



**Accomplishments Report  
(2005-2010)**  
National Program 215:  
Pasture, Forage, and Rangeland Systems



Agricultural Research Service  
U.S. Department of Agriculture

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## **Introduction**

### **Purpose of this Report**

This report summarizes selected accomplishments of the Agricultural Research Service's national program focusing on Pasture, Forage and Rangeland Systems. A key purpose of the report is to provide information for an external assessment of the program's performance over the past 5 years. This assessment will also provide recommendations for the future direction of the program. Because of space limitations not all of the program accomplishments have been presented.

### **The Agricultural Research Service (ARS)**

The ARS is the principal in-house research agency of the U.S. Department of Agriculture (USDA). It is one of the four component agencies of the Research, Education, and Economics (REE) mission area. An overview of the ARS mission, structure, planning process, and relationships with other agencies is provided in Appendix One.

### **National Program 215: Pasture, Forage and Rangeland Systems**

Our Nation's grass and shrub lands- including pastures, hay, feedstock, turf and range lands- provide forages, bioenergy, open spaces and ecological services that contribute significantly to our agricultural, environmental, economic, and social well-being. Rangeland, pasture, and forages together comprise about 55% of the total land surface of the United States, about a billion acres. Privately owned lands comprise about 45% of this amount. The Nation's 30-40 million acres of turf lands are found around homes, schools, municipal and commercial buildings, in our parks, greenbelts and recreational areas, and along our roadsides, airports and right-of-ways. These lands represent the largest and most diverse land resources in the U.S.

The goals of this national program are to contribute significantly to meeting national needs for food, fiber, fuel and ecosystem services by restoring, conserving and using the Nation's pasture, hay, turf, and range lands in a way that meets the objectives of the U.S. Department of Agriculture, and the REE (Research, Education and Economics) Mission Area. Goals and objectives are met through basic and applied research that develops and transfers economically viable and environmentally sustainable conservation, production and monitoring practices and strategies that incorporate an improved understanding of ecological and agronomic processes, enhanced plant materials, advanced technologies, and user-friendly decision-support tools based on sound science.

Outputs of National Program 215 (NP215) research support the "Actionable Strategies" of the ARS Strategic Plan associated with the following objective and performance measures:

#### **Objective 6.3: Conserve and Use Pasture and Range Lands Efficiently**

Healthy, vigorous plant communities on diverse lands protect soil quality, prevent soil erosion, and provide sustainable forage and cover for livestock and wildlife. They also provide fiber and a diverse habitat for wildlife, improve water quality and sequester atmospheric carbon dioxide. The four serious threats that pose an increasing risk to the values, goods, and services provided by public and private pasture and range lands are wildfire, invasive species, loss of open space, and reduced profitability. ARS works with public and private land stewards to maintain/enhance watersheds and landscapes and their environmental services. The Agency produces the scientific knowledge needed to actively manage pasture and range lands and maintain the health, diversity, and resilience of these ecosystems.

#### **Performance Measure 6.3.1: Improved management practices and technologies for managing pasture and range lands to improve economic profitability and enhance environmental values.**

**Actionable Strategies:** Provide new environmentally acceptable practices and technologies for controlling invasive weeds and reduce wildfires. Provide science-based information and technologies to better manage environmental services while improving the economic effectiveness of forage-based livestock, bioenergy, and turf systems. Provide improved grass and forage legume germplasm to meet economic and environmental objectives including conserving and improving pasture, forage, turf, and rangeland condition; plant vigor and diversity; water management; and environmental stewardship. Accelerate development of methodologies and technologies to measure and monitor pasture and range land health and assess and document the benefits of conservation practices. Provide cost-effective practices and strategies for restoring degraded pasture, forage, and range lands. Develop and promote grazing and harvested forage practices and technologies for livestock and bioenergy production that reduce input costs and increase sustainability. Develop and promote management practices and technologies that reduce animal exposure to toxic plants.

**NP215 Collaboration**

Solving today’s complex problems requires sustained research by interdisciplinary teams working at multiple scales. Creating such teams requires collaboration between (1) ARS scientists and locations; (2) between ARS scientists and scientists in other federal, state, local and private research institutions; and (3) between scientists and those citizens who use the products of research. An overview of ARS locations with projects in NP215 and internal and external collaboration is provided in Appendix Two.

**Beneficiaries of this National Program:** Since 2/3 of the world’s agricultural land is pasture, hay and range lands, the research done in this research program will benefit the country and the rest of the world in meeting their economic, social and environmental needs. Livestock producers and Natural Resource Conservation Service and Extension workers that provide information to land owners will benefit. The products of this program will benefit federal land stewardship agencies such as the Bureau of Land Management, Forest Service, National Park Service, Fish and Wildlife Service, Bureau of Indian Affairs, Department of Defense, and U.S. Geological Survey (USGS) that are responsible for almost a billion acres of public land. Beneficiaries include state land management agencies responsible for state-owned grazing lands, and policymakers and rural and urban community organizations that need information and technologies to assess and improve the management of the nation’s lands. Finally, the public at large will benefit through improved management of the Nation's pasture, forage, turf and range lands through greater economic opportunities, access to high quality food, and improved recreational opportunities, new bioenergy resources, and enhanced ecological services.

**Summary of Technology Transfer Activities**

<b>Activity</b>	<b>Number of activities</b>	<b>Total attendance</b>
Field days	276	14,484
Popular press articles	389	
Radio-TV items	1284	
Decision support tools	11	
Presentations to customers	980	44,192
K-12 School Events	439	19,496
Patents and CRADAS	27	
Germplasm releases	28	
Exhibits at Fairs	21	

## **Component 1: Rangelands Management Systems to Improve Economic Viability and Enhance the Environment**

**Integrating Across Research Problem Areas to Meet National Needs:** Developing science-based databases and decision-support tools for formulating land management strategies at a national level is an increasingly important priority. The research from its national network of laboratories over the past 50 years has positioned ARS scientists to make significant contributions in working with other Federal agencies to develop nationally oriented analyses, databases and decision-support tools essential to meet policy needs. Two examples where ARS grazingland research has contributed significantly to aiding other Federal agencies in meeting their mission objectives are CEAP (Conservation Effects Assessment Project) and ESDs (Ecological Site Descriptions).

### **Problem Statement A**

Need for economically viable rangeland management practices, germplasm, technologies and strategies to conserve and enhance rangeland ecosystems.

**Objective A.1.** Develop management and monitoring strategies and decision support tools that conserve natural resources while maintaining economic objectives.

### **Management Strategies and Decision-Support Tools**

Research in this section is focused on issues such as soil erosion, woody plant management, carbon sequestration, and estimating precipitation over broad areas. Some accomplishments provide decision support models, which can be used directly by managers, and some provide background information of value to both managers and policymakers.

**Saving the Nation's soil resources.** Nationally 20% of non-federal rangelands generate over 65% of the average annual soil loss, and between 23 and 29% (92-106 million acres) of the Nation's rangelands are vulnerable to accelerated soil loss. ARS scientists in Reno, NV with ARS scientists in Tucson, AZ and Boise, ID, and NRCS scientists developed the Rangeland Hydrology and Erosion Model (RHEM), a process-based model for assessing soil erosion rates on rangelands. RHEM allows NRCS to target the most vulnerable rangeland areas for soil erosion. RHEM provides a risk index which can describe which sites are most vulnerable and when the sites are most vulnerable (time of year) to soil loss. The RHEM tool also provide a means of evaluating alternative conservation practices to determine which practices are most effective at lowering the risk of accelerated soil loss. This new technology will allow USDA and its conservation partners to be proactive in preventing accelerated soil loss rather than concentrating on repairing degraded lands which is a far more costly approach. (Weltz et al., 2008)

**Assessing Erosion Risk after Fire.** In the year following a wildfire, soil erosion and excessive runoff are the primary risks. Public-land management agencies need tools to estimate this risk to identify high-risk land areas for mitigation and rehabilitation. ARS scientists at Boise, ID evaluated the ARS-developed WEPP model for simulating rangeland fire impacts and collaborated with the USFS to develop the WEPP-based Erosion Risk Management Tool (ERMiT). ERMiT is now used by land managers and Burned Area Emergency Response (BAER) teams in predicting erosion and sedimentation from burned areas on forest and rangeland ecosystems in the western United States. Since 2005, the web-based ERMiT has logged over 3400 simulation runs. (Moffet et al., 2007; Robichaud et al., 2007)

**Selecting Post-fire Erosion Barriers.** Resource managers are challenged with selecting the most effective erosion control treatments to implement following wildfires. The relative effectiveness of commonly used erosion barrier treatments was measured by ARS scientists in Boise, ID and the USFS using artificial and natural rainfall events. Results provide Burned Area

Emergency Response (BAER) Teams the ability to better select which erosion control treatments to apply to stabilize a landscape following fire. (Robichaud et al., 2008)

**Hydrologic Vulnerability of Juniper Encroachment.** ARS researchers from Boise, ID and Burns, OR investigated the hydrologic impacts of juniper encroachment into sagebrush steppe at multiple sites in the Great Basin. Runoff and erosion from degraded sites increased exponentially where tree dominance resulted in more than 50% exposure of bare ground. Tree removal by cutting was found to stimulate understory vegetation, facilitate intercanopy litter recruitment, and reduce bare ground area resulting in improved infiltration, decreased runoff volume and velocity, and reduced soil loss. Depending on vegetation response, tree cutting represents a potentially viable option for recruiting cover, decreasing runoff and erosion, and improving overall rangeland health on degraded wooded shrublands. (Pierson et al., 2010 and 2007a). ARS scientists also worked as part of a multidisciplinary effort to develop a science-based decision-making framework for implementing juniper control. This framework is used to prioritize sites to be treated, to select treatments, and predict response. (Miller et al., 2007)

**Impacts of Mechanically Shredding Juniper on Soil Erosion.** ARS scientists in Boise, ID investigated the impacts of using mechanical shredding for juniper control on soil compaction, infiltration and surface runoff and erosion. Experimental results indicate that minor soil compaction can result from the use of mechanized shredders; however, the shredded tree material sufficiently mitigates these impacts while also potentially reducing runoff and erosion in areas where shredding spreads tree residue beyond the canopy of the tree. Limited adverse and some positive hydrologic responses to shredding at the small plot scale indicate that shredding is a potentially hydrologically viable method that land managers can use for juniper fuel control. (Cline et al., 2010)

**Carbon sequestration on rangelands.** Increasing woodland cover in the West may increase carbon storage, but knowledge is limited about carbon distribution in these landscapes, especially in below-ground pools. ARS scientists in Reno, NV quantified the spatial distribution of soil carbon in pinyon-juniper woodlands, and determined prescribed fire's effect on soil C and N. Prescribed burning caused immediate increases in surface soil C and N, but over intermediate to longer time periods, no statistically detectable change in soil C or N occurred from burning. This research indicates that proposed fuel load reduction treatments within the pinyon-juniper woodlands of the Great Basin should have no impact on long-term carbon sequestration. (Rau et al., 2010)

