lost, at least in the short term, to other processes.

Table 1. Percent of applied N recovered and root length density as a function of depth in the turf-soil profile for Kentucky bluegrass. Values are means of 3 samples ± standard deviation.

	Applied N % recovery		Root Length Density
N Source:	$(NH_4)_2SO_4$	$Ca(NO_3)_2$	cm/cm3
Layer:			
Foliage	3 ± 0	3 ± 0	
Thatch	48 ± 7	30 ± 2	
Soil 0-1 cm	35 ± 8	36 ± 2	410 ± 8
1-2 cm	2 ± 1	5 ± 3	270 ± 12
2-3 cm	1 ± 0	2 ± 1	154 ± 16
3-5 cm	1 ± 1	3 ± 2	122 ± 13
5-10 cm	3 ± 2	4 ± 2	95 ± 8
Total recovery	93 %	83 %	



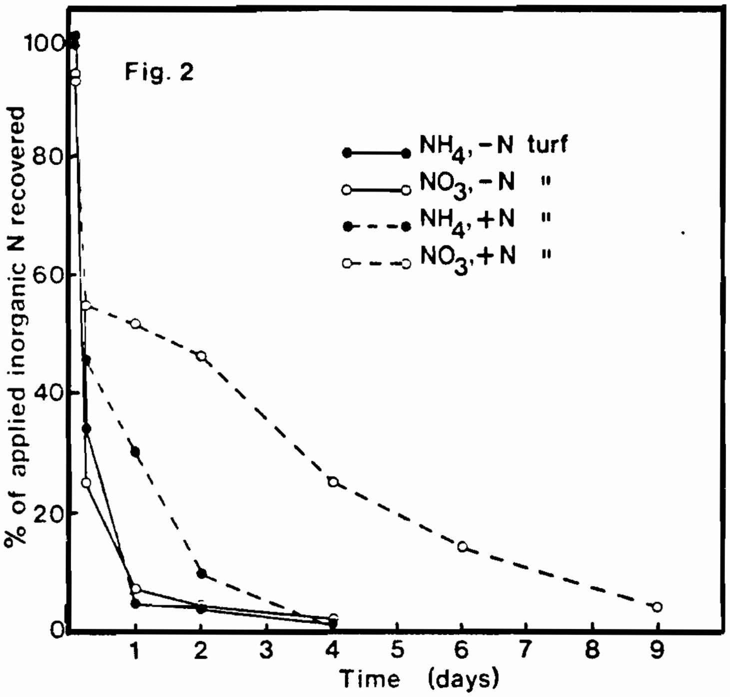


Fig. 1. Depletion of inorganic N applied as either ammonium sulfate or calcium nitrate to Kentucky bluegrass (Fig. 1A), tall fescue (Fig. 1B), and perennial ryegrass (Fig. 1C). Values are means of 9 samples.

Fig. 2. Depletion of inorganic N applied as either ammonium sulfate or calcium nitrate to N-sufficient and N-deficient Kentucky bluegrass. Values are means of 3 samples.

PREDICTING PHYSICAL AND CHEMICAL PROPERTIES OF CONTAINER SOIL MIXES

J. R. Jenkins
Consultant
Soil and Plant Laboratory, Inc.
352 Mathew Street
Santa Clara, California 95050

Soil, usually high in sand, is often one of the basic components in container media for many outdoor nursery growers and research institutions. It is often amended with organic and mineral materials to improve physical and chemical characterisitics to meet the demanding needs of container plant culture. Physical properties of primary importance include bulk density, total and air-filled porosities, water retention and saturated hydraulic conductivity (K_{sat}). Critical chemical properties are pH and cation exchange capacity (CEC).

Though these properties are considered important and practical for routine testing, there have been few attempts to determine the mathematical relationship between properties of a mixture and the component properties. There is a need for a simple, accurate method to create the ideal mixture for a container grower's particular situation using readily obtainable components. In answer to this need, a straightforward mathematical model was formulated to predict potting mixture properties:

$$\text{Mixture property} = \sum_{i=i}^{n} (\text{component volume ratio}_i)(\text{component property}_i),$$

where n=number of observations and i=observation index.

The ideal container mixture has properties which most closely match the needs of the crop at the least cost. This model may allow container mix producers and users to custom-formulate suitable mixtures more cheaply and with less trial and error.

