SOLVING TODAY'S PROBLEMS WITH TOMORROW'S TECHNOLOGY

Understanding Water Stress

The competition for Arizona's limited water resources is clearly increasing as the state continues to grow. It is increasingly important to develop means for reducing water usage by crop and household plants. Higher efficiencies in water use can be achieved by modifying cultural practices such as irrigation scheduling, or by developing new lines of plants through selection or by newer methods such as genetic engineering. In any case, it's necessary to know when a plant becomes stressed enough so its performance falls below desired levels. Our major goal is to understand how deficits in water regulate growth and metabolism of plants so that logical approaches can be used to modify plants.

Stress-caused growth responses of plants have been of major concern to physiologists because water stress almost always reduces plant size, and therefore, productivity. As a result of many studies, stress effects on growth were considered primarily in terms of the turgidity of the cell, a biophysical property. This made it difficult to imagine how genes could be modified to confer stress resistance. The data used to relate growth to tissue water status, however, were obtained almost wholly from tissues which had already expanded. Through use of combined biophysical, biochemical and anatomical approaches, we have found that the stress responses of plant parts actually involved in growth are much different than those of expanded tissues, and the results have led to major modification in thinking about how stress regulates growth. As a result of these studies, growth is now seen as being a combined biochemical and biophysical process, and it is now possible to suggest how growth processes can be regulated directly by gene action. We are currently trying to identify the specific growth related processes that are affected by water stress.

Dr. Kaoru Matsuda
Plant Sciences

A. Fertig

Geminivirus Component Synthesis

Geminiviruses are an important group of plant viruses which have become prominent in the Southwest in recent years. They are responsible for diseases of cotton, squash, melons and other important plants in Arizona. These viruses are transmitted by the sweet potato whitefly or, in one case, the beet leafhopper. It is not understood why geminiviruses cause disease in some plants but not in others, how insect vector specificity is determined, or how interactions with different plants result in different disease severities. These are subjects of ongoing research in this laboratory.

Fortunately the geminiviruses are genetically simple with only five or six viral genes code for the production of proteins in infected plants. One of these proteins is the coat protein which assembles to form the protective shell for viral DNA. This shell is uniquely shaped to resemble twin spheres, thus providing inspiration for the name geminivirus. Other proteins are produced in much lower quantities and, though they are predicted from our studies of the viral genes, have escaped detection by traditional methods. Progress in understanding the functions of these proteins is hindered by their low concentrations.

Molecular methods involving recombinant DNA experiments are being used to solve this problem. Genes of bean golden mosaic virus (BGMV) are being cloned into bacterial plasmids in ways that direct the bacteria to make viral proteins. Proteins synthesized in this way can be produced in large quantities, purified and used for making specific antisera. These antisera are used as tools in sensitive detection schemes to bind to the authentic viral proteins in extracts of infected plants. Characterization of these viral proteins will advance our understanding of their roles in viral pathogenesis.

Another aspect of this work is to determine the functions of geminiviral proteins. We are deliberately mutating viral genes to study the changes these mutations have on the function of their corresponding proteins. Many of these mutations are lethal to the virus and are, thus, of limited usefulness in telling us about function. We can avoid the lethality problem by inserting mutated viral genes into the chromosomes of plants to allow the plant to direct the replication and expression of the genes. This approach uncouples the production of a functional protein from the virus replication. These experiments are underway in our laboratory using plasmids of the soil bacterium, *Agrobacterium tumefaciens*, as vectors for moving genes into plant chromosomes.

A third facet of our work involves detection of geminivirus infections in plants collected in the field. Using computer analysis of many different geminivirus DNA sequences we have identified DNA sequences which all of the viruses have in common. We are testing whether a cloned copy of this sequence can be used to detect virtually any geminivirus. A simple method of purifying geminiviruses from field samples is being tested by itself or in connection with the cloned DNA probe for rapid and accurate diagnosis.

Dr. Alan J. Howarth
Dr. Joseph G. Utermohlen
Plant Pathology

Macrofibers, A Multi-Use Bacterial Material

Experimental methodologies involving bacteria, including their use in biotechnology have focused almost exclusively on bacteria as individual cells. Many bacteria found in natural settings, however, grow in aggregate forms in which the biomass behaves more like a multicellular organism rather than as individual cells. Surprisingly little is known about the growth and physiology of such forms or their potential applications.

About 10 years ago I discovered that *Bacillus subtilis*, a spore-forming bacterium commonly found in soil can be made to grow in the form of a highly ordered multicellular fiber-like structure. We have established a number of new ways to study macrofibers that provide complement information to that gained using conventional bacterial genetics and physiological protocols. Of primary importance has been the use of biomechanics.