**North American rangelands as carbon dioxide sinks.** In a collaborative regional experiment, ARS scientists used Bowen ratio micrometeorological tower methodology to measure carbon flows in four rangelands of the Great Plains, two in the desert Southwest, and two Northwest sagebrush steppe sites. Both sagebrush steppe sites functioned as carbon sinks, capturing carbon dioxide from the air through plant activity. Three of the four Great Plains grasslands were sinks, but the two Southwest hot desert sites showed a net annual release of carbon into the atmosphere. These results clarify the role of rangelands in the global carbon cycle, and provide information on how climate can affect carbon sequestration. (Poley et al., 2010a and 2010b; Svejcar et al., 2008; Gilmanov et al., 2006)

**Rainfall variation and soil carbon sequestration in rangelands.** Shifts in precipitation patterns accompanying global climate change are predicted to affect rangeland ecosystems significantly. Little work has been done on how soil processes could change as a result. At Burns, OR, ARS scientists observed that shifts in precipitation timing from winter to spring in cold desert ecosystems resulted in lower carbon inputs to the soil by plants while increased microbial activity led to increased decomposition. This resulted in as high as a 14% increase in carbon loss from the soil. This information will help develop regional and global models of the carbon cycle that aid policy makers in evaluating carbon sequestration strategies and evaluation systems. (Aanderud et al., 2010)

**Precipitation Estimation on Western Rangelands.** The timing and amount of precipitation are principal drivers of most rangeland processes, but the availability of rainfall-gauge data over rangelands is limited. The National Weather Service operates a network of Doppler radar stations that produces hourly rainfall estimates covering 96% of the conterminous U.S. ARS scientists in Boise, ID evaluated radar precipitation estimates at ARS research watersheds across the United States and determined that the network does a relatively good job of detecting high-intensity rainfall events, but uniformly underestimates total annual precipitation. Additional modifications of the NWS precipitation-processing procedures might improve utility of these data for rangeland modeling applications. However current data are useful for estimating high-intensity events that greatly affect soil erosion and flooding. (Hardegree et al., 2008b)

### **Monitoring and Assessing Rangelands**

The Conservation Effects Assessment Project (CEAP) has identified a need for improved monitoring of the impacts of rangeland management practices along with evaluating current condition and trends in rangeland status. This section focuses on multi-scale measurements (from remote sensing to plot level), emphasizing increased accuracy and cost-effectiveness.

**State-of-the-art remote sensing for rangeland management.** Research by ARS scientists at Mandan, ND has focused on modeling remote sensing-based data to estimate forage quality and quantity at the plant leaf scale, and has moved up to pasture and ecoregions scales. In cooperation with the U.S. Forest Service, these techniques were used to assess and monitor rangeland on the 150,000-acre Grand River National Grassland. An intensive field campaign was initiated in 2010 and preliminary results indicate the utility of space-borne and aerial optical data for estimating rangeland forage quality, quantity, and structure is excellent. The technology is being transferred to the USFS for a pilot test by field personnel. If the pilot is successful, the model will be extended to other national grasslands. (Phillips et al., 2009; Beerli et al., 2007; Phillips et al., 2006a)

**Quantifying shrub recovery after wildfires and management fires.** Precise estimates of shrub recovery rates after wildfires are unavailable. ARS scientists at Dubois, ID have validated using georeferenced, high-resolution aerial (VLSA) and satellite (SPOT) imagery to rapidly and accurately quantify shrub cover across large areas of sagebrush-steppe. Combining large-sample imagery and a 70-year historical fire record provided precise recovery-rate estimates for general shrub, horsebrush, bitterbrush, and sagebrush cover after fires in the mountain big sagebrush steppe. These recovery estimates are applicable to predict the optimal interval for prescribed burns and to plan landscape-scale prescribed fires to manage shrub cover in sagebrush steppe that is critical habitat for sage grouse, and used simultaneously for livestock production. (Moffet, 2009; Clark et al., 2008a)

**Landscape-level monitoring.** ARS scientists in Las Cruces, NM, working with BLM, developed a monitoring protocol based on ecological site descriptions and spatial processes. This protocol is being applied to restoration efforts for the Restore New Mexico program that is targeting over 1,000,000 acres at an anticipated cost of over \$20,000,000. The protocol examines how local patchiness and landscape context interact to determine remediation success. This science-based guidance will save millions of dollars by more effectively targeting management applications and will be used at other BLM locations. (Bestelmeyer et al., 2009a and 2009b; Herrick et al., 2008)

**Reducing costs in monitoring rangelands.** Low-cost monitoring using remote sensing has been limited by insufficient resolution for tracking changes in soils and vegetation. ARS scientists at Las Cruces, NM evaluated a combination of sensors and found that five-centimeter resolution aerial photographs from Unmanned Aerial Vehicles (UAV) provided adequate measurements of gap and patch sizes as well as percent bare soil and vegetation ground cover.

Software was developed for constructing mosaics from multiple images to monitor large study areas. Using a combination of images taken at different resolutions provided lower-cost and timely information needed to help managers identify rangeland areas at risk of degradation and degraded areas with good potential for restoration. (Laliberte et al., 2010; Rango et al., 2006)

**Applications of New Rangeland Monitoring Tools.** Measurements from images acquired using new tools and methods allow economical extensions of ground-based monitoring for uplands, riparian areas, allotments, and watersheds. These aerial tools have been used to systematically capture tens of thousands of aerial photographs over management areas in 7 western states. The aerial method is rapid, quantitative, repeatable, economical, and applicable at a variety of spatial scales. Extensive collaboration with other ARS units and with federal land management agencies, private enterprise, and NGOs has allowed the system to be tested for a variety of applications including rangeland health for uplands and riparian systems, status of invasive species and wildlife habitat, forecasting biofuel yield from juniper woodlands, and bringing new power to ecological investigations. (Booth et al., In Press, 2009, and 2008; Cox and Booth, 2008b; Sivanpillai and Booth, 2008; Blumenthal et al., 2007; Seefeldt and Booth, 2006; Booth et al., 2006a, 2006b, 2006c, and 2006d; Booth and Cox, 2006; Booth et al., 2005; Booth et al., 2004)

**Assessment Tool for Juniper Encroachment.** Western juniper has encroached upon over 8 million acres of sagebrush/bunchgrass rangeland in the western United States. Development of remote-sensing tools to map these ecosystems is essential for modeling current and future juniper distribution and to assign priorities for on-the-ground juniper management activities. ARS scientists in Boise, ID in collaboration with Idaho State University, and the Bureau of Land Management have developed new tools based on Light Detection and Ranging (LiDAR) remote sensing technology for detecting juniper invasion trajectory and phase, and the health and status of underlying sagebrush-steppe vegetation. They developed new algorithms for measuring lower-stature sagebrush vegetation that is at the current limit of LiDAR detection; quantified historical patterns and phase of juniper invasion; and, developed models to predict the trajectory of future juniper invasion. This information will be used with other remote sensing tools to identify and prioritize areas for western juniper management by BLM. (Glenn et al., 2010; Sankey et al., 2010; Spaete et al., 2010)

**Quantifying Stream Shading.** Existing techniques for assessing stream shading require costly equipment and instrumentation and only make indirect assessments. ARS scientists at Boise, ID developed and evaluated new, effective, and low-cost field and image analysis techniques for assessing stream surface shading from digital images. These techniques will allow researchers, natural resource managers, and environmental quality regulators to more efficiently and effectively monitor stream shading and investigate its relationship with stream temperature variability. (Clark et al., 2008b)

**Efficient Vegetation Biomass Sampling.** Tradition methods for sampling vegetation biomass are time-consuming, expensive, and, because they are destructive, they prevent repeated measures. ARS researchers at Boise, ID examined cost effectiveness of using a non-destructive, point sampling technique to stratify variability in subsequent biomass sampling on a sagebrush-bunchgrass rangeland sites. Double-sampling strategies where half of the point-sampled plots were also measured for biomass yielded a cost savings of 39% with relatively minor reduction in biomass sample precision ( $18 \pm 4\%$ ). Agricultural producers, natural resource managers, and research will benefit from cost savings and increased availability of biomass information accruing when sampling cost constraints are reduced. (Clark et al., 2008a)

## **Plant Genetic Diversity**

Restoring rangeland requires knowledge of appropriate genetic materials, and a broad base of plant materials from which to select. Recent research has been focused on applying newer genetic techniques to quantifying characteristics of both native and introduced rangeland plants.

**Genetic diversity of plant material.** Public land managers, seed companies, and other stakeholders are concerned about the genetic identity and diversity of plant materials used for large-scale revegetation in the western U.S. With the increased emphasis to use locally adapted germplasm, an understanding of genetic diversity and species relationships is critical. DNA fingerprinting techniques can identify and characterize natural groups of key plant species (Weising et al., 2005; Lee et al., 2005), which provide land managers with a better knowledge base for making revegetation and management decisions. Researchers in Logan, UT have used high-throughput DNA sequencing and DNA genotyping to characterize natural population structure of native and introduced plant materials across this region. Species studied include purple needlegrass (Larson et al., 2001a); bluebunch wheatgrass (Jones et al., 2002; Larson et al., 2000; Larson et al., 2004); Utah sweetvetch (Karudapuram and Larson, 2005; Bushman et al., 2007b); basalt milkvetch; western prairie clover (Bhattarai et al., 2010b); Searls Prairie clover (Bhattarai et al., 2010a); western wheatgrass (Larson et al., 2003b); Altai wildrye (Jensen et al., 2005a); Russian wildrye (Jensen et al., 2005b; Jensen et al., 2006a); meadow brome (Jensen et al., 2004); crested wheatgrass (Jensen et al., 2006b; Jensen et al., 2009a); Siberian wheatgrass (Jensen et al., 2009b); differentiation between big squirreltail (*Elymus multisetus*), bottlebrush squirreltail (*E. elymoides*), four landraces of *E. elymoides* ssp. *brevifolius* (Larson et al., 2003a), and eight other *Elymus* wheatgrass species. Among other things they found that North American Roemer's and Idaho fescues are genetically distinct; that cultivars of Joseph and NezPurs are hybrids of *Festuca roemerii* and *F. idahoensis*; and, identified hybrids between Idaho fescue and naturalized sheep fescue (*F. ovina*). Other outputs of this line of work included developing a simplified taxonomic treatment of the phylogeny of the South American *Bromus* sect. *Certachloa* taxa (Massa et al., 2001; Massa et al., 2004); identification of bottlebrush squirreltail landraces used in developing and releasing 'Fishcreek' and 'Toe Jam' germplasms and the reevaluation of subspecies classifications (Jones et al., 2004a,b); and, Fescue research confirmed recognition of Roemer's species and identified need for more natural fescue varieties. (Jones et al., 2008).

