Plant Biological and Molecular Processes
FY 2002 National Program Annual Report

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Introduction

This program includes much of the Agricultural Research Service's fundamental research that is necessary for practical advances, but is too far upstream to provide direct solutions for practical problems. The research focuses on mechanistic understanding of specific plant processes and properties. The knowledge and tools developed can be used to improve functions and properties of plants. This program is divided into three components: Analysis and Modification of Plant Genomes (functional genomics, focusing on the molecular end of the spectrum; technology for modifying plant genomes); Biological Processes that Determine Plant Productivity and Quality (mechanisms of plant growth and development, photosynthesis, productivity, and environmental responses that relate processes and attributes of the whole organism to their genetic and metabolic underpinnings and provide the context for molecular manipulations); and Mechanisms of Plant Interactions with Other Organisms (plant defensive reactions to pests and pathogens, emphasizing those that reduce the need for applied pesticides; interactions with beneficial organisms; secondary metabolism and products). Together, the results of the research approaches under this National Program provide a continuum of understanding of plant function from genes to phenotype (plant attributes and performance).

During 2002, there were many important discoveries and advances, some of which are described below. By no means do these selected accomplishments capture the important achievements of the entire research program. Instead, they highlight the type of activities carried out under this program and the type of benefits that result. The advances are grouped by program components.

Selected Accomplishments by Component

Component I. Analysis and Modification of Plant Genomes

Evaluation of herbicide-resistant rice systems. Hybridization between rice and red rice, a dominant weed in the southern United States, may become problematic when herbicide-resistant rice systems come into use. DNA/PCR microsatellite fingerprinting analyses were conducted to quantify rates of outcrossing between three imidasolinone-resistant rice cultivars and red rice at the Dale Bumpers National Rice Research Center, Stuttgart, Arkansas, in cooperation with the University of Arkansas. It was determined that outcrossing between these imidazolinone-resistant rice cultivars and red rice occurred at very low levels and decreased with decreasing synchronization of flowering under field conditions. Thus, a key management consideration in herbicide-resistant rice systems may be to plant rice cultivars that are least likely to flower during the same period as the infesting population of red rice.

Developing genetic tools to protect soybean from attack by nematodes. The soybean cyst nematode is the most devastating pest of soybean and is responsible for about $1 billion losses in U.S. farm income annually. ARS scientists in Beltsville, Maryland, constructed microarrays containing 3,000 cDNA clones from gene libraries derived from soybeans infected with soybean cyst nematode, race 3. Use of these microarrays revealed that resistant (but not susceptible) cultivars induce specific genes soon after nematode attack. This suggests that these particular genes are involved in effecting a defense response. These genes have been pinpointed to an area of the soybean genome, which has been cloned and partially sequenced. Genetic markers have been developed from this genomic data to aid soybean breeders in selecting soybean plants resistant to the soybean cyst nematode.

Development of sophisticated microarrays with genes that regulate soybean seed composition. Many genes govern traits for improved soybean oil and protein quality. It is very difficult to determine the impact of beneficial natural or genetically engineered mutations in these genes on the expression of other traits in soybean. ARS scientists in West Lafayette, Indiana, used advanced molecular genetic techniques (suppression subtractive hybridization) to generate libraries of differentially-expressed gene messages at several periods during seed development. Three thousand high-quality clones of expressed genes in those libraries were printed to glass-slide microarrays. This technology provides an important tool for assessing the overall impact of genetic modification of one trait on all other traits in soybean seed, and will help identify potential effects of the modified gene that may be detrimental to food quality.

Component II. Biological Processes that Determine Plant Productivity and Quality

Using biological mechanisms to protect soybeans from attack by nematodes. Soybean cyst nematode is the major pest that limits U.S. soybean production causing about $1 billion losses in U.S. farm income annually. ARS scientists in Beltsville, Maryland, discovered two cell-wall hydrolase genes that are expressed during infection of soybean cyst nematode susceptible soybeans. The promoter sequence for these genes, which controls when and where they are expressed, is being isolated to attach to genes conferring resistance to the nematode. When genetically engineered into the host plant, this construct should stop nematode-caused damage early, but without changing any properties of the plant in the absence of the pest.