This study was undertaken to gain data, along with published values, to test the hypothesis that each mixture component proportionally adds its own property to that of the mixture. Previously published data, as well as original data, were used to test the model hypothesis. Reported results are from graduate research in the Soil and Environmental Sciences Department at the University of California, Riverside.

Comparison of Predicted and Measured Values

Physical and Chemical Property Measurement

Twenty experimental mixtures were prepared consisting of different combinations of sandy loam soil (Typic Xerothent), sand (Typic Xeropsamment), white fir bark, and perlite. Each component and mixture were tested for bulk density, total and air-filled porosities, container capacity, available water, saturated hydraulic conductivity (K_{sat}), pH and cation exchange capacity (CEC). Multiple regression was used to determine the relationship between the measured properties and the mixture components. To compare the measured and predicted data, linear regression, ratio of the average predicted over the average measured data, and average percent difference between individual observations were used.

Current Experiment Comparison

There was good agreement between predicted and measured values for bulk density, total porosity, container capacity and available water. Air-filled porosity, for the sandy loam and sand mixtures, and K_{sat} were greatly over-predicted by the model. Both pH and CEC have highly significant regression coefficients and predicted to measured value ratios close to unity for all the mixture groups. Values predicted by the model deviated relatively little from the measured values.

Published Research Comparison

To determine if the model were valid for published data, additive model predicted values were evaluated using the comparative indices. A diverse array of components had been used in the published studies including clay loams, sands, perlite, sawdust, and peat mosses. For bulk density, the model tended to slightly under-predict results with little deviation from measured values. Averages of the predicted and measured values are close. The average predicted total porosity was within 4.6% of average measured total porosity. No significant interaction between components was found. The predicted to measured ratio for air-filled porosity from the data gathered in this experiment was substantially greater (1.56) than the previously published research (1.03). A scatter diagram of the air-filled porosity against total porosity indicated that much of the deviation from linearity is associated with media total porosities below 57%. This value coincides with the lowest mixture total porosity reported in the published studies, which was 57%. Deleting experimental air-filled porosity values of mixtures below 57% total porosity reduced the overall predicted to measured ratio from 1.56 to 1. 24. Model prediction of container capacity and available water literature data was good, though a trend was indicated for slight under-prediction of available water and a higher deviation as compared to the container capacity prediction. Insufficient data was found to effectively evaluate model prediction of K_{sat}, pH, and CEC. Though researchers often determine these properties for mixtures, the values of the property for individual components are often not reported.

Conclusions

The results of this study indicate that the additive model is valid for several important physical and chemical of container mixtures. Component interaction was not significant in most cases, even though a wide range of typical components were analyzed. Model prediction was unsatisfactory for K_{sat} and air-filled porosity for mixtures with total porosity below 57%. For prediction of these properties, the appropiate multiple regression equation could be utilized if the mixture components are of similar type.

CAPITAL AND MANAGEMENT INTENSIVE SYSTEMS FOR DESERT VEGETABLE PRODUCTION

Alfonso Durazo III
Farm Advisor
University of California Cooperative Extension
Imperial & Riverside Counties

Specialty vegetable crop producers in the Colorado Desert areas of Southern California have been using intensive growing systems to push crops to early market windows for over fifty years. Traditional practices included the use of peaked beds running east and west with a 45° angle on the south side. This south-sloped bed provides maximum exposure to the sun, which is in the southern part of the sky during winter months. An even earlier market window is reached by erecting craft papers and redwood lath heat traps on top of these beds. This method further increases soil temperature on the south side of the bed, as well as providing frost protection while the plants are small.

Over the past six years, new, innovative technologies developed in Israel have begun to replace the old systems of intensive production in the desert. These new systems may include drip irrigation, plastic mulch, row covers and bed fumigation.

In the Coachella Valley, bell peppers are traditionally direct seeded on ground that has been fumigated with methyl bromide in October. The lath and paper heat traps are erected, the seeds germinate and grow slowly through the winter. After the threat of frost is over, the plants are thinned to a ten inch in-row spacing. This method provides for an early April harvest.

With the new system, bed tops are fumigated only, and the plastic is utilized as a mulch to heat the soil during the growing season. Thus the plastic is not wasted, as with the broadcast method. Fumigation is accomplished with methyl bromide injected into the beds or metham applied through drip irrigation lines. Transplants are planted through holes in the mulch, in February, for harvest in mid to late April. Yield increases of five to eight hundred boxes per acre have been achieved on well managed fields. Furthermore, the ground is not tied up for the fall season, as with the old system, and double cropping is possible. Other crops currently being produced under these intensive systems include melons, eggplant, cucumbers, squash and tomatoes.