**Characterization of perennial grass species.** Grass performance across multiple locations and environments is paramount when making seeding recommendation. Due to the chronic and often severe disturbance of western North American rangelands, revegetation with perennial grasses is used to rapidly stabilize soil resources and provide competition against annual invasive plant species. An understanding of native and introduced perennial grasses over multiple environments and ecotypes is often limiting. ARS scientists at Logan, UT; Lincoln, NE; Mandan, ND; Miles City, MT; and El Reno, OK brought together results of simulated revegetation plantings at numerous sites across the Great Plains and Intermountain regions of the U.S. to document the performance and adaptation of commonly used native and introduced cool-season grass species. Performance and adaptation was species specific. Some species such as crested wheatgrass (*Agropyron* spp.) exhibited broad adaptation by resulting in successful revegetation at a wide range of locations. Other species showed more limited revegetation success and range of adaptability. Additionally, for most of the species the interaction between the species and the environments (genotype by environment interaction) was large. Thus, the implications are that for large-scale revegetation projects, land managers should have a good understanding of the site characteristics of the project to ensure proper selection of appropriate perennial grass species. With these results land managers can make better informed decisions when choosing which species to include in mixtures and which to avoid. (Robins et al., 2007)

**Classifying *Kochia*.** Forage kochia is not closely related to two other *Kochia* species. Introduced from central Eurasia, forage kochia—*Kochia prostrata*—is a useful winter forage as well as wild-fire deterrent. However, environment conservationists are worried about the possibility of its contamination by another introduced species *Kochia scorparia*, which became a

noxious annual weed. Greenmolly or perennial summer cypress (*Kochia americana*) is probably the U.S. native equivalent of forage kochia. Scientists at Logan, UT determined the genomic relationships among these three *Kochia* species and concluded that they are so distantly related that there is little chance of hybridization between any two of them. This information removed the obstacle for the acceptance of forage kochia in the revegetation planting after wildfires in the semi-arid regions of the U.S. (Lee et al., 2005)

**Distinguish native from introduced *Glyceria* species.** Mannagrass species native to Europe have been introduced into North America, and the *Glyceria declinata* species has become invasive. It is impossible to accurately distinguish *G. declinata* from several native species using taxonomic measurements. Scientists at Logan, UT developed DNA molecular markers to distinguish native from introduced *Glyceria* species, and especially the invasive *G. declinata* from visually similar species. These markers saved California conservation efforts over \$1 million by showing that they were inadvertently planting the invasive *G. declinata*. Using these markers helped Oregon annual ryegrass growers eliminate seed lots contaminated with unknown *Glyceria* so international shipments could be made. Since 2006, 56 seed lots valued at \$8,000 per lot were approved for shipment to Australia. (Bushman et al., 2009; Gerlach et al., 2009; Bushman et al., 2007a; Whipple et al., 2007)

**DNA monitoring and endangered species.** Monitoring strategies are needed to better understand biological relationships between endangered species, their wild relatives, and the potential impact on rangeland function. DNA fingerprinting techniques can identify and characterize biologically these species. The proposed listing of slickspot peppergrass (*Lepidium papilliferum*) as an endangered range plant species could have a significant impact on land management agencies and ranching activities in southwestern Idaho. In cooperation with the BLM and Idaho Office of Endangered Species, ARS Logan, UT researchers used DNA sequencing and DNA fingerprinting techniques to test genetic similarities between slickspot peppergrass and other plants in the widespread *L. montanum* complex. With this information, and other data collected by the U.S. Fish and Wildlife Service, the U.S. Fish and Wildlife Service decided not to list slickspot peppergrass as an endangered species. This decision prevented significant regulatory costs and disruptions in livestock production, military activities, and other land use activities on over 20,000 acres. (Larson et al., 2010; Smith et al., 2009; Weising et al., 2005)

### **Management Framework**

There is a critical need for rangeland scientists and managers to have a common language and vision for discussing vegetation change, response to treatments, and landscape scale. If the profession is to adjust management based on monitoring information, there is a need for an adaptive framework which is clear and straight-forward.

**State-and-transition models (STMs) to describe rangeland dynamics.** Long-term data are often unavailable to test STMs. ARS scientists in Las Cruces, NM used experimental and inventory data to test the limits of resilience in a desert rangeland STM, providing a clear illustration of how diverse datasets can be incorporated in STMs. This approach provides a mechanism for interpretation of monitoring and assessment data for the NRCS Conservation Effects Assessment Program (CEAP) for grazing lands. Interpretations enable decision-making to target restoration activities and to modify management to increase rangeland sustainability. (Bestelmeyer et al., 2009b; Briske et al., 2008a)

**Managing complex problems on sagebrush steppe rangelands.** Current-day management of rangelands is made more difficult by the increasing complexity of problems that managers are confronted with due to environmental variability in space and time. ARS scientists at Burns, OR examined the dynamic nature of complex problems on rangelands and proposed techniques for addressing topical issues. When the causes of problems vary in space and time, solutions must

remain dynamic and are best tackled with adaptive approaches set within an appropriate ecological framework. Implementing adaptive management programs will involve overcoming institutional paradigms, emphasizing biological measures of success, forming strong interdisciplinary partnerships, and working within process-based ecological frameworks that allow advancement through both validation and constructive dissent. (Boyd and Svejcar, 2009b; Reeve Morghan et al., 2006.)

**Objective A.2.** Determine impact of livestock grazing, fire, mechanical treatments, and drought on ecological integrity and watershed structure and function.

### **Livestock Grazing**

Livestock grazing remains one of the most widespread land uses in the western United States. There are many questions about the specific impacts of livestock grazing on ecological services such as wildlife habitat, carbon sequestration, and water quality. Public land managers require scientific documentation to use in NEPA documents if the documents are to stand up to public scrutiny. Compatibility of grazing with sage-grouse habitat and with the Clean Water Act have become big issues in the western US.

**Moderate livestock grazing compatible with sage-grouse habitat.** Livestock grazing has been indirectly blamed for sage-grouse declines in the western United States and southern Canada; however, there is a lack of scientific research that directly relates the two. ARS scientists in Burns, OR conducted a study to determine the level of utilization at which cattle begin to access herbaceous vegetation under the canopy of sagebrush plants; these herbaceous plants can provide important screening cover for nesting sage grouse. Results suggest that cattle use of understory plants was minimal until pasture utilization exceeded about 35% utilization by weight. These results can be used to help land managers develop grazing plants which minimize the impacts of grazing livestock on herbaceous cover important to nesting sage-grouse. (France et al., 2008)

**Grazing and environmental effects on soil carbon dynamics:** Long-term livestock grazing in the dry, shortgrass ecosystem increased ecosystem carbon storage compared to non-grazed areas, but ecosystem carbon storage decreased with long-term livestock grazing in the relatively wet midgrass and tallgrass ecosystems. Sagebrush steppe and Great Plains rangelands functioned as sinks for carbon whereas Southwest hot desert sites were sources of carbon on an annual basis. Periods of carbon uptake during any one year are brief, often lasting less than three months. These findings show that proper grazing management of rangelands can enhance soil organic carbon sequestration and reduce carbon dioxide in the atmosphere. (Morgan et al. 2010; Schuman et al., 2009; Ingram et al., 2008; Morgan et al., 2008; Derner and Schuman, 2007; Potter and Derner, 2006; Derner et al., 2006b)

**Animal bedding and stream crossing.** Data have not been available to characterize the environmental effects of concentrated animal bedding and stream crossing. ARS scientists in Dubois, ID, in collaboration with scientists at ARS-Kimberly, ID and ARS-Boise, ID quantified nutrient loading and infiltration, soil erosivity, and hydrologic dynamics in bedground sites, and sediment and microbial translocation in stream-crossing sites. Subsequently, translocation rates of nutrients and soil, and sediment and bacteria were developed for animal bedding and crossing activities, respectively, in subalpine ecosystems. These translocation estimates are applicable for identification and selection of appropriate bedding and stream crossing sites for livestock that are grazing subalpine ecosystems. (Leytem and Seefeldt, 2008)

**Minimizing Water Quality Impacts of Rangeland Sheep Production.** Excessive suspended solids and pathogens (e.g., *Escherichia coli*) are among the most prevalent water quality impairments in rangeland streams worldwide. Effects of herded stream crossings by domestic sheep on suspended solids and *E. coli* in stream water, however, are largely unknown. ARS

researchers from Boise and Dubois, ID evaluated the spatial and temporal effects of herded, one-way crossings by sheep bands on total suspended solids (TSS) and *E. coli* concentration and load responses in water downstream from crossing points on perennial streams in the Centennial Mountains of eastern Idaho/southwestern Montana. Crossings produced substantial, but short-lived elevations in TSS and *E. coli* concentrations and loads in downstream waters but these effects declined rapidly over time and with downstream distance. Survival of *E. coli* introduced during crossings was quite limited. These results conclude that herded stream crossings by sheep are not a serious water quality concern in cold water streams. (Clark et al., 2011c)

## **Fire**

Next to livestock grazing, prescribed fire may be the most widely applied land management practice on rangeland. And if wildfires are considered, the effects of burning on rangeland resources are of wide interest to many rangeland users and managers. Many rangeland biomes evolved with fire, but human settlement and introduction of invasive species such as cheatgrass have drastically altered fire cycles.

**Hydrologic Impacts of Fire.** Many land managers of sagebrush rangelands assume burning creates soil water repellency and that post-fire reductions in infiltration capacity are directly attributable to fire-induced repellency. ARS scientists in Boise, ID conducted multiple studies of the hydrologic impacts of wildland fire and prescribed fire used to reduce fuel loads on mountainous sagebrush rangelands. Fire consistently increased concentrated overland flow and erosion compared to unburned conditions until groundcover returned to above 60%; this typically took at least two growing seasons. Soil water repellency decreased infiltration capacity by up to 50%, but was found to be naturally occurring on unburned sagebrush rangelands with burning typically reducing or having no effect on the strength of post-fire repellency. Soil water repellency and infiltration capacity were highly variable over time for unburned as well as burned conditions and pre-fire repellency strength exerted more influence on post-fire infiltration than burning. This information advances the fundamental understanding of burning sagebrush rangelands, provides land managers an improved ability to assess the hydrologic effects of fire, and is the foundation for development of fire erosion modeling tools. (Pierson et al., 2009, 2008a, and 2008b).

**Effects of prescribed fire on sagebrush steppe goods and services.** ARS scientists in Burns, OR conducted a series of studies to evaluate the effects of prescribed fire on wildlife habitat, forage production, and resistance to invasion by annual grasses. The results provide information land managers can use to make informed decisions about the appropriate use of prescribed fire. This information has been used in a variety of land use planning documents developed by state and federal agencies. (Rhodes et al., 2010; Davies et al., 2009a; Davies and Bates, 2008; Davies et al., 2008; Davies et al., 2007b)

**Fire as a range management tool.** Wildfires are part of rangeland ecosystems, and prescribed fires are part of programs to manage rangeland ecosystems, including shrub dynamics. Scientists at Dubois ID, and their collaborators used data from very-large-scale-aerial imagery to characterize the changes in horsebrush, bitterbrush, and sagebrush cover after wildfires and prescribed fires. (Manuscript in review by a peer-reviewed scientific journal.)

