

1999 Symposium of the Oklahoma Academy of Science Plant Responses to the Environment: From Molecules to Populations

Preface

The following is a collection of abstracts of presentations by invited speakers in a Symposium held in conjunction with the 88th Annual Technical Meeting of the Oklahoma Academy of Science. The subject of the 1999 Symposium is *Plant Responses to the Environment*. The Symposium was organized to provide a forum for researchers who are studying plant-environment interactions. The subtitle of the Symposium, *From Molecules to Populations*, is intended to convey the diversity in the questions these researchers address. The goal of the Symposium organizer was to bring together researchers, teachers and students who use a wide range of experimental approaches, model systems and field and laboratory techniques to investigate fundamentally-related scientific questions. It is hoped that this meeting will stimulate increased communication and cooperation between everyone with an interest in plant-environment interactions. The most difficult task in organizing the Symposium was determining what topics should be included. Universities and research centers across Oklahoma sponsor research on many different aspects of plant-environment interactions. It is unfortunate that the constraints of time permitted presenting only a cross-section of the richness of that subject matter. The easiest and most rewarding task was issuing the invitations to the Symposium speakers: every single research group that was invited to participate accepted the invitation. The 1999 Symposium was made possible through the sponsorship of the Academy and the host institution, Oklahoma City University. The Symposium organizer is indebted to Ron Tyrl, President of the Academy, for his enthusiastic support and encouragement; and to Ed Nelson, Executive Secretary-Treasurer of the Academy, for his assistance with the arrangements. And above all, the invited speakers are to be commended for taking time from their busy schedules to share their knowledge and expertise with the Academy membership.

13 November 1999
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PLANT-SOIL INTERACTIONS IN SEMIARID ECOSYSTEMS: THE IMPORTANCE OF INDIVIDUAL SPECIES. Anne Fernald Cross; Department of Botany, Oklahoma State University, Stillwater, OK 74078,

Vegetation plays a central role in structuring semiarid ecosystems, such that plant-induced changes in structure tend to alter ecosystem-level processes, including soil nutrient cycling, with time. My research, conducted in *Bouteloua gracilis* - *Bouteloua eriopoda* grassland and *Larrea tridentata* shrubland in northern New Mexico, tested two hypotheses. First, I postulated that the spatial distribution of plant-essential elements (N, P, and K) and those taken up in the transpiration stream (Cl and Na) would mirror plant distribution. And second, I postulated that differences in the distribution of soil nutrients between grassland and shrubland would reflect different mechanisms of nutrient acquisition. I measured the spatial heterogeneity of soil nutrients and vegetation using spatially-explicit sampling combined with remote aerial photography. To examine mechanisms of nutrient acquisition, I applied the stable isotope of nitrogen, ^{15}N , to surface soils and traced its movement over 10 years. Relative contagion analyses showed distinct differences in the spatial heterogeneity of the three species. Geostatistical analyses of soil nutrient distribution demonstrated close links between plant spatial heterogeneity and soil nutrient distribution in both the vegetation types, but a lack of "islands of fertility" in the grassland. Finally, Raleigh distillation analyses of ^{15}N showed that plant uptake dominated nitrogen acquisition in the grassland, but that other processes, most likely passive capture of atmospheric nitrogen deposition, dominated nitrogen acquisition in the shrubland. Understanding how individual species affect nutrient acquisition and redistribution in semiarid ecosystems may be used to re-evaluate current models of desertification for the southwestern United States.

FRUIT MATURATION AND PATHOGENESIS. Charlie Biles (1), Benny D. Bruton (2), and J.X. Zhang (2); (1) Biology Department, East Central University, Ada, OK 74820, and; (2)USDA, Agricultural Research Service, Lane, OK 74555.