These components are expensive, thus the term capital intensive. The high initial expense is one of the obstacles a grower must deal with when converting to the new systems, but the biggest deterrent to their conversion is the intensive management that must be followed in order to achieve optimum results from these different components.

This paper will address some of the management practices that have been developed in the desert to maximize production under capital and management intensive systems.

VEGETABLE CROP MANAGEMENT INTERACTIONS WITH PLANT DISEASES

A. O. Paulus
Extension Plant Pathologist
Plant Pathology Department
University of California
Riverside, California

Moisture and temperature are two of the most important factors in the development of fungal and bacterial diseases in California. Warm-season crops grown in California during the summer-dry period, under furrow irrigation, are generally free from wet-weather diseases. Diseases such as halo blight and anthracnose of beans, early blight of tomato and bacterial spot of tomato disappear in successive plantings maintained in the warm dry climate of the coastal and interior valleys of California under these conditions. Bacterial spot of tomato and pepper caused by the bacterium, Xanthomonas campestris pv. vesicatoria, is rarely found in California but causes severe epidemics in Florida. Florida experiences periods of continuous rainfall and heavy dews and bacterial spot becomes rampant.

In recent years because of labor costs some crops have been grown in the summer under sprinkler irrigation. In effect, such crops are not longer being produced in an arid climate. The humid conditions and freewater provided by overhead sprinkling convert these fields into humid, "rainfall islands" well suited to the infection, spread and development of wet-weather diseases. Disastrous losses have occurred under sprinkler irrigation where infected seed from outside sources has been used. For example, striking contrasts in the field development of haloblight of bean have been observed where the same lot of infected seed was sown in two nearby fields with one under furrow irrigation and the other overhead sprinkling. Before pod development was completed, the field under sprinkler irrigation was a total loss from blight. The other field showed no evidence of blight at harvest (4).

Bacterial speck of tomato caused by the bacterium, Pseudomonas syringae pv. tomato, is usually only important in the spring following early rains. Consequently, an early application of a copper fungicide reduces the inoculum potential of the bacteria on the plant and subsequent sunny days without accompanying rainfall reduce the disease to a level of uneconomic importance. Later plantings to escape the rainfall periods would also minimize the effects of bacterial speck.

Late blight of celery caused by the fungus, Septoria apiicola, is a serious economic problem during the rainy fall and winter months. Spore germination and penetration of the plant requires a relative humidity above 90% for about 2 days or free moisture for at least 24 hours. Some celery growers have used sprinkler irrigation because of high labor costs and occasionally late blight can be a problem in some of the non-rainfall times of the year. If at all possible and economically feasible growers should furrow irrigate since this practice keeps the foliage dry during the nonrainfall periods and inhibits the development of late blight.

Buckeye rot of tomato caused by the fungus, Phytophthora parasitica, can cause serious economic losses to tomato growers in northern California. The incidence of fruit infections during the final days before harvest in 1980 was significantly higher after a 4-day interval between irrigations than after a 25-day interval between irrigations. In 1982, irrigations were applied every 4, 8, 16 or 32 days throughout the last 74 days of crop growth. The rate and incidence of infection of fruit sampled during the final irrigation increased significantly as the interval between prior irrigations was decreased. Furthermore, the final incidence of infection on fruit grown in the field increased from 24 to 48% as irrigation frequency increased from once every 32 to once every 4 days. Yield of healthy fruit declined as irrigation frequency increased from every 16 to every 4 days. The results suggest that the inoculum of P. parasitica, probably in the form of zoospores, is formed more rapidly and abundantly when previous irrigations have been frequent and have not allowed soil to dry extensively (2).

Tomato and pepper planted on high beds and on soil with excellent internal drainage tend to have less yield loss from the effects of Phytophthora root rot that those planted on low beds and soils with poor internal drainage. Careful attention to keeping the top of the beds dry near the end of the growing cycle helps to alleviate the problem of pink rot of celery and drop of lettuce caused by Sclerotinia sp.