The structure of a bacterial macrofiber is not very different from that of a helically twisted multifilament textile fiber, consequently I have established a collaborative effort with Professor J.J. Thwaites of Cambridge University, a mechanical engineer-fiber scientist. We have been able to measure the material properties of bacterial cells such as the visco-elastic nature of the cell wall, and to precisely quantitate the twist states of macrofibers and their formation. These fundamental properties, many never before measured in bacterial cells, have been useful in providing clues to the way in which bacterial cells grow and assemble their cell walls.

From studies of the biomechanics of macrofiber formation it became apparent that the motions associated with growth of the multicellular form could be use to make the structure adhere to substrates. A new method of cell immobilization was developed based upon the principle of entanglement. Immobilized macrofibers have important potential in biotechnology where it is often desired to keep the products made by the bacteria separate from the cells themselves. We have had some initial success as well in producing associations of macrofibers with plant roots. In view of the ability to use genetic engineering techniques with *B. subtilis* to produce strains that make various desired products, it seems likely that macrofibers may find applications in many different systems. However, the most obvious application, the use of bacterial macrofibers as fibers, has yet to be explored.

Dr. Neil H. Mendelson
Nutrition and Food Science

Remote Sensing and U.S.-Mexican Land Use

Concern has recently increased over the apparent trend toward desertification in the world's arid lands. Desertification may be resulting from overgrazing on fragile arid and semiarid rangelands and the subsequent irreversible loss of vegetative cover. To help understand the situation, remote sensing, a technology that uses data gathered from aircraft or earth-orbiting satellites, is being used to explore the interaction between vegetation and climate. Remote sensing allows the monitoring of changes in land surface reflectance and soil moisture, both symptoms of severe land degradation.

At the UA's Remote Sensing Center (ARSC), researchers have been studying the effects of vegetation removal on surface reflectance and its effect on the proportion of solar energy that is gained or lost along a portion of the Arizona-Sonoran border. This boundary area is an ideal place to study the results of different land use practices on surface characteristics, as a 70-year-old fence cuts across areas that were once virtually identical. ARSC's research focus has been on rangeland areas with a long history of high stocking rates on the Mexican side, adjacent to rangeland that has been less intensively used on the U.S. side. In some locations, this contrast is striking, even on satellite imagery.

The recently completed first phase involved the documentation of vegetation differences between selected border sites, and the ability to measure these differences using remotely-sensed data. This phase focused on three of the major plant communities found between Lochiel, Arizona, and the border city of Nogales: an oak woodland, a mesquite grassland, and a mixed tall and short grass plains grassland. Within each vegetation community, paired study sites were located on both sides of the border that shared similar soils, vegetation, topography, hydrology; the only difference was the intensity of land use. Throughout 1987, these sites were intensively sampled to measure plant biomass, green leaf area, vegetation cover and soil type. Coincident with the field work was a series of overflights with a light aircraft that provided surface reflectance and surface temperature data that was then related to ground observations.

Results of the ground studies showed significant biomass and percent cover differences between Mexican and U.S. sites; the ground-spectral analyses indicated that these differences were detectable using the reflectance data. The project's next phase will focus on the relationship between surface vegetation cover and local surface solar-energy budgets. Intensive field measurements of radiation, evaporation (latent heat), sensible heat, and soil heat will be made at the Lochiel site to establish the effect of surface reflectance differences on local surface solar-energy budgets.

Louisa Beck
Dr. Charles Hutchinson
Arizona Remote Sensing Center
Arid Lands Studies
Dr. Alfredo Huete
Soil and Water Science

Smart Machines

Agricultural engineers and other agricultural scientists all over the country are developing expert systems to help the farmer of the future. At the University of Arizona we are combining expert systems with computer vision so that some day agricultural machines can "see" and "think".

Agricultural engineers in Florida have developed a machine that can "see" ripe oranges and pick them off the tree. Other agricultural engineers in New York have developed a machine that can "inspect" apples for bruises at the rate of 30 per minute. It takes the grocery shopper longer to select good fruit in the grocery store. In addition, the computer apple "inspector" can even measure the area of a bruise on the apple.

UA researchers are classifying the characteristics of vision devices so that a computer program "expert system", can be created that will select the appropriate components of vision systems for each unique application in agriculture—planting, cultivating, harvesting, sorting and packing.

Combining vision and reasoning the way humans do is a task that industrial engineers throughout the world are working to perfect. Intelligent robots, smart machines, are being perfected to make cars, to inspect food packaging, and to inspect the soldering in new television sets.

To apply these principles to agricultural machines in the field with poor lighting and rough terrain is a formidable task. Don't expect your dealer to have these smart machines of the future on the showroom floor next time you visit him. If these smart machines don't turn out to be cost effective you won't see them in the showrooms at all. The farmer won't be replaced by smart machines. He will just be able to do his job better.