**Controlling grasshoppers with fire.** Grasshoppers in rangeland systems play important ecological roles in nutrient cycling and as a source of prey for other animals. When their numbers get too high, however, they become serious pests, consuming more than 20% of the forage needed by wildlife and livestock. ARS scientists at Miles City and Sidney, MT studied the interrelationship between fires and the population of *A. deorum*, a grasshopper that lays its eggs just below the soil's surface. They found that when there was a prescribed burn with 4,500 kg/ha of plant biomass to serve as fuel, none of the eggs of *A. deorum* hatched in 86% of the trials. In

contrast, the eggs of *M. sanguinipes*, a grasshopper that lays its eggs deeper in the soil, were not affected by the heat. Other grasshopper species that lay their eggs at intermediate depths have moderate rates of mortality. These data show a clear relationship between the depth of the egg in the soil and mortality during fires. Land managers can still use fire to help control species that lay their eggs at greater depths by timing the fire to coincide with the eggs hatching and the wingless nymphs emerging on the surface. (Branson and Vermeire, 2007)

### **Grazing by Fire Interactions**

Because these two land management practices are so widespread on rangelands, it is critical to understand some of the interactions. There are many questions, especially on public land, about how to manage grazing after wild- and prescribed fire. Some agencies have prescriptive rest periods following fire, but there is a limited research basis to help managers make decisions about grazing management following fire.

**Prescribed Fire Effects on Cattle Resource Selection.** Prescribed fire is commonly applied world wide as a tool for enhancing habitats and altering resource selection patterns of grazing animals. A scientific basis for this management practice has been established in some rangeland ecosystems (e.g. montane grasslands, tall grass prairie, mixed prairie, shortgrass prairie, shrub steppe, and savanna) but its effectiveness has not been rigorously evaluated on mesic sagebrush steppe rangelands. ARS researchers in Boise, ID evaluated resource selection of beef cattle using global positioning system (GPS) collars for 2 years prior to and for 5 years after fall prescribed burns. Resource selection functions (RSF) developed from these data indicated cattle selected for lightly to moderately burned areas for all 5 post-fire years. Cattle had previously avoided or were neutral towards these burned areas during the 2 years prior to the fire. Resource selection responses to the fire lasted 2-3 years longer than expected based on work in other ecosystems. This work demonstrates that prescribed fall burning is an effective tool for managing cattle distribution on mesic sagebrush steppe and for shifting concentrated cattle use away from critical resources such as stream riparian areas and moist upland meadows. (Clark et al., 2011a and 2011b)

**Post-fire grazing management.** Livestock producers in the Great Basin are frequently not allowed to graze livestock on public rangeland for at least two years after a wildfire, adversely affecting their profitability. ARS scientists at Burns, OR found that livestock grazing the year after a fire need not damage sagebrush rangeland. A 5-year study showed that moderate grazing either one or two years after a fire was not different from deferring the rangeland for two full years. The research was conducted on sagebrush rangeland in a good condition. Results could vary depending on rangeland type and condition prior to the fire. These findings show that deferring grazing for two-years is not always necessary. Consequently, grazing deferral decisions after fire should be based on local conditions. (Bates et al., 2009)

**Grazing reduces fire impacts and risk.** ARS scientists from Burns, OR evaluated the influence of grazing on fire impacts and risk in rangelands. To measure grazing effects on fire impacts, rangelands were either grazed until the year prior to burning (1993) or have been excluded from grazing since 1937. Vegetation measurements were taken 12, 13, and 14 years after the fire. Burning caused a huge increase in cheatgrass (an invasive annual grass) in the ungrazed areas, but not in the grazed areas. In a related study, they also found grazing exclusion on sagebrush rangeland can result in more frequent and larger fires in native rangelands that would be more difficult to suppress because of fuel buildup and smaller gaps between fuels. More frequent fires can increase invasive weeds and reduce desirable forages for livestock and wildlife. (Davies et al., In press; Davies et al., 2009c)

### **Soil and Weather**

Rangelands are characterized by large spatial and temporal variation; both site and yearly weather variation have large impacts on productivity and habitat quality. In many biomes,

knowledge of plant productivity as influenced by site and weather is largely anecdotal and not well documented in the scientific literature. Reducing soil erosion is critical to maintaining adequate site productivity.

**Site potential of sagebrush plant communities.** There is wide variation in the ability of sagebrush steppe plant communities to produce the ecological goods and services demanded by society. Scientists at Burns, OR conducted a series of studies which will help agricultural and natural resource managers set realistic expectations for productivity and habitat values for a variety of sagebrush vegetation types. (Davies and Bates, 2010a; Davies et al., 2007a and 2007c)

**Plant symbiotic relationships.** Microbial endophytes are often undetected but influence numerous plant survival mechanisms. ARS scientists at Las Cruces, NM have been studying the impacts of endophyte colonization on the survivability of desert shrubs to improve plant establishment on degraded sites. Metagenomic analysis of seed-borne endophyte communities revealed a complex consortium of 24 species that persist and systemically colonize micropropagated plants. The consortium includes nitrogen fixing and thermophilic bacterial strains in addition to putative halophytes. This previously unrecognized seed-borne endophyte diversity has potential for optimizing microbial endophyte interactions to enhance plant adaptation and productivity. The symbiotic relationship between endophytes and desert plants may provide opportunities for managing microbial populations to improve host plants' ability to regulate water loss and improve the establishment and persistence of desirable plants on degraded sites. (Barrow et al., 2008; Lucero et al., 2008a and 2008b; Barrow et al., 2007)

**Drought Risk Management.** Drought is common in the northern Great Plains and is an important risk for livestock producers and rangeland managers. Traditionally, following a spring drought, managers delay making decisions such as reducing livestock numbers in the hope that summer rains will compensate for production lost in the spring. ARS scientists at Miles City, MT found that summer rains can increase forage production but these increases were seldom large enough to compensate for the 50% reductions caused by spring droughts. For example, the probability of getting 6 inches of rainfall in July and August which would have a significant impact on forage production is less than 1%. This information can aid livestock producers and land managers in making timely and realistic risk-management decisions on when livestock numbers should be reduced because of drought. (Heitschmidt and Vermeire, 2006)

**Impacts of Rangeland Seeding Practices on Erosion:** Mechanical seeding treatments used to establish rangeland vegetation can disturb surface soils and negatively impact runoff and erosion rates. The amount and persistence of impacts caused by different mechanical seeding treatments were quantified by ARS scientists in Boise, ID using artificial rainfall. The rate of recovery in hydrologic response for each treatment was controlled by the rate at which total surface cover increased until the amount of bare soil was reduced below critical levels. Results help land managers to evaluate revegetation projects in terms of their efficiency in obtaining the desired plant community balanced against the consequences of short-term elevated runoff and erosion risk associated with bare soil exposure. (Pierson et al., 2007b)

**Objective A.3:** Identify factors such as landscape position, species composition, land use history, management strategies, and climatic variables that can be used to predict and minimize the risk of degradation of rangeland ecosystems.

Rangeland environments are recognized as highly heterogeneous both over time and across landscapes. Typically, management practices respond to temporal heterogeneity, such as periodic droughts or wildfires. Increasingly, though, research has been devoted to better understanding heterogeneity of location on effects of management and how those effects should be evaluated and managed.

**Ground based indicators of ecosystem processes.** Cost effective vegetation and soil measures are needed that reflect status of underlying processes. New indicators of soil susceptibility to wind erosion were identified by ARS scientists at the Jornada unit in Las Cruces, NM. These indicators were combined with vegetation indicators previously developed by the Jornada. This information was incorporated with data from the National Resource Inventory to provide assessment of resource status across the western US. One observation was that non-native species currently occur on nearly 50% of non-federal rangelands, and account for at least 50% of plant cover on over 5% of these lands. This information will be used by the BLM, NRCS, Department of Defense, and NPS to evaluate effects of management practices on the status of over 100 million acres of western US rangelands. (Herrick et al., 2010; Li et al., 2010)

**Shrub microsite influences post-fire perennial grass establishment.** Woody plants can cause localized increases in resources that may persist after fire and create a heterogeneous environment for restoration. ARS scientists at Burns, OR tested the hypothesis that burned sagebrush subcanopies would have increased seedling establishment and performance of post-fire seeded perennial bunchgrasses compared to burned interspaces. Results indicated that one year post-seeding, subcanopy microsites had dramatically higher seedling density and taller seedlings than interspace microsites. These data quantitatively underscore the importance of shrub conservation for providing microsites with elevated chances of restoration success, particularly in low elevation sagebrush communities where establishing perennial grasses can help reduce susceptibility to invasion by exotic annual plants. (Boyd and Davies, 2010)

**Importance of landscape connections in successful restoration.** Remediation success will benefit from improved understanding of landscape connectivity and effects of prior management activity. ARS scientists at Las Cruces, NM analyzed runoff barriers installed over 70 years ago in southern New Mexico using time series aerial photography and satellite imagery. Persistent effects on vegetation patterns were observed but the magnitude of the effect varied with landscape location. Improved interactions between sites can increase resource retention and plant establishment at plant and patch scales, and land managers can use this information to identify landscape locations where conservation practices that modify soil and vegetation are more likely to improve soil and water resource retention. (Rango and Havstad, 2009)

**Simulation modeling of species composition over time.** ARS scientists at Las Cruces, NM developed a soil-water dynamics simulation model to integrate knowledge on factors affecting historic shifts in plant composition to predict future dynamics. The establishment of black grama, an important native grass, under historic (1850s) and current vegetation and soil conditions was compared to explain the shifts in vegetation dominance from grass to shrubs. Plant establishment appeared to be affected more by changes in soil properties than by changes in the vegetation present. These results can be used to help identify landscape locations where soils favor restoration efforts. (Peters et al., 2010)

## **Problem Statement B**

Need for improved livestock production systems for rangelands that provide and use forages in ways that are economically viable and enhance the environment sustainable.

**Objective B.1:** Develop monitoring and decision-support tools and management strategies for land managers.

Management practices need to recognize the influences of specific uses, and these studies build on history of rangeland research conducted by the ARS to examine interactive effects of livestock grazing as an influence on management responses to specific practices.