SOIL REACTION AND CHEMICAL APPLICATIONS. Club root disease of crucifers caused by the fungus, Plasmodiophora brassicae, is an economically important disease in San Mateo, Santa Cruz, Monterey and Santa Barbara counties. Infected plants become stunted, bluish in color, and may wilt slightly during the warm part of the day. Roots become enlarged with knots or spindleshaped swellings called clubs. The disease is favored by wet soils with a neutral-to-acid reaction and is seldom important on soils at pH 7.3 or above. Clubroot in the Salinas valley has been controlled by single applications of lime ($CACO_3$) for up to 3 years and was more effective than pentachloronitrobenzene or calcium cyanamide. Lime applied at 5-33 metric tonnes per ha to severely infested Placentia sandy loam soils gave control for 2-3 years (1).

Fusarium yellows caused by the fungus, Fusarium oxysporum f. sp. apii, is the most important problem facing California celery growers. The initial symptom is a lag in growth, usually followed by yellowing of foliage. Severely infected plants frequently have an orange-brown color in the interior of the crown and vascular discoloration which often extends into the petiole. Numerous greenhouse experiments have shown that the addition of potassium and chloride to soil under a nitrate-nitrogen regime can substantially reduce the severity of Fusarium yellows. Field trials in the Salinas valley indicated the application of 400 lbs/acre calcium chloride in the irrigation water resulted in a significant reduction in disease severity but unfortunately did not satisfactorily control the disease. Resistant cultivars appear to be the solution to the control of Fusarium yellows of celery (3).

CONCLUSIONS

The appearance of wet-weather diseases in California requires the simultaneous involvement of a susceptible plant, its particular bacterial or fungus pathogen, and the proper temperature and free moisture environment. Growers should reduce disease potential by growing in the summer-dry periods and careful use of water management at all times of the year.

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FERTILIZING TOMATOES THROUGH DRIP IRRIGATION

Jewell L. Meyer
Extension Irrigation and Soils Specialist
University of California, Riverside, CA

Hunter Johnson, Jr.
Extension Vegetable Specialist
University of California, Riverside, CA

Steven E.C. Post[1]
National Rainbird
Glendora, CA

The use of trickle/drip irrigation on vegetable crops is increasing annually and much research has been conducted by both public and private institutions throughout the world on the use of this practice. There is a great deal of information available in publications from various universities, in commercial brochures or manuals, and in professional journal reports covering design, crop responses, soil moisture management, filtration and clogging prevention, the injection of various chemicals and fertilizer materials. This information and extensive field experience by growers and manufacturers alike has made trickle/drip irrigation a successful economic practice for a number of vegetable crops.

One of the crops which has made extensive use of trickle/drip irrigation is fresh market tomatoes, especially along California's south coast, and in Baja, California. The practice provides many well-known practical advantages over furrow irrigation, one of which is the ease and efficiency of supplying soluble fertilizers, especially nitrogen, to a crop during its growth, without interfering with other operations. Crops can be "spoon fed" on whatever schedule

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[1] previously Area Irrigation and Soils Specialist, University of California, Riverside, CA

the grower chooses. Mixed fertilizers, including acidified phosphorus materials, are regularly applied through drip irrigation systems, in addition to the application of dry mixed fertilizers which are applied to the soil prior to planting. A survey of these practices indicates that rates as high as 400-500 kg per hectare each of N, P, and K may be applied to a crop of tomatoes through a combination of preplant and drip injection. These rates appear to be considerably in excess of plant requirements for high yield. Some growers have questioned these rates, as well as the need and value of the extra P and K applied through the drip system. Another question which arises is the method of apportionment of nutrients to the plants during crop growth and what effects the apportionment method has on the efficiency of satisfying plant needs in terms of yield, fruit size and quality.

With these questions in mind a research project was initiated in 1983 with the cooperation of Extension Irrigation Specialists at UCR and farm advisors in San Diego County. The initial experiment was conducted on a commercial farm near Carlsbad, and subsequent experiments have been conducted annually at the U.C. South Coast Field Station near Tustin. This paper will present the results and conclusions of the past two year's experiments, 1985 and 1986.

The objectives were: 1) to evaluate nitrogen rate effects on yield; 2) to compare methods of apportioning nitrogen during crop growth; and 3) to test the effects of phosphorus applied through the drip irrigation system.