Dr. Kenneth A. Jordan
Dr. Wayne Coates
Agricultural Engineering

Biotech Nematode Control

Root knot nematodes are among the most destructive pests of cultivated crops. More than 3,000 plant species are attacked and annual losses exceed $3 billion nationwide. Cotton, Arizona's premier crop, is highly susceptible.

For years, control of these microscopic parasites has depended upon chemical treatment of infested soil. However, increasing public concern and governmental regulation have eliminated many of the most effective nematicides. Consequently, researchers are looking for alternative methods of control.

Nematodes are attracted to the roots of host plants which they penetrate to initiate feeding sites. The mechanisms of attraction, host recognition, penetration, and subsequent modification of host tissue are affected or modulated by secretions from the infecting nematodes. Since disruption of these processes would lead to virtual control, there is considerable interest in defining the nature and mode of action of nematode secretions.

In the College of Agriculture, a program is underway to isolate and characterize two primary secretions, amphidial and salivary exudates. Amphidial exudates are thought to play an important role in host-finding and recognition since they originate from the nematodes' main sensory receptors, the amphids. Salivary exudates originate in the salivary glands and are extruded from the stylet, a hollow, needle-like feeding apparatus that the nematode uses to puncture plant cells. Salivary exudates bring about the dramatic changes in host tissues that are characteristic of root-knot infections and that probably are responsible for most of the damage that the nematode inflicts on its host, although other secretions may be involved as well.

A major breakthrough in this study has been the discovery that salivary secretions can be induced by treating nematodes with synthetic and naturally occurring substances, many of which resemble neurotransmitters in mammalian systems. Using materials such as catechol or resorcinol to stimulate salivation, it will now be possible to collect and purify quantities of material adequate for analysis and characterization.

Monoclonal antibodies will aid this effort since secretions from a single nematode are so small that even the best microscopic techniques may not be able to detect them. Monoclonal antibodies, produced in our laboratories, are being used to detect and isolate amphidial secretions and to determine their fate in host tissues.

Dr. Michael A. McClure
Plant Pathology

Desert Plant Antibiotics

During the past 40 years, an intensive effort has been made to discover novel and safe antibiotics useful against clinical diseases caused by bacteria, fungi and mycobacteria. This work has led to the discovery of approximately 7,000 naturally occurring and more than 50,000 semi-synthetically derived antibiotics. Of this number, only about 150 are currently in clinical use.

In spite of all the success in treating many infections with antibiotics of microbial origin, new types of antibiotics with different activity spectra and new mechanisms of action are required. These drugs are needed to combat multiple resistant strains, allergic reactions, systemic mycoses or serious side effects. These various factors provide us with powerful incentives to search for active agents from previously underexplored sources.

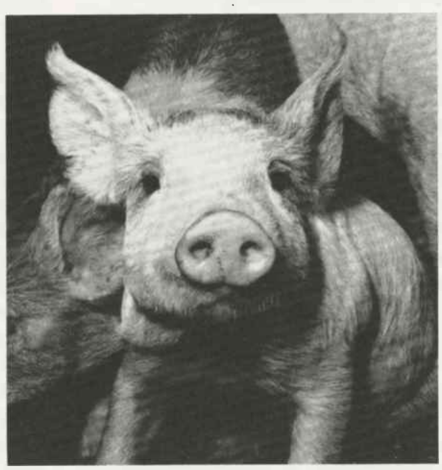
We suspect that the antibiotics known today represent only a portion of those to be found in nature. The plant kingdom still seems to be a potentially rich source of antimicrobial agents as demonstrated by the long history of plant use against bacteria and fungal infections documented in folk medicines throughout the world. Natural plant products are an attractive source of potential leads to new antibiotics, not only for the diversity and novelty of chemical structures produced by these living organisms, but also for the potential specificity of biological action. Alternatively, the natural product might provide a template or assemblage of key functionalities, stereochemistry and conformation which are responsible for the observed activity and can be synthesized in simple analogs with retention of activity.

With these concepts in mind and with the notion that arid-adapted plant species might be an untapped reservoir of biologically potent compounds, we embarked on an effort to evaluate the potential of natural products as anti-infective agents.

Our preliminary screen is designed to identify potential drugs that would be useful in combatting fungal infections (*Candida brassicae, C. albicans*), tuberculosis (*Mycobacteria*), and gram positive and negative bacteria (*Staphylococcus aureus, Bacillus subtilis, E. coli, Klebsiella pneumoniae*). During the course of this screen, we collected, extracted and tested 174 different plant species representing 108 different genera. From this preliminary screening we identified 22 active plants that belong to the tribes Astereae, Heliantheae, Helenieae, Anthemideae and Inuleae of the Asteraceae.

Dr. Barbara N. Timmermann
Arid Lands Studies

A. Fertig



A. Fertig