**Decision-support systems for simulation of forage production and livestock gains.** A newly developed decision support system for the central Great Plains, the Great Plains

Framework for Agricultural Resource Management (GPFARM) model was used to simulate forage production in shortgrass prairie and cow-calf production in northern mixed-grass prairie. The forage module displayed good agreement in tracking plant functional group growth and senescence trends, although warm-season grass biomass peaked earlier compared to observed data. The cattle module performed well and tracked cow and calf pre- and post-weaning weights. This model has functional utility for rangeland and livestock production system managers as a decision support system to manage risk. (Andales et al., 2006 and 2005)

**Biochemistry of juniper diet selection by livestock.** Juniper is a widely available plant but not typically used by livestock. ARS scientists at Las Cruces, NM studied supplementation options to increase juniper consumption. Sheep and goats were fed one-seeded juniper branches following basal diets with either a rumen degradable protein supplement, nondegradable protein supplement, or a control with no additional protein. Goats had higher juniper intake than sheep, and intake of juniper was greater for both species with protein supplements compared to controls. Juniper intake was lowest when the juniper secondary chemical compounds were greatest. Land managers can use these results to modify livestock foraging habits to increase juniper consumption by varying the type of supplement used. Increasing juniper consumption when other rangeland forages are in short supply will reduce the need to feed hay or reduce livestock numbers. (Utsumi et al., 2010 and 2009)

**GPS Animal Tracking Systems for Managing Livestock-Ecosystem Interactions.** Tracking wildlife and livestock movements and habitat use patterns accurately and affordably is essential to effective ecosystem management. ARS scientists at Boise, ID published the manufacturing details of the Clark Animal Tracking System (ATS), a GPS-telemetry animal tracking system that solves many of the problems associated with GPS technology in the past. Previous systems were very expensive, had relatively small data storage capacity, and were often difficult to use in the field. The ARS scientists have transferred the new technology to the Idaho Department of Fish & Game, USDA/APHIS/WS, USFS, and USDI/FWS. These agencies are working collaboratively to use the data being collected to evaluate habitat use and activity patterns of livestock and wildlife in response to prescribed fire treatments and programs to reintroduce predators. (Clark et al., 2006b)

**Measuring Thermal Environments of Animals on Rangelands.** Air temperature varies across a landscape and can influence where livestock spend time grazing. Mapping spatial changes in temperature can help to predict grazing animal distribution and use patterns. Measurement of air temperature requires equipment that must be shaded from the sun using shields that are fragile and expensive. Low-cost, custom solar shields were constructed by ARS scientists in Boise, ID and tested as alternatives to commercially available shielding. Instruments were able to measure air temperature to within approximately 3°C of actual air temperature and were approximately 10% cost of deploying commercial shields. These shields can significantly reduce the overall cost of intensively measuring air temperature at many points across extensive and complex landscapes. (Clark et al., 2006a)

**Pre- and post-weaning Cow behavior.** Information on spatial and temporal use of landscapes by livestock during different production phases is crucial for rangeland management and optimizing animal distribution. ARS scientists in Las Cruces, NM continuously monitored location of cow-calf pairs using GPS electronics attached to individual cows before and after weaning to document foraging behavior and rate and distance traveled. Distance traveled increased and spatial use of the landscape for foraging changed after weaning. Understanding the effects of husbandry practices on animal behavior can be used to more effectively manage livestock on rangelands.

**Objective B.2:** Develop and evaluate germplasm for improving forage quality, availability, and production.

One of the primary limits to rangeland animal production is the low forage quality typically inherent to native rangeland forages. One factor in evaluating improvement to forage production are resulting improvements in forage quality that can be achieved through plant breeding or targeted forage management practices.

**Increasing forage production and quality on marginal lands under reduced irrigation.**

Recently, 'Cache' Meadow brome was released by ARS scientists at Logan, UT for use on non-irrigated or irrigated pastures with increased seedling establishment, forage production under limited irrigation and forage quality. It "greens up" 2 to 3 weeks earlier extending the grazing season. It is attractive to western US producers because it is adapted to slightly acidic to mildly alkaline soils on dryland pastures, extremely winter hardy, and recovers quickly after grazing. This cultivar is quickly becoming the meadow bromegrass of choice on marginally production cropland. Based on Foundation Seed sales through 2010 (5,372 lbs), revenue from certified seed sales are currently estimated at greater than \$2,500,000. More recently, the yellow-flowered alfalfa subspecies variety 'Don' was released because of its persistent and non-aggressiveness in mixed plantings (legumes, forbs, and grasses) for use with limited or no irrigation on disturbed rangelands and irrigated pastures. In grazing situations, stands of 'Don' persist while sativa-type alfalfa decreased by 40% in grass mixtures over three years. (Jensen et al., 2004; Peel et al., 2009)

**Forage kochia improves sustainability of livestock production.** Low forage production and poor nutritional content of western rangelands limits the value of these rangelands for fall and winter grazing. Forage kochia provides increased amounts of nutritious forage during the fall and winter ARS scientists in Logan, UT in cooperation with animal scientists with Utah State University, compared the carrying capacity and livestock performance of traditional winter pastures dominated by crested wheatgrass (*Agropyron desertorum*) and cheatgrass (*Bromus tectorum*) versus similar rangelands seeded with forage kochia (*Kochia prostrata*) and crested wheatgrass. The rangelands with forage kochia had higher crude protein, six-fold higher carrying capacity, and cattle were in better body condition as compared to the non-treated rangelands). Additional studies documented that cattle grazing forage kochia during fall and winter reduced daily winter feeding costs by 25% while resulting in comparable body condition and back fat as those that were fed alfalfa hay. New, larger-statured ARS-developed forage kochia germplasms are highly palatable to cattle and have potential to revegetate highly eroded rangelands in the US and elsewhere. Range livestock producers throughout UT, ID, WY, CO, OR, and NV who manage an estimated 8 million acres of rangeland and 300,000 head of cattle and sheep have now planted hundreds of thousands of acres of forage kochia on public and private rangelands. (Bailey et al., 2010; Waldron et al., 2010a, 2010b and 2006b; Monaco et al., 2003c)

**Objective B.3.** Reduced economic losses to the livestock industry caused by rangeland poisonous plants and enhance food and feed safety.

**Poisoning of Livestock by Various Larkspur Species (*Delphinium*)**

Larkspur (*Delphinium* spp.) poisoning causes serious economic losses approaching \$10 million to livestock producers from dead animals and lost grazing opportunities.

**Factors affecting cattle susceptibility to larkspur intoxication.** Studies by ARS scientists in Logan, UT are determining the molecular mechanism of larkspur poisoning. Individual animal variation, breed differences, livestock species variability (sheep vs. cattle) and rodent models are being utilized to evaluate the physiological and molecular basis for this difference. Sheep are 4-5 times more resistant than are cattle; individual cow variation and breed differences have been identified and 4 mouse strains have been evaluated demonstrating a 2X difference between strains. Breed differences may be substantial. This genetic difference will be exploited in cattle to reduce losses by selecting resistant individuals or breeds to graze larkspur infested pastures.

**Physiological effects and toxico-kinetics of larkspur alkaloids in cattle.** Intoxication of cattle by larkspur alkaloids is highly related to dose and rate of elimination of individual alkaloids. ARS scientists at Logan, UT evaluated physiological effects and measured absorption and elimination of the two major alkaloids from larkspur in cattle: methyllycaconitine (MLA) and deltaline (DLT). Maximum serum concentrations were reached 10 hours after dosing and MLA was eliminated at a slower rate ( $E-t_{1/2} = 20.5$  hours) when compared to DLT ( $E-t_{1/2} = 8.2$  hours). This research determined that 99% of the alkaloids are eliminated within 7 half lives. Understanding clearance times for natural toxins is important for food safety considerations, and to understand alkaloid kinetics for use in management recommendations about treating intoxicated animals. (Green et al., 2009a, 2009b, and 2009c)

**Methyllycaconitine (MLA) potentiates nicotinic acetylcholine receptors (nAChR).** Relatively little research has been done on the pharmacology of MLA at muscle-type nAChRs that results in the collapse and death from larkspur poisoning. Cell culture-based studies showed that MLA at low concentrations potentiates the response of specific receptors to agonists, suggesting that MLA acts to increase the activity of these receptors indirectly through activation of a site on the protein. At high concentrations, MLA does not provoke a response itself after binding to a receptor, but dampens agonist-mediated responses. Understanding the mechanism of MLA action provides insight into the clinical signs of cattle poisoned with larkspur and was used to refined treatment protocols. (Welch et al., 2010; Green et al., 2009c)

**Additive toxic effects of the two most important larkspur alkaloids.** MDL (less toxic) and MSAL (highly toxic) alkaloids occur together in larkspur species. Most toxic effects of larkspur have been attributed to MSAL alkaloids alone, although MDL alkaloids are often a substantial component of the total alkaloid mixture. Research using mouse models determined that the less toxic MDL alkaloids exacerbate larkspur toxicity through an additive effect thus increasing toxicity of the more toxic MSAL alkaloids. So when assessing the toxic potential of larkspur plants both the MSAL alkaloid content and the MDL alkaloid content need to be determined. (Welch et al., 2009)

**Consumption patterns and toxicity of tall larkspur.** *Delphinium occidentale* has been problematic for cattle producers on many western Montana rangelands. Grazing studies were conducted on timothy grass-dominated rangelands to determine the timing and amount of larkspur ingestion in relation to phenology and quality of other forage. It was determined that cattle grazed larkspur during the vegetative and bud stages because timothy grasses are slow to grow and other palatable forbs are lacking. This is different than other larkspur infested ranges where cattle generally do not graze larkspur before flowering. The results of this research have been incorporated into management recommendations in this region and have reduced cattle losses. (Welch et al., 2008)

**Consumption of *Delphinium andersonii* by cattle.** Grazing studies of cattle consumption of low larkspur (*D. andersonii*) on western U.S. rangelands measured timing and consumption. Cattle consumed low larkspur in relation to availability, and yearlings consumed more low larkspur than did adult cattle. Further, cattle consume low larkspur in a cyclic manner, with periods of high, but non-fatal, consumption followed by several days of detoxification and reduced consumption. This research by ARS scientists at Logan, UT provided new management recommendations that have significantly reduced cattle losses on these rangelands. (Gardner and Pfister, 2007)

**Drug therapy for intoxicated cattle.** ARS Scientists at Logan, UT determined that neostigmine given i.m. at 0.04 mg/kg body weight was effective at reversing the toxic response to larkspur in cattle. Neostigmine is inexpensive, approved for veterinary use for i.m. injection, and has fewer side effects than physostigmine, an alternative treatment. Neostigmine therapy is useful for livestock producers and veterinarians in dealing with persistent larkspur problems on risky rangelands. (Green et al., 2009a, 2009b, and 2009c)

**Sheep grazing as a possible tool to reduce cattle deaths.** Anecdotal accounts indicate that if sheep eat the flowering heads of low larkspurs (e.g., *D. nuttallianum*), the plants senesce quickly, and toxicity and acceptability to cattle decrease. In theory co-grazing would reduce risk to cattle. However, grazing studies conducted over several years and in several states indicate that sheep grazing will not diminish risk for livestock producers with dense low larkspur populations. (Pfister et al., 2010b; Ralphs, 2005)