Experiments during the past two years were conducted on adjacent pieces of land, both mapped as sand Emigdio (coarse, loamy, mixed calcareous), thermic, Typic Xerofluvents. Plot preparation was the same in both years. After pre-cropping with barley to deplete nitrogen, the land was fumigated with methyl bromide chloropicrin for weed control and as a precaution against soil-borne diseases. Preplant phosphorus treatment was applied as a band of treble superphosphate 10 cm deep under the plant row before planting to supply 50 $kg.ha^{-1}$. Trickle irrigation tubing (T-Tape®, high flow, 20 cm outlet spacing) was installed 15 cm to one side of the bed center on raised beds spaced 1.5 m apart. The beds were mulched with black polyethylene film and 'Jackpot' hybrid tomatoes were transplanted in a single row 40 cm apart. Amount and frequency of irrigations was based upon Class A pan evaporation modified by a crop coefficient estimated to maintain soil moisture tension at or below 25 cb. Prior to planting, soil in the experimental areas was sampled to determine nutrient status. Nitrate and ammonium N were in the range of 6 to 10 ppm each, phosphorus was in the range of 4 to 6 ppm, and potassium about 300 ppm. Saturation percentage was 30-32%, pH 7.5, and EC_e less than 1 dS/m.

The treatments applied in both years are shown in Table 1. In treatments which received phosphorus through the drip system, the source was TVA 17-44-0 urea phosphate. Nitrogen in those treatments was supplemented with urea ammonium nitrate (UN32) to supply the total amount required, and UN32 was used as the sole N source in other treatments. Treatments were apportioned over a ten-week period in both experiments. The graduated schedule of application was designed to apply increasing amounts of nitrogen weekly and peak with the largest amounts during the period of fruit development. Apportionment in other treatments was in weekly one-tenth increments of the total to be applied. In 1985, all of the phosphorus (17-44-0) was applied during weeks two, three, and four following transplanting. In 1986, the phosphorus was apportioned in equal one-tenth increments over ten weeks beginning one week after transplanting. Leaf samples were taken every two weeks (1985) or weekly (1986) to evaluate the effects of treatments on nutrient status. Fruit was harvested weekly as mature greens or colored fruit, sized by industry standards and weighed.

Results 1985:

The effect of nitrogen rate on yield was linear from zero through 224 $kg.ha^{-1}$ (r=0.98) and curvilinear through the 448 kg rate (r=0.98). The regression curve implied an optimum response at a nitrogen rate of about 217 $kg.ha^{-}1$, but since a treatment rate between 224 and 448 $kg.ha^{-1}$ was not included, a more accurate estimate of optimum rate could not be determined. Petiole nitrate nitrogen was deficient throughout growth for zero and 56 kg rates. The plants in the 112 kg rate fell below the sufficiency level (6000 ppm) during early fruit development. The 224 and 448 kg rates maintained tissue levels in the adequate range until harvest. In spite of the pre-plant phosphorus band application in the root zone petiole P remained near the deficiency level for most of the treatments throughout growth. The treatment which received extra phosphorus in the irrigation water maintained tissue levels in the sufficiency range. There was no effect of method of apportionment of nitrogen on total yield or fruit size. There was no effect of nitrogen rate on fruit size distribution. There was, however, a significant increase in percentage of extra large fruit at the 224 kg rate with additional phosphorus through the drip system as compared with the same rate without added phosphorus. Yield results are provided in Table 2.

Results 1986:

Total yields and fruit sizes were significantly larger in 1986, perhaps due to an improvement in soil moisture management. In the nitrogen rate series (treatments 1-4), the 112 kg rate was significantly lower in yield than the 336 and 448 kg rates, but no difference in yield between the two high rates. The reduction in yield which occurred at the 448 kg rate in 1985 was not observed in 1986. The linear yield increase with nitrogen rate was due to highly significant linear increases in numbers of three fruit sizes: extra large, large and medium. There was no rate effect on maxi size fruit. Nitrogen apportionment method had no effect on yield or fruit size distribution (treatments 2 and 3 compared with treatments 6 and 7). The method of supplying phosphorus had no effect on yield or fruit size (treatments 6 and 7 compared with treatments 8 and 9).

Petiole P tended to be in the sufficient range throughout growth for all treatments, but the treatments which were supplied with phosphorus through the drip system were significantly higher in petiole levels than all others during the fruit development period. Petiole N was fairly well related with nitrogen rate and apportionment method. Both 112 kg rate treatments and the 200 lb. rate, with graduated apportionment, were deficient at all sample dates. Treatments at rates of 224 kg N or higher supplied in equal weekly amounts maintained sufficient levels through early fruit set, but only those receiving 336 Kg N and 448 Kg N (graduated supply) remained at adequate levels until harvest.