Approximately 10% of the harvested and marketed fruit and vegetables in the U.S. soften and rot on the shelf because of natural fruit softening processes or postharvest decay organisms. The percentage of the crop lost after harvest is greatest in developing nations. The goal of our research program is to investigate melon fruit physiology in regard to fruit development and decay caused primarily by fungal pathogens. Cantaloupes are usually harvested *ca.* 40 days after pollination. The fruit has a relatively short shelf-life of 12-14 days that may be further shortened by fungal infection. There are two general categories of fungal pathogens; latent and non-latent. Latent pathogens are thought to invade the fruit epidermis (exocarp) during net development; 12-25 days after pollination. The fungus remains dormant until the fruit is harvested. Subsequently, the fungus begins to cause decay which may not be discovered until the fruit reaches market. Non-latent pathogens infect the fruit at all stages of development. Both latent and non-latent pathogens cause severe losses. Our laboratory has investigated several fungal cell wall-degrading enzymes that play a role in fruit decay. Polygalacturonase (PG) has been found to be synthesized in fruit tissue by the latent pathogen *Phomopsis cucurbitae*. PG produced by the fungus appears to be inhibited until the fruit is about 40 days old. The data suggest that there may be fruit PG-inhibitors that delay the onset of the latent pathogen. Two plant defense related enzymes that have been investigated in our laboratory are peroxidase and chitinase. Peroxidase activity peaks 30 days after pollination. HPLC anion exchange chromatography indicates that an anionic peroxidase declines in 40 and 50 day fruit when compared to 30 day fruit. The time of anionic peroxidase decline in activity corresponds to the activation of the latent pathogen *Phomopsis cucurbitae*. Chitinase is an exo-chitinase that has been found throughout the cantaloupe fruit. Early in fruit development, two chitinase isozymes were evident according to HPLC anion-exchange chromatography profiles. However, the isozyme disappears in 20 day fruit and did not reappear in later developmental stages. If these enzymes are involved, it may be possible to use genetic bioengineering to maintain or increase the level of these enzymes to inhibit fungal colonization of the fruit.

GENOMIC ANALYSIS OF PLANT RESPONSES TO SALINITY AND DROUGHT STRESS. John Cushman (1), Robert Burnap (2), Eduardo Misawa (3), Rolf Prade (2); Ray Bressan (4), Paul Hasegawa (4), Jose Pardo (4); Jian-Kang Zhu (7), David Galbraith (7), Hans Bohnert (5,6,7); (1) Departments of Biochemistry/Molecular Biology, (2) Microbiology/Molecular Genetics and (3) Mechanical and Aerospace Engineering, Oklahoma State University, Stillwater, OK 74078; (4) Department of Horticulture, 1165 Horticulture Building, Purdue University, W. Lafayette, IN 47907; (5) Departments of Biochemistry, (6) Molecular /Cellular Biology, and (7) Plant Sciences, University of Arizona, 1041 East Lowell Street, Tucson, AZ 85721; USA.

Salinity and drought stresses are responsible for greater reductions in crop productivity than any other biotic or abiotic factor. To improve plant stress tolerance in the future, a greater understanding of complex stress signaling and adaptation processes will be required. Technical advances in high-throughput DNA sequencing, gene chips, and functional genomics now permit a global understanding of the complex ways in which plants perceive and respond to salinity and drought stresses. The University of Arizona, Oklahoma State University, and Purdue University have formed a consortium to isolate and functionally analyze all stress-related genes in halophytic and glycophytic plant (*Arabidopsis*, Ice plant, and Rice), and non-plant models (*Aspergillus*, *Dunaliella*, *Synechocystis*, and *Saccharomyces*). Comparisons among these diverse organisms should reveal both evolutionarily conserved and unique stress defense mechanisms. Large scale sequencing of stress-specific Expressed Sequence Tags (ESTs) and microarray analysis are being used to link structural genomic information with functional analyses. Functional analyses of genes important for stress signaling and tolerance include random and targeted mutagenesis screening, complementation assays, reverse genetic screens, and detailed analysis of reporter gene fusions.

MOLECULAR ANALYSES OF AN ARBUSCULAR MYCORRHIZAL SYMBIOSIS IN *MEDICAGO TRUNCATULA*. Ignacio E. Maldonado-Mendoza, and Maria J. Harrison. Plant Biology Division. The Samuel Roberts Noble Foundation. P.O. Box 2180.2510 Sam Noble Parkway. Ardmore, OK 73402. USA.