**Predicting risk of larkspur poisoning.** Duncetop larkspur, *Delphinium occidentale*, occurs throughout the Intermountain West and northwestern United States. *D. occidentale* has two chemotypes, one containing the MSAL-type alkaloids (the more toxic of the larkspur alkaloids) and one lacking the MSAL-type. Both chemotypes appear to occur in definable geographical regions. An initial survey of herbarium specimens (n=>500) representing the distribution of *D. occidentale* clearly indicated there is a geographical region wherein larkspur plants contain little or no MSAL alkaloids. Similarly, a geographical region was identified where the larkspur contains high levels of the MSAL type that are a high risk for cattle. Additionally, reciprocal gardens were established to determine the influences of environment and genetics on larkspur alkaloids in various populations. Toxicity, or lack thereof, of these geographically isolated populations has been confirmed in cattle studies using heart rate and muscle weakness parameters and using toxicological comparison in mice. A more than a 25-fold difference in toxicity was demonstrated between the two plant populations. These important research results are used to define geographical regions where tall larkspurs are of low risk for cattle poisoning allowing livestock producers to utilize these highly productive spring and summer ranges with little risk. (Cook et al., 2009a)

**Low larkspur density related to weather.** Cattle losses to low larkspur are directly related to plant density, as density is a major factor in whether or not cattle can ingest a lethal dose. This multi-state long-term study examined the influence of weather variables on low larkspur density at 4 sites located near NRCS SNOTEL weather stations and a history of substantial cattle losses from low larkspur. Density measurements were taken yearly for 8 years at Yampa and Collbran, CO; Huntington, UT; and Ashton, ID. Years of higher winter and spring moisture, and conversely lower temperatures resulted in greater low larkspur density and greater risk of cattle losses. Understanding the weather conditions that promote greater low larkspur densities has increased the awareness of livestock producers to recognize high-risk years. (Gardner et al., 2008)

### **Livestock Losses From Abortifacient and Teratogenic Plants**

Estimated annual losses associated with abortifacient and teratogenic plants include \$20 million from ponderosa pine in the western U.S and over \$40 million from broom snakeweed due to abortions in west Texas and New Mexico alone. One to three percent of livestock born have some level of birth defect with most related to teratogenic plants. Occasional regional or individual ranch losses have been catastrophic, for example, on the Channel Scablands of Washington State, over \$5 million was lost in a single county in one spring from lupine-induced "crooked calf syndrome". Individual ranch calf losses of 10% to 100% were recorded and similar losses have been reported periodically over the last 50 years.

**Effects of isocupressic acid metabolites in cattle.** Isocupressic acid (ICA) is the primary abortifacient compound in ponderosa pine needles (PN). ARS scientists at Logan, UT determined that Utah juniper (high in agathic acid) was abortifacient in cattle and this supports the hypothesis that metabolites of ICA or other labdanes contribute abortions in cattle. This research helps ranchers to better manage PN abortion in cattle and understand the potential sources of other related plants that induce abortions. (Gardner et al., 2010)

**Concentration of the abortifacient toxin (isocupressic acid; ICA) in pine needles.** ARS scientists at Logan, UT established a sampling protocol for Ponderosa pine (PN) and determined

that ICA concentrations are not uniform between populations, between individual trees, or even between parts within an individual tree. So a composite sample is most representative of a single tree's ICA content. ICA concentration also varies by location and season of the year. This work partially explains the variability of incidence of pine needle abortion in the west and the challenge in making management recommendations to ranchers. (Welch et al., 2009a)

**Characterization of diterpene acids in broom snakeweed.** ARS scientists at Logan, UT characterized diterpene acids in broom snakeweed plants. It is hypothesized that some of these diterpene acids are abortifacient in livestock similar to isocupressic acid from pines. This research will provide information to research scientists linking the abortifacient activity reported from broom snakeweed to that of ponderosa pine. (Davis et al., 2009)

**Factors affecting broom snakeweed population cycles.** Studies on two contrasting sites allowed ARS scientists at Logan, UT to identify two triggers (fire and spring grazing pressure) for snakeweed invasion and develop a new phase for the state-and-transition model for upland big sagebrush ecological sites. Managers should maintain a healthy bunch grass component in the plant community to resist snakeweed invasion. The new snakeweed phase in this transition model provides guidance to land managers and ranchers to maintain healthy rangelands. (Ralphs, 2009; Thacker et al., 2008)

**Herbicide control of broom snakeweed.** ARS scientists at Logan, UT compared herbicide efficacy of Grazon, Milestone and Escort to that of Tordon in controlling broom snakeweed. Tordon was most effective when applied in either spring or fall, while Grazon, Milestone and Escort were acceptable when applied in fall. It was also shown that grass cover and production were greatest in Tordon plots, and were correlated with the degree of snakeweed control. (Thacker et al., 2009a and 2009b)

**Characterizing teratogenic alkaloids from lupines and other species.** Numerous alkaloids have been identified in lupines, poison hemlock, and *Nicotiana* spp. that cause similar birth defects, i.e. cleft palate and skeletal contractures. Many of these alkaloids occur as enantiomers and for this reason need to be fully evaluated including structural characterization, toxicological evaluation and teratogenic potency. Research by ARS scientists at Logan, UT using specialized cell lines (TE-671 cells express muscle-type receptors and SH-SY5Y cells express fetal neuronal receptors) determined the biochemical mechanism of action and allowed the comparison of teratogenic potency of alkaloids and their enantiomers. The utilization of these cell lines has reduced the number of live animals needed for toxicology studies, reduced the cost of this research and increased the output while decreasing turn-around time. (Green et al., 2010; Rader et al., 2008 and 2007) Over the last 5 years ARS scientists have evaluated 4 enantiomeric pairs using a mouse bioassay as well as two cell lines. The enantiomers of a given pair possess different toxicity and level of activity. This information is critical in evaluating overall toxicity of plants and making management recommendations to ranchers, extension agents and veterinarians. (Lee et al., 2008a, 2008b, 2007a, 2007b, 2007c, 2006a, and 2006b)

**Identifying conditions and factors that cause cattle to graze lupines.** On the channel scablands of east central Washington a 5 to 8% calf loss from lupine-induced malformations is common and catastrophic losses of 25 to 100% have been recently recorded. ARS scientists identified *Lupinus leucophyllus* as the species responsible for the losses and surveys of the rangelands in this region identified high risk and low risk pastures. Grazing studies determined when and why cattle graze *L. leucophyllus*. Lupine is nutritious containing 15-18% and up to 40% protein in vegetative parts and seed pods respectively. Lupines are the preferred forage during mid to late summer and this overlaps the susceptible stages of pregnancy. ARS scientists recommended using high-risk lupine pastures for stockers and open heifers and this has resulted in improved utilization of these rangelands and decreased losses to crooked calves. (Gorniak et al., 2008; Lee et al., 2009c and 2008c; Panter et al., 2009 and 2007c; Pfister et al., 2008 and 2007a; Ralphs et al., 2007c)

**Influence of climate on population cycles of lupines in east central Washington.** ARS scientists at Logan, UT determined that lupine populations cycle with climate patterns dying off during dry periods and increasing when precipitation will support seedling establishment. Weather patterns can be used to predict years when risk of crooked calf syndrome is high. (Cook et al., 2009d)

**Understanding the mechanism of lupine-induced crooked calf syndrome.** Using a Spanish goat model, ARS scientists at Logan, UT elucidated the mechanism of action of lupine-induced crooked calf syndrome. Using ultrasound imaging it was shown that fetal movement is inhibited resulting in cleft palate and or skeletal malformations. Studies to determine if intermittent grazing will prevent the malformations were conducted in goats, and fetal movement returns to normal if lupine ingestion is interrupted for 36-48 hours. In addition to the impact on cattle production systems, biomedical researchers use the Spanish goat model as an important tool for development of improved treatments for cleft palate in children. (Hanes et al., 2008 and 2007; Weinzweig, et al., 2008 and 2006)

**Evaluating the role of genotype and environment on alkaloid profiles in lupines.** The *Lupinus* genus is complex and difficult to classify taxonomically. ARS scientists from Logan, UT evaluated specimens of *L. sulphureus* from field collections and herbaria using taxonomic and chemical comparisons. While taxonomy was consistent between specimens and collected plants, alkaloid profiles were not and a total of seven distinct alkaloid profiles were identified. Each alkaloid profile was unique in its geographical distribution and its potential risk to livestock. These results are being used to define geographical regions where lupines grow and to assess risk of crooked calf syndrome. Livestock producers will be able to graze cattle on pastures where lupines contain no teratogenic alkaloids with little risk of losses from lupine, thus providing valuable summer forage. (Gay et al., 2007a and 2007b; Launchbaugh et al., 2007)

**Characterization of *Veratrum* alkaloids in serum of sheep.** *Veratrum californicum* causes multiple birth defects in sheep, the most noted being a cyclops. Cyclopamine is the putative teratogen but other similar steroidal alkaloids occur in many other plant species. ARS scientists at Logan, UT determined absorption and elimination profiles for cyclopamine in sheep. ARS research eliminated the once large losses to the sheep industry, but occasional reports in sheep, cattle, and camelids still occur. Cyclopamine is an important biomedical research tool in elucidation of the Sonic Hedgehog gene pathway, and its chemical derivatives are now being used in phase II clinical trials for cancer chemotherapy in people. (Welch et al., 2009c; Zhang et al., 2008)

### ***Astragalus* and *Oxytropis* Poisoning in Livestock**

There are over 400 species of *Astragalus* and 22 species of *Oxytropis* in North America. All major rangeland communities in North America, South America, China and other countries contain some of these species that negatively impact livestock production. There are 3 syndromes associated with these species: locoism, 3-nitro toxicity, and selenium poisoning.