Conclusions:

The results of these experiments indicate that phosphorus supplied through the drip irrigation system or as a pre-plant band application near the plant row can be equally effective in terms of tomato yield and fruit size. They also show that phosphorus supplied through the drip system in addition to a preplant application may promote larger fruit sizes, although not total yield. This effect needs to be re-examined in a phosphorus rate experiment and may provide the added value of clarifying the relationship between phosphorus tissue levels and yield. Under the conditions of these experiments, the optimum rate of nitrogen appears to be in the range of 224 to 336 $kg.ha^{-1}$. The results have clearly shown that there is no added economic yield or fruit size value, and a potential yield reduction, to a nitrogen rate as high as 448 $kg.ha^{-1}$. Based on yield and fruit size distribution, the two methods of apportioning nitrogen to the crop are equally effective in satisfying plant requirements.

Table 1. Experimental treatments

A. 1985:

	$N(kg.ha^{-1})$	$P(kg.ha^{-1})$	Nitrogen Apportionment Method
1.	0	45[1]	-----
2.	56	45[1]	Graduated
3.	112	45[1]	Graduated
4.	228	45[1]	Graduated
5.	448	45[1]	Graduated
6.	228	45[1] + 45[2]	Graduated
7.	112	45[1]	Equal
8.	224	45[1]	Equal

B. 1986:

	$N(kg.ha^{-1})$	$P(kg.ha^{-1})$	Nitrogen Apportionment Method
1.	112	45[1]	Graduated
2.	224	45[1]	Graduated
3.	336	45[1]	Graduated
4.	448	45[1]	Graduated
5.	112	45[1]	Equal
6.	224	45[1]	Equal
7.	336	45[1]	Equal
8.	224	45[2]	Equal
9.	336	45[2]	Equal

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[1] Pre-plant band application
[2] Supplied in irrigation water

Table 2. Marketable yield and size distribution

1985:

	Treatment	t.ha^{-1}	% Size ExLg	Lg	Med	Sm
1.	No N	23.3 c	14.3	53.6	26.9	5.1
2.	56 N (G)	26.7 abc	16.0	48.5	33.1	2.4
3.	112 N (G)	27.9 abc	17.0	43.1	33.3	6.5
4.	224 N (G)	30.9 ab	11.4	43.2	36.9	8.5
5.	448 N (G)	25.4 bc	18.4	47.6	33.2	0.8
6.	224 N,P + (G)	29.1 ab	22.5	48.4	26.6	2.6
7.	112 N (E)	32.1 a	14.9	44.2	33.9	7.0
8.	224 N (E)	28.5 abc	17.6	49.3	30.5	2.6

1986:

	Treatment	t.ha^{-1}	% Size Maxi	ExLg	Lg	Med
1.	112 N (G)	34.9 b	3.7	54.3 w	32.8 z	9.2
2.	224 N (G)	42.0 ab	2.9	44.7 yz	38.4 y	13.9
3.	336 N (G)	44.9 a	2.7	49.7 wxyz	33.6 yz	14.0
4.	448 N (G)	45.7 a	2.8	50.5 wxy	34.5 yz	12.1
5.	112 N (E)	41.4 ab	3.4	52.1 wx	31.5 z	13.0
6.	224 N (E)	44.4 a	4.0	48.9 wxyz	32.4 z	14.6
7.	336 N (E)	40.4 ab	1.8	43.2 z	38.0 y	17.0
8.	224 N,P + (E)	44.8 a	2.9	51.8 wx	34.0 yz	11.2
9.	336 N,P + (E)	46.9 a	2.9	45.4 xyz	36.2 yz	15.4

NUTRIENT REQUIREMENTS OF ONION AND GARLIC CROPS

K. Tyler, D. May, J. Guerard, D. Ririe, and J. Hatakeda
University of California Cooperative Extension

Introduction

California leads the nation in the production of garlic and onions. The state also leads in the production of dehydrator garlic and dehydrator onions. Although Gilroy is still known as the garlic capital of the world, most of the garlic production has shifted to the San Joaquin Valley. Two large dehydrators remain in Gilroy, however almost all of the garlic is trucked in from Fresno and Kern Counties for processing. Dehydrator onions are produced in Fresno, Kern, Riverside, Kings, Modoc, and Monterey Counties.