Improving the nutritional value of soybean protein. Soybean seed meal is deficient in sulfur-containing amino acids that are nutritionally essential for monogastric animals and humans. ARS scientists in Columbia, Missouri, characterized a gene that encodes APS reductase, a key enzyme involved in sulfur metabolism. Genetic manipulation of the expression of this gene or the activity of its enzyme product could lead to higher levels of cysteine, a dietary essential amino acid, in soybean protein.

Biological mechanisms to protect plants from heat-stress. Plants have an inducible, multi-component heat protection system that can reactivate partially denatured enzymes or remove fully denatured enzymes from the cell, thereby preventing cell injury or death. ARS scientists in Lubbock, Texas, identified four genes with distinct mutations that regulate plant response to high temperatures. These acquired thermo-tolerance deficient mutants are currently being used to identify the genes responsible for heat-stress protection in plants.

Selection of germplasm to help produce peanuts with less water. Water is the most limiting factor in U.S. peanut production. ARS scientists in Dawson, Georgia, showed that peanut varieties vary in water use efficiency. Varieties were selected for their ability to produce high yields under differing levels of water availability. These lines will be useful in future breeding efforts to develop productive 'water-saver' peanut varieties.

Reducing 'trans' fat in soy-based food products. Soybean oil is often hydrogenated in many food applications to improve flavor and functional properties of the oil. Unfortunately, some of the fatty acids are converted to 'trans-fat' during hydrogenation, which is a health concern. ARS scientists in Raleigh, North Carolina, developed a non-transgenic soybean with oil that requires less processing (hydrogenation) for food use. This germplasm is being used by soybean breeders throughout the United States to develop productive soybean varieties with higher levels of oleic acid. This trait provides vegetable oil processors added flexibility for achieving better flavor, improved frying stability, and healthier food products.

Discovery of a signaling protein between plant cells. ARS scientists in Albany, California, in collaboration with researchers at the University of California-Riverside, cloned a gene (CLV3) for a signaling protein between plant cells. CLV3 protein was shown to be located in the extracellular space of plant cells. This is the first demonstration in plants that an extracellular signaling protein is required for cell-to-cell communication during plant development. Plant scientists can use this new gene to understand plant growth and eventually to enhance shoot and flower structure in crops such as corn and rice.

Protein identified that helps wheat plants survive freezing temperatures. ARS scientists in Beltsville, Maryland, identified a protein that helps protect winter wheat cells from freeze-damage by interacting with water channel proteins in the cell. Discovery of this protein that can protect plant cells from freezing damage may accelerate the development of more cold tolerant crops and horticultural species.

A biochemical basis for heat tolerance in cotton. The Cotton Belt of the United States stretches across the southern tier of States from the Atlantic to the Pacific Oceans. High temperatures in the Cotton Belt often impair cotton growth. ARS scientists in Urbana, Illinois, discovered the "weakest link" in the photosynthetic chain of events leading to productivity. It is the enzyme rubisco activase, which allows the plant to maintain a high capacity for taking up carbon dioxide from the air at normal temperatures. The scientists are working with a private-sector company to modify this enzyme in a way that stimulates photosynthesis at high temperatures.

Engineering increased resveratrol content in blueberries. Reveratrol, a natural plant compound with chemotherapeutic properties is also an antifungal agent. ARS scientists in Oxford, Mississippi, in collaboration with the University of Idaho, genetically inserted a modified gene in blueberries that encodes the enzyme reveratol synthase, which elevates the level of resveratrol. Transformation of this gene into different cultivars should result in the development of pathogen-resistant blueberry varieties.

Determining how cancer-fighting drugs work. Mayapple (Podophyllum peltatum) produces the compound podophyllotoxin, which serves as a biochemical structural backbone for anti-cancer drugs. ARS scientists in Oxford, Mississippi, found that this compound inhibits plant growth by affecting the formation of mitotic microtubular organizing centers. This was discovered using immunofluorescence observation of the formation of tubulin in cells treated with podophyllotoxin.