**Improved extraction method for swainsonine analysis.** ARS scientists at Logan, UT developed a streamlined procedure for swainsonine analysis. A simple one step solvent extraction is more efficient for large numbers of samples for swainsonine screening. No differences were detected between the new procedure and the previously published method. This new method greatly enhances current research output in the area of locoweed/endophyte/swainsonine interactions and current international locoweed plant sample surveys. (Garrosian et al., 2007)

**Evaluation of the endophyte, *Undifilum* in locoweeds using real-time PCR.** The locoweed toxin, swainsonine, is synthesized by a fungal endophyte (*Undifilum oxytropis*). A real-time PCR method was developed to quantify the fungus in plant tissue. The endophyte resides in the

crown of the plant and grows into the new stems, leaves, flowers and is deposited in the seeds. Within all locoweed populations studied, some plants have very low amounts of the endophyte and correspondingly low concentrations of swainsonine. Other plants have high amounts of endophyte and corresponding high concentrations of swainsonine. This suggests that the endophyte must grow and colonize the plant at some critical threshold before swainsonine is produced at toxic levels. This basic research will advance future research in understand the biology of the fungus, its growth and transmission, and its synthesis of swainsonine so that improved management practices can be developed. (Cook et al., 2009b and 2009c; Ping et al., 2009; Ralphs et al., 2007b)

**Conditions under which livestock graze locoweeds.** Late fall, winter and early spring are critical times when actively growing locoweed are preferred to dormant grasses. Cool season grasses produced significant competition against new locoweed seedlings. Germination of locoweed seed was lower in grass treatments than in the bare ground and control plots. Pubescent wheatgrass reduced the vigor and increased mortality of locoweed seedlings more than other seeded grasses, however all grasses were shown to suppress reestablishment of indigenous locoweed plants. Using this research, adjustments to current management strategies have reduced losses from locoweeds in New Mexico and Colorado and improved utilization of these rangelands. (Pfister et al., 2010a and 2007b)

**Animal species differences in susceptibility to locoweed toxicity.** Horses are uniquely sensitive to locoweed poisoning and cattle and sheep are intermediate in their susceptibility. Rodents and mule deer are relatively resistant and develop few neurologic lesions when compared to other species. This research is important to livestock producers and wildlife managers to prevent or reduce losses from locoweeds. (Stegelmeier et al., 2007d)

**Effects of locoweeds on reproductive function in livestock.** ARS scientists at Logan, UT have evaluated locoweed-induced reproductive dysfunction in multiple farm species. Reproductive efficiency in all species is negatively impacted and severity of reproductive effects and prognosis for recovery are species, time and dose dependant. . (Furlan et al., 2007; Hafez et al., 2007; Pfister et al., 2005)

**Ipomoea fetotoxicity.** *Ipomoea* species are widely distributed throughout tropical and sub-tropical areas worldwide. Many of these species contain the locoweed toxin, swainsonine and related calystegines. Research done at the University of Sao Paulo, Brazil, in collaboration with ARS scientists in Logan, UT determined that *Ipomoea carnea* cause embryonic death, inhibits neonatal/maternal interactions and reduces overall reproductive performance. This research has improved livestock production in countries where toxic *Ipomoea* species grow. (Barbosa et al., 2007 and 2006; Hueza et al., 2007)

**Development of new diagnostic tests for locoweed intoxication.** ELISA technology and proteomic analysis was used to investigate potential diagnostic tests for locoweed poisoning. Linkers were synthesized and reacted with swainsonine to form swainsonine haptens, however products of reactions were not clean and satisfactory swainsonine haptens have not been isolated. Differences in blood proteins in poisoned cattle, sheep and horses have been detected, but a reliable marker for the abnormal glycosylation of these proteins has not yet been identified. This research will provide advanced technology to better diagnose locoweed poisoning and to predict prognosis for recovery. (Landau et al., 2009; Soto-Blanco et al., 2007)

**Poisoning of livestock by *Astragalus* species containing nitro-propionic acid.** The nitrotoxins from certain *Astragalus* species were neurotoxic to horses and resulted in enlarged and abnormal neurons of the basal ganglia. This research has improved diagnosis of plant poisoning in animals, particularly nitro-containing *Astragalus* in horses. This research provides awareness to veterinarians, extension agents, and horse owners of the potential toxicity of these plants and improves the diagnosis of plant poisoning. (Cook et al., 2009e)

**Poisoning of livestock from seleniferous forage.** Many selenium accumulator plants store high concentrations of selenium as selenate and methylselenocysteine. These forms of selenium are toxic to livestock when ingested and depending on the rate of ingestion will induce chronic or acute toxicoses. Scientists at Logan, UT determined rates of absorption, distribution, and elimination of multiple forms of selenium including selenite, selenate, selenomethionine, methylselenocysteine and selenium from *Astragalus* species and Western Aster. This research provides new and important information for diagnostic labs throughout the U.S. on toxico-kinetics and pathology of different forms of selenium. Additionally, this research provides information to predict clearance of these forms of selenium from tissues for food safety issues. (Tiwarly et al., 2007 and 2005)

### **Toxicity of Pyrrolizidine Alkaloid-containing Plants and Other Hepatotoxic & Neurotoxic Plants**

Plants containing pyrrolizidine-alkaloids (PAs) are globally distributed and are the greatest worldwide poisonous plant threat, costing hundreds of millions of dollars annually in poisoned animals, decreased livestock production, inefficient use of resources, lost trade and human poisonings. PA's and their metabolites contaminate feeds, foods and herbal products and some countries have regulated exposure limits, creating trade barriers for imported grains and food products. PA associated human health issues (liver disease, fetal toxicity, carcinogenesis, etc.) are significant as well as the many animal production issues created by these plants. Other hepatotoxic and neurotoxic plants, such as white snakeroot in the Midwest, rayless goldenrod in the Southwest, water hemlock with worldwide distribution, and photosensitizing plants also contribute to the overall losses of poisonous plants on a worldwide basis.

**Improved methods for monitoring and characterizing pyrrolizidine alkaloids.** Plants containing toxic pyrrolizidine alkaloids (PAs) are found throughout the world and commonly poison livestock, wildlife and humans. The toxicity of many PA containing plants including *Senecio*, *Crotalaria*, *Cynoglossum*, *Ageratum* and *Amsinkia* species has been described. Improved methods have been developed for plant extraction, alkaloid isolation using solid phase extraction and detection and quantification of alkaloids using liquid chromatography/mass spectrometry. Class specific and alkaloid specific antibodies and ELISA's have also been developed to screen large numbers of feed and food samples. These methods will improve the detection of PAs in herbal products, feed and food to reduce the risk of poisoning in animals and humans. (Stegelmeier et al., 2009a)

**Characterization of PAs of specific *Symphytum*, *Senecio* and *Ageratum* spp.** ARS scientists developed chemical and immunologic techniques to better analyze and characterize the pyrrolizidine alkaloid (PA) contents of two main comfrey species (*Symphytum* spp). These comfrey plants are commonly used as herbal products and have been associated with PA poisoning in people. ARS scientists at Logan, UT characterized the PAs in an invasive plant commonly called "fireweed" (*Senecio madagascariensis* Poir.) that has heavily infested rangelands on the Hawaiian Islands. Fireweed has become a serious weed problem and heightened the concern for possible poisoning of grazing livestock in the area. The PAs, clinical disease and histologic lesions produced in cattle poisoned by *Ageratum houstonianum* have also been characterized and described. This is a toxic Central American plant and recently several similar *Ageratum* species have been associated with human and livestock poisoning in Africa, Nepal and Afghanistan. (Provenza et al., 2009; Stegelmeier et al., 2009a; Gardner et al., 2006)

**The carcinogenicity of pyrrolizidine alkaloids (PAs).** Pyrrolizidine alkaloids are found in many plant genera and in hundreds of species throughout the world and have been responsible for many outbreaks of poisoning in animals and people. Many PAs are carcinogenic; pilot studies have been completed by ARS scientists in Logan, UT using P53 knockout mice to study the

potential carcinogenesis of PAs. Information from these small animal models will be used to better characterize and understand the toxicity and potential carcinogenicity of PAs in animals and people. (Stegelmeier et al., 2007)

**Chemical analysis and toxicological evaluation of rayless goldenrod and white snakeroot.**

White snakeroot (*Ageratina altissima*) and rayless goldenrod (*Isocoma pluriflora*) cause “trembles” and “milk sickness” in livestock and humans. The suspected toxin complex in both species is called tremetol and is a mixture of sterols and derivatives of methyl benzofuran ketones. A quantitative HPLC method was developed to measure these compounds in plants. Three major benzofuran ketones, tremetone, dehydrotremetone, and 3-oxangeloyl-tremetone were isolated from rayless goldenrod, 11 different tremetone-like compounds were isolated from white snakeroot and quantified. Dose response studies in horses and goats using electromyography and electrocardiography were conducted. Poisoning produces extensive myonecrosis of many skeletal and cardiac muscles. This research provides new results and current information about the toxicity of rayless goldenrod and white snakeroot in livestock for veterinarians and ranchers to improve diagnosis of plant poisoning and avoid animal losses. This information is essential for livestock producers and veterinarians to identify and avoid exposure to toxic populations of these plants. With increasing emphasis in society on natural foods, farmers markets, hobby farms, etc., the potential for human exposure to these compounds is increasing. (Stegelmeier et al., 2010)

**Toxicity of *Adonis*, cutleaf nightshade and water hemlock seed to livestock.** *Adonis aestivalis* (pheasant’s eye) contains cardio toxins and can contaminate hay resulting in livestock poisoning. Histologic and ultrastructure studies demonstrated the classical cardioglycoside-associated myocardial lesions. Cutleaf nightshade (*Solanum triflorum*) poisons horses and contaminates feed and hay. A rodent model was used to verify toxicity, describe clinical effects and define pathology. Ingestion of immature water hemlock (*Cicuta maculata*) seed caused a 20% death loss in a herd of cattle in central Utah. Chemical analysis and research using a rodent model determined that water hemlock seed contained the highly toxic cicutoxin and multiple cicutol-like compounds typically found in toxic hemlock tubers. This information has been disseminated to producers, veterinarians and extension agents alerting them of the potential toxicity of these plants. (Panter et al., 2007; Stegelmeier et al., 2007b and 2007c)

**Analysis of saponins in switchgrass cultivars.** ARS scientists in Logan, UT developed a rapid analytical method to isolate, characterize and quantify steroidal saponins in switchgrass. Three major steroidal saponins were quantified in several cultivars of switchgrass (*Panicum virgatum*). Steroidal saponins were also quantified in *Panicum maximum* and *Brachiaria* spp. that are important forage species in South America and Africa. Steroidal saponins can be toxic to livestock causing hepatogenous photosensitization. The U.S. Department of Energy has identified switchgrass for development into bio-fuel that could reduce the nation’s dependence on foreign oil. Estimates suggest switchgrass could be grown on over 40 million acres in the U.S. Switchgrass grown on this scale will result in residues or hay used for animal feed, or pastureland used livestock grazing. *Brachiaria* is planted on 50-70 million hectares in Brazil and is used for livestock grazing. This research and the analytical methodology is used to screen switchgrass cultivars for saponin profiles and concentrations and grass breeders use this information to select cultivars with low saponin content for further development for bio-fuel production. Collaborative research with scientists in Brazil is improving identification of toxic grasses, reducing livestock losses and improving utilization of important rangelands where these grasses grow. (Cerqueira et al., 2009; De Almeida et al., 2009; Jani et al., 2009; Lee et al., 2009b; Stegelmeier et al., 2007a and 2009c; Faeth et al., 2006)

**Development of phyloerythrin methodologies.** Plants containing photodynamic chromophores or toxins that alter liver function result in dermal accumulation of photodynamic plant metabolites. These molecules react with sunlight producing photosensitization and sunburn in light pigmented areas of the body or areas not covered by hair such as the nose, tip of the

ears, udder or vulva. Phylloerythrin assays using fluorometric spectrometry were further developed. Veterinarians, researchers, and diagnostic labs will use these assays to determine the extent of liver damage and to diagnose poisoning by plants that cause photosensitivity. (Liang et al., 2006)

**Objective B.4:** Assess near- and long- term animal productivity, well-being and product quality under alternative rangeland management strategies.