Ten field experiments with dehydrator garlic and six experiments with dehydrator onions were conducted between 1979 and 1985 in the San Joaquin and Salinas Valleys. All experiments were in grower's fields except for several located at the UC West Side Field Station in western Fresno County. Objectives of the experiments were: (1) to provide information on the mineral nutrient requirements of these crops; and (2) to establish macronutrient deficiency ranges in leaf tissue for dehydrator garlic and onions. For those who desire more detailed information than will be provided here, two detailed reports, Vegetable Crops Series Number 221, "Garlic Mineral Nutrition," and Vegetable Crops Series Number 223, "Dehydrator Onion Mineral Nutrition," are being prepared for distribution early in 1987.

Garlic Results

A five-year study of garlic mineral nutrition, encompassing ten field experiments and using both California Early and Late Garlic varieties, has shown that crop responses to application of fertilizer for garlic are moderate. In fact, because of the crop's excellent ability to forage for or efficiency in utilization of phosphorus and zinc, the application of these nutrients should be required only when soils are extremely depleted of bicarbonate extractable phosphorus and DTPA extractable zinc. Addition of nitrogen at rates between 100 and 200 lbs. per acre appears adequate for the crop since most yield responses in the field experiments leveled off at either 100 or 200 lbs. of nitrogen per acre and some of the test soils were found to be very deficient in nitrate-nitrogen. Garlic produced from virus-free cloves was shown to be more responsive to higher rates of nitrogen and may benefit from rates as high as 300 lbs. of nitrogen per acre.

Plant tissue analysis of garlic leaf samples may be useful in determining the crop's nutrient status and should be used in combination with preplant soil analysis for helping to manage fertilizer programs for garlic. A leaf analysis guide below, which lists deficient, intermediate, and sufficient levels of total N, soluble phosphate-phosphorus, and soluble potassium, provides information by which garlic leaf analyses may be interpreted.

Garlic leaf analysis guide for diagnosing a crop's nutrient status.

			Nutrient Level		
Sampling time	Plant part	Plant nutrient	Def.	Inter.	Suf.
Early season (pre-bulbing)	Newest fully elongated leaf	Total N, %	4	4-5	5
		PO_4-P, ppm	2000	2000-3000	3000
		Soluble K, %	3	3-4	4
Midseason (bulbing)	Newest fully elongated leaf	Total N, %	3	3-4	4
		PO_4-P, ppm	2000	2000-3000	3000
		Soluble K, %	2	2-3	3
Late season (post bulbing)	Newest fully elongated leaf	Total N, %	2	2-3	3
		PO_4-P, ppm	2000	2000-3000	3000
		Soluble K, %	1	1-2	2

Onion Results

These studies clearly show that dehydrator onions are very responsive to fertilizer application in soils where available nutrient levels are low or deficient. In soils of low nitrogen availablity, substantial yield increases can be expected with nitrogen application of 100 to 240 pounds per acre, with little or no benefit from rates of 300 to 500 pounds per acre. On some fields yields and total solids have been reduced by rates of 400 and 500 pounds of nitrogen per acre.

Dehydrator onions are much more likely to require phosphorus fertilizer than garlic produced in the same fields. Certainly, soils with bicarbonate extractable phosphate-phosphorus levels lower than 8 ppm should have phosphorus fertilizer applied before planting dehydrator onions. Application rates should be in the range of 100 to 200 pounds P_2O_5 per acre.

The onion leaf analysis classification presented below should serve as a useful guide for sampling dehydrator onions and for interpreting analyses for nitrogen, phosphorus and potassium. Because the growth period for onions is longer than for many vegetable crops, leaf analyses can be more useful for diagnosing the nutrient status of onion crops and assuring that they are adequately fertilized with nitrogen.

Onion leaf analysis classification for diagnosing the nutrient status of onion crops.

Sampling time	Plant part	Plant nutrient	Nutrient Range Def.	 Inter.	 Suf.
Early season (100-120 days)	Tallest leaf	Total N, %	<3	3-4	>4
		PO_4-P, ppm	<1000	1000-2000	>2000
		Soluble K, %	<3	3-4	>4
Midseason (120-140 days)	Tallest leaf	Total N, %	<2½	2½-3	>3
		PO_4-P, ppm	<1000	1000-2000	>2000
		Soluble K, %	<2½	2½-4	>4
Late season (140-180 days)	Tallest leaf	Total N, %	<2	2-2½	>2½
		PO_4-P, ppm	<1000	1000-2000	>2000
		Soluble K, %	<2	2-3	>3

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