Determining how Fusarium toxins work. ARS scientists in Oxford, Mississippi, described the complex mechanism of action of the lethal phytotoxin 2,5-anhydro-D-glucitol, which is produced by the pathogenic fungus Fusarium solani. Very small amounts of this toxin causes plant cell death. This toxin is a structural analogue of the simple sugar, fructose. When phosphorylated by plant kinase enzymes to form 2,5-anhydro-D-glucitol-bisphosphate, this bioactivated compound inhibits the key metabolic enzyme fructose-1,6-bisphosphate aldolase, leading to plant death.

Allergen removed from soybean. Food allergies are an increasing concern for oilseed crops. ARS scientists in Beltsville, Maryland, and St. Louis, Missouri, in collaboration with the Arkansas Children's Hospital, DuPont and Pioneer Hi-Bred International, genetically engineered soybean to remove a major human allergen. Their biotechnological approach is called gene silencing. The new allergen-minus soybean germplasm can be used to develop soy-based foods that have a lower allergenic risk to consumers.

Genetically engineered tomatoes high in antioxidants. The use of genetic engineering to improve quality traits in foods is still in its infancy. ARS scientists in Beltsville, Maryland, engineered tomatoes to express a common gene from yeast. The yeast gene increased the vine life, juice quality, and antioxidant level of the field-grown tomatoes. Earlier studies showed that the gene also increases the level of lycopene, an anti-cancer agent, in field-grown tomatoes. These may be the first reports of genetic engineering to improve healthfulness of foods by enhancing phytonutrient content.

A better tasting tomato in our future? Current production and shipping practices require that tomatoes be harvested before ripening, then ripened later using ethylene gas to initiate the process. While this allows long-term storage and the ripened tomatoes achieve full color, they often do not achieve full flavor. ARS scientists in Ithaca, New York, isolated and characterized a gene that regulates multiple ripening processes. Learning how to control this gene is the key last step in making ripening tomatoes develop full flavor, as well as ripe color.

Component III. Mechanisms of Plant Interactions with Other Organisms

Disease resistant peanut varieties with improved oil quality. Sclerotinia blight is a fungal disease that severely limits peanut production in the southwestern United States. Increasing the level of oleic acid in peanut oil is a health benefit and improves the frying value of peanut oil. ARS scientists in Stillwater, Oklahoma, in cooperation with Texas A&M and Oklahoma State Universities, will release two peanut cultivars with enhanced resistance to Sclerotinia blight and also high oleic acid content. These lines have the potential to enhance the cash flow to peanut growers in Oklahoma and Texas by $6 million annually.

Biotechnological approaches to protect peanuts from attack by viral diseases. Tomato-spotted-wilt-virus is a significant viral pathogen on peanut in the southeastern United States and is becoming a threat in the southwest. ARS scientists in Stillwater, Oklahoma, used biotechnological techniques to introduce the nucleocapsid gene from the disease organism into a runner-type peanut. This approach should produce transgenic plants with coat-protein mediated resistance to tomato-spotted-wilt-virus.

Development of natural products to control growth of algae. ARS scientists in Oxford, Mississippi, developed new technology for controlling algal blooms in farm ponds. The control agents are derivatives of anthraquinones, compounds that help destroy cells grown under sunlight. Two patents have been filed for a new algicide and a new class of antifungals for plants. Negotiations are ongoing with an industrial partner to license the anthraquinone derivative for use as an algicide.

Determining how aquatic weeds develop resistance to herbicides. Hydrilla verticillata, an invasive aquatic weed in the southern United States, has evolved resistance to the herbicide that is most commonly used and effective for its control. ARS scientists in Oxford, Mississippi, discovered a gene in the weed with a natural mutation that confers resistance to the herbicide. This gene encodes the enzyme phytoene desaturase. Studies have revealed other areas of the gene that can be mutated to give resistance and have demonstrated that there is cross-resistance to various herbicides.

Targeting transgenes to non-grain tissue in barley. A promoter that targets gene expression specifically to the lemma and palea, the outer covering (chaff) of barley grains, has been identified by ARS scientists in Madison, Wisconsin, in collaboration with the University of Wisconsin. This promoter can be used to insert protective and antifungal genes in the chaff around the barley grain. This promoter will enable plant scientists to better protect grain from pests that can harm cereal grains, without inserting the protective genes into the grain that is consumed.