The arbuscular mycorrhiza (AM) is the most common association found in plants and the predominant type of mycorrhiza found in crops of commercial importance. The association is considered mutually beneficial: the plant supplies the fungus with carbon, while the fungus assists the plant with the uptake of phosphate and other nutrients from the soil. The development of the functional symbiosis requires a complex series of interactions between the two symbionts. When the fungal hyphae contact the root surface they differentiate to form an appressorium, and subsequently penetrate the root. Once inside the root, the hyphae grows inter- and intracellularly throughout the cortex and differentiate within the cortical cells to form highly branched structures known as arbuscules. The arbuscule/cortical cell interface is predicted to be the site at which nutrient exchange occurs. We have selected *Medicago truncatula*, a model legume, and the AM fungi, *Glomus versiforme* and *G. intraradices* to study genetic and molecular aspects of the association. Several *M. truncatula* genes that show differential expression in the symbiosis have been identified: a xyloglucan endotransglycosylase (XET)-related protein, a putative arabinogalactan protein (AGP), and a putative homologue of a mammalian translation initiation factor (eIF3). cDNA clones encoding both plant and fungal phosphate transporters have also been identified and characterization of these clones is providing insight into the mechanisms of phosphate transport in the symbiosis. The obligate biotrophic nature of AM fungi imposes limitations to study gene expression in fungal parts. We have characterized a cDNA encoding a fungal phosphate transporter expressed in external hyphae using a monoxenic root organ culture system. Our results suggest that the fungal external hyphae have the ability to sense the levels of phosphate surrounding them and that phosphate availability can regulate phosphate transporter gene expression in AM fungi. The use of this system in which a fungal gene is analyzed in a "separate" context from its plant host will allow us to gain an understanding of the mechanisms that regulate the cross talk between the two symbionts during this association.

APOMIXIS: ITS TRANSFER AND EXPRESSION IN MAIZE-*TRIPSACUM* BACKCROSS HYBRIDS. Bryan Kindiger; USDA-ARS, Grazinglands Research Laboratory, El Reno, OK 73036.

Specific reproductive characteristics associated with the diplosporous form of apomixis in *Tripsacum dactyloides* were compared to those same characteristics in a set of apomictic maize-*Tripsacum* backcross hybrids carrying 36, 18, and 9 *Tripsacum* chromosomes. Chromosome karyotypes and random amplified polymorphic DNA (RAPD) analyses were used to assess the chromosome constitution, genetic uniformity, and mode of reproduction of the apomictic maize-*Tripsacum* backcross hybrids representing four differing lines of descent. Data relevant to the number of progeny generated by apomixis sexual developmental behaviors, sexual polyploidization, and polyembryony were obtained. The data indicate that the number of apomictic sexual, BIII derived hybrids and twin offspring occur at similar frequencies regardless of their maize and or *Tripsacum* chromosome constitution. However, some differences were observed when data obtained from apomictic *Tripsacum* was compared to the apomictic maize-*Tripsacum* backcross hybrids. Preliminary comparisons on the type and frequency of progeny obtained from identical or closely related families during various growing seasons suggest that normal environmental components do not obviously affect apomictic expression. In addition, chromosome and molecular variations were observed in apomictic progeny obtained from apomictic parents, indicating that an infrequent level of partial meiotic activity is occurring in apomictic individuals.

GLOBAL ENVIRONMENTAL CHANGE AND TERRESTRIAL ECOSYSTEMS. Yiqi Luo; Department of Botany and Microbiology, University of Oklahoma, Norman, OK 73019.

Human activities have resulted in dramatic changes in the environment of planet Earth. For example, carbon dioxide (CO₂) concentration in the atmosphere has increased at an unprecedented rate, rising from 276 parts per million (PPM) in the pre-industrial time to 365 PPM now. It is predicted that atmospheric CO₂ will reach 700 PPM by the end of the 21st century. Climate anomalies, such as El Niño, hurricanes, and sizzling summer heat, occur much more frequently and at more devastating magnitudes now than before.

In facing the global environmental change, biologists have to answer two questions. First, how will global environmental change alter the function and structure of the biosphere, the system capable of supporting living organisms? Second, how will biosphere functioning regulate global environmental change?

In this talk, I will focus on the research that my group has conducted on carbon (C) exchange between the atmosphere and terrestrial ecosystems. Each year, fossil fuel burning and deforestation release approximately seven billion tons of C into the atmosphere, half of which remains in the atmosphere. Terrestrial ecosystems and oceans presumably take up the other half. Our research is designed to identify the mechanisms underlying C uptake in the terrestrial ecosystems, as well as to quantify the amount of C sequestered by global terrestrial ecosystems. My talk will present three examples of our research contributions. The first is on identification of the physiological mechanism in controlling photosynthetic acclimation to elevated CO₂. The second is on discovery of an invariant function that is universal for all C₃ plants. The function enables us to cut across interspecific variation and environmental heterogeneity to estimate the CO₂-stimulated increment in the global terrestrial C influx. The third is on development of deconvolution and inverse analysis in ecology. The novel approach will help extrapolate results from manipulative experiments in ecology to prediction of ecosystem responses to global change in the natural world.