Livestock animal performance can be influenced by specific management practices. One key factor is stocking rate, and evaluation of these kinds of influences requires long-term studies. ARS is in a unique position to conduct these long-term studies because of its history and long tenure at specific locations with an adequate land base for these types of studies.

**Grazing strategies and forage/animal production in semi-arid rangelands.** The preponderance of grazing research documents indicates weather variation and stocking rate affect vegetation and animal responses independently of grazing system in semi-arid landscapes. Forage production is reduced with increasing stocking rate, as are individual gains whereas livestock gains per unit land area are increased with increasing stocking rate. Experimental evidence does not support implementation of rotational grazing to enhance production goals on western rangelands. However, this evidence does not effectively address all potential management benefits arising from rotational grazing systems because these systems have seldom been investigated as a component of an entire ranch enterprise nor has this type of research investigated social/human factors of grazing management systems. Rangeland managers and policy makers need to recognize that the potential benefits of grazing systems are derived from sound management models, rather than from ecological phenomena. (Smart et al., 2010; Hart and Derner, 2008; Briske et al., 2008b; Derner et al., 2008; Derner and Hart, 2007a and 2007b)

**Using livestock as ecosystem engineers in rangelands.** Management of rangelands has emphasized livestock production with the uniform use of vegetation, but emerging ecosystem objectives for these lands may not benefit from these prior management practices. Research by ARS scientists at Cheyenne, WY is providing land managers with important tools to achieve desired contemporary conservation objectives. For example, livestock can be used as ecosystem engineers (tools) to alter structure and function of rangelands can improve habitat for declining native grassland birds in the western Great Plains. The use of livestock as ecosystem engineers to alter vegetation structure and function for additional ecosystem services beyond food and fiber is feasible for application by private and public land managers. Adapting these management activities within the context of current livestock operations will require some management modifications. (Toombs et al., 2010; Augustine et al., 2010; Derner et al., 2009a and 2009b; Derner and Whitman, 2009; Derner et al., 2006a and 2006b)

**Rangeland Biodiversity: management practices to enhance rangeland wildlife habitat while sustaining livestock production.** In recent years, the impact of fire and prairie dogs on wildlife populations has not been clear. ARS scientists at Cheyenne, WY implemented prescribed burning programs on five National Grasslands encompassing >1 million acres of public rangelands in the western Great Plains to enhance wildlife habitat and control unpalatable plant species. Comparison of yearling steer weight gains on pastures with and without prairie dogs demonstrated a linear decline in gains with increasing percentage of the pasture colonized by prairie dogs. At the same time, our studies showed that prairie dog colonies are a preferred habitat for the mountain plover (supporting densities similar to prescribed burns). These findings indicate that available plover nesting habitat associated with prairie dog colonies closely tracks the area actively occupied by prairie dogs. Collectively, these studies have quantified tradeoffs between management for livestock versus some declining wildlife species, and provide insights to landscape-scale management patterns that can be used in managing rangelands for multiple

ecosystem services. (Augustine, 2011; Toombs et al., 2010; Augustine et al., 2010, 2008a, 2008b and 2007; Augustine and Milchunas, 2009; Derner et al., 2006a)

**Altering calving date impacts calf weaning weight.** Calving date and weight affects subsequent management of heifer calves for their entry into the breeding herd. ARS scientists at Miles City, MT evaluated the impact of differing nutrient patterns from birth until first breeding on weight gain and reproductive performance of heifers born into mid-winter, early spring, or late spring calving systems and weaned at different ages. Heifers from varied calving, pre- and post-weaning management strategies performed similarly in initial reproductive performance and subsequent calf production, even though growth patterns and body weight at breeding differed. There are many options available to producers for rearing heifers by adjusting calving dates to meet cow nutritional needs. (Grings et al., 2006)

**Evaluating different breeds of cattle for arid lands.** Increasing costs for fuel, grains, and supplemental feeds are requiring livestock producers to consider new strategies to maintain profitability. An ARS scientist at Las Cruces, NM has been evaluating Criollo cattle that evolved in arid environments as an alternative to traditional breeds. Criollo cattle traveled farther to use a greater diversity of habitat types, and spent less time near water/riparian areas compared to British breeds. Criollo cattle also mature earlier with a greater probability of reaching puberty at a younger age. These traits suggest that Criollo cattle may be a heritage breed well suited for beef production systems in arid lands and merit further study.

## **Problem Statement C**

Need for improved rangeland restoration, rehabilitation and mitigation practices, germplasm, tools and strategies to restore rangeland integrity in a manner that is economically feasible and environmentally acceptable.

**Objective C.1.** Understand mechanisms of weed invasion and develop management strategies that can be used to restore rangelands that have been degraded by weeds and other disturbances.

### **Ecology and Control of Undesirable Invasive Plants**

Weeds pose a major threat to rangelands throughout the world. Combating invasive plants in rangelands requires understanding the causes of invasion so improved management strategies can be developed.

**Understanding what makes a plant invasive on rangelands.** The introduction of non-native species can result in the breeding among closely related native species or populations that have historically occurred in different areas. This genetic mixture can result in the formation of new genetic forms that are more invasive than either one of their parental species or populations. ARS scientists in Reno, NV have found that multiple introductions and subsequent gene flow provide a serious mode of the evolution of new invasive genotypes. Researchers developed a two molecular marker system, random amplified polymorphic DNA (RAPD) and microsatellites to characterize the genetic diversity and population structure of cheatgrass from four northern Nevada sites. Both techniques proved useful in assessing population genetics. RAPDs proved most useful in revealing genetic diversity. Microsatellite markers were successful in identifying heterozygous individuals, products of cross-pollination, which is an important aspect regarding the production of offspring that may be able to colonize new habitats. This process can present a serious problem for control efforts due to the novel genetic variation found in these new evolved populations. (Ashley and Longland, 2007)

**Mechanistic links between resource availability and plant invasion.** Combating invasive plants in rangelands requires understanding and addressing the causes of invasion. A primary cause of plant invasion is thought to be increased availability of resources, such as water,

nitrogen and light. ARS scientists at Burns, OR have expanded upon this basic idea by linking it to the enemy release hypothesis. This hypothesis states that plants that escape from resource-rich habitats are likely to benefit most, due to lower predation from their natural enemies. Consequently, anthropogenic disturbances that increase plant resource availability may play a larger role in invasion than previously thought due to exotic species having a greater ability to capitalize on resources because they are not as limited by natural enemies as native species. In a related study, ARS scientists found that invasive species are also evolving traits that allow them to take advantage of high resource availability. Utilizing genetic material from the USDA-ARS National Plant Germplasm System, ARS scientists found that among 14 invasive plants that are invading North America, there was a general trend towards evolution of higher growth rates. Finally, ARS has tested whether resource availability can be manipulated to control invasion. ARS scientists demonstrated that reducing nitrogen availability can limit or even preclude invasion by two problematic invaders: diffuse knapweed and Dalmatian toadflax. The scientists then reviewed management strategies for manipulating nitrogen availability, and concluded that strategies that encourage slow-growing native plants, such as restoration or prescribed burning, are most effective at immobilizing nitrogen, and combating invasion. (Perry et al., 2010; Blumenthal et al., 2010 and 2009; Blumenthal, 2009, 2006 and 2005)

**Nutrient management in restoring degraded rangelands.** A major problem facing land managers is choosing the appropriate species/mixtures to seed into disturbed rangelands that will restore ecosystem function. There is evidence that invasive weeds, e.g. cheatgrass and dyer's woad, out-compete the native plants for nutrients, particularly nitrogen. ARS scientists at Logan, UT are studying the interaction between rangeland plants and invasive weeds. This research is focusing on degraded rangelands in the Snake River Plain and Great Basin by identifying mechanisms of weed dominance, and enhancing the knowledge base of soil nutrient dynamics (Norton et al., 2003, 2004, and 2007), with emphases on how weeds differentially utilize mineral forms of soil N (Monaco et al., 2005a, 2003a, and 2003b; MacKown et al., 2009). In addition, this research provides land managers with ecologically based interpretations of how weeds operate on western US rangelands, and potential options for manipulating soil N to assist in weed reduction (Perry et al., 2010; James et al., 2010). Comparisons of a broad array of perennial grasses, forbs, and shrubs illustrate tremendous variability and suitability for revegetating disturbed, weed-infested ecosystems (Monaco et al., 2005b; Waldron et al., 2005; Ralphs et al., 2007b; Leonard et al., 2008; Thacker et al., 2009a and 2009b; Gunnell et al., 2010). These team efforts with rangeland scientists, ecologists, and plant geneticists in Logan, UT have also yielded new innovative ways that plant material specialists, and land managers can readily choose the correct plant materials for the first line of defense against the spread and dominance of invasive plants (Jones and Monaco, 2007 and 2009; Jones et al., 2010a). Integrated invasive plant management provides land managers with multiple tools working in unison to improve rangeland resources (Monaco et al., 2003c; Monaco and Creech, 2004; Monaco et al., 2005c; Morris et al., 2009; Vasquez et al., 2010).

**Escape from soil-borne pathogens aids plant invasion.** Enhanced performance of invasive plants has been explained by the release from the harmful effects of natural enemies typical in their native ranges. However, this has rarely been validated for soil-borne pathogens. Soil samples from numerous populations of black cherry in its native and non-native ranges were collected by ARS scientists in Miles City, MT and used to develop a bank of soil-borne pathogen (*Pythium* spp.) isolates. Comparative pathogenicity experiments tested the effect of isolates which were identified by DNA sequencing. Results from the pathogenicity trials revealed the presence of highly harmful pathogens among *P. serotina* populations in its native range but a lack of similar diseases for populations in its non-native range. This indicates the increased dominance by black cherry in its non-native range is due in part to the absence of effects by harmful pathogens compared to populations in its native range. This study provides some of the first empirical evidence to suggest that biogeographical variation in plant performance is explained at least in part by non-native populations escaping from the effects of virulent soil-borne pathogens. (Reinhart et al., 2010; Reinhart and Clay, 2009)

**Introduced plants may help reduce cheatgrass in the Great Basin.** ARS scientists in Reno, NV documented that crested wheatgrass is among a select few perennial grasses that can effectively suppress cheatgrass. The team tested the hypothesis that crested wheatgrass reduces available mineral soil N to levels below which cheatgrass is competitive. Relative to other ecosystems tested, soil beneath crested wheatgrass does not have appreciably lower mineral N. In fact, the molar proportion of ammonium-N in the total mineral pool is relatively high in the tested crested wheatgrass sites, averaging 84% throughout the year. The team is now exploring if crested wheatgrass reduces nitrification and thereby decreases the competitive stature of cheatgrass. (Blank and Young, 2009)

**Effects of the Northern Tamarisk beetle as a biological control agent on ecosystem processes and weed physiology.** The invasion of western riparian areas by the exotic species, *Tamarix*, is an issue of serious concern due to alterations in biodiversity, water use, channel processes