TOWARD UNDERSTANDING THE POSSIBLE ROLE OF PHOSPHOINOSITIDE-SPECIFIC PHOSPHOLIPASE C IN PLANTS. Toshiro Shigaki, Christian Dammann and Madan K. Bhattacharyya; Plant Biology Division, The Samuel Roberts Noble Foundation, P.O. Box 2180, Ardmore, OK 73402.

Phosphoinositide-specific phospholipase C (PI-PLC) regulates many important cellular functions in animal cells through the production of inositol (1,4,5)-trisphosphate (IP₃) and the subsequent release of calcium. PI-PLC genes, with the highest similarity to mammalian PI-PLC-d isoforms, are cloned from several plant species. This indicates the possible involvement of this signal pathway in plants. We have observed from both radio-receptor assay and HPLC analysis that IP₃ contents increased rapidly to several folds over water control in soybean cells following replenishment of growing medium with Murashige-Skoog inorganic salts. This increased IP₃ content correlates with the increased DNA synthesis. On the contrary, infection with a bacterial pathogen, *Pseudomonas syringae* pv. *glycinea*, resulted in a rapid reduction of cellular IP₃ contents in soybean cells. It appears that PI-PLC, which may be involved in cell growth and other housekeeping functions, is inhibited to a sustainable period following infection. It is possible that signals of plant or/and pathogen origin(s) may be involved in altering PI-PLC-regulated host metabolisms in infected cells to meet the new needs of a host-pathogen interaction. Recently, we have cloned a gene that encodes a soybean PI-PLC1-associated protein. No significant homology to genes with known function could be found in the GenBank. However, a search in the "prints" database revealed six footprints of the rhodopsin super-family of G-protein-coupled receptors. Interestingly, α1-adrenergic receptors, shown to regulate mammalian PI-PLC-d isoforms, also belong to this super-family. Preliminary results indicate that PI-PLC activity itself can effect the transcription of this putative receptor protein. This suggests the presence of a regulatory feedback mechanism that ensures the stable level of PI-PLC activity in cells. We are currently investigating the possible function of this candidate regulatory protein.

PHYTOREMEDIATION OF POLYAROMATIC HYDROCARBONS: PLANTS AND MICROORGANISMS. Paul E. Olson, John S. Fletcher, Michael D. Kyle, and David P. Nagle; Department of Botany and Microbiology, University of Oklahoma, Norman, OK 73019.

Examination of volunteer vegetation growing on industrial sludge contaminated with polyaromatic hydrocarbons (PAHs) has been used as a means of identifying particular plant species whose roots foster the restoration of contaminated soil and the degradation of organic contaminants. Examination of aerial photographs taken over the last 25 years has been used to characterize the time and nature of plant invasion and plant succession that has resulted in the development of a diverse and healthy community of plants currently growing at the site. The study is designed to determine the influence of different plant species ranging in age from 5 to 15 years on the organic contaminants present in industrial sludge at depths ranging from 30 to 180 cm. The distribution of roots and their influence on the rhizosphere microbial ecology in the soil/sludge at this site is also being examined. The results of this field work will be discussed in relationship to earlier laboratory studies (Donnelly et al. 1994, *Chemosphere* 28:981, and Fletcher and Hegde 1995, *Chemosphere* 31:3009) that have shown the importance of root phenolics in fostering the growth and action of desired bacteria that degrade recalcitrant organic pollutants. The goal of this research is to identify particular plant species and plant husbandry practices for use in establishing plant communities on industrial sludge and/or contaminated soil that will foster the sustained, gradual degradation of organic contaminants. An established plant community with root-associated microorganisms provides an ecologically stable, low cost, intrinsic bioremediation system.

PLANT RESPONSE TO HERBIVORY: FROM INDIVIDUAL TO ECOSYSTEM. Linda L. Wallace; Department of Botany and Microbiology, University of Oklahoma, Norman, OK 73019

Herbivory was once thought to be a simple process of tissue removal from plants. However, we now know that herbivory processes occur across several temporal and spatial scales. Similarly, plant response to herbivory tracks across these multiple scales. A great deal of controversy concerning herbivory is predicated on misinterpretation of which scale is being considered. I will report on some of my work looking at individual plant response to herbivory and how that individual response can be scaled up to community and ecosystem levels. For example, plant transpiration rates are positively and linearly correlated with nitrogen uptake rates. Thus, herbivory effects on leaf area indices will influence soil nitrogen and competitive interactions among plants within a community. Comparisons of studies in Yellowstone National Park and the tallgrass prairies of Oklahoma have shown that in systems where plants compensate for herbivory (i.e. increase photosynthesis, transpiration and growth), community structuring rules are vastly different from systems in which species do not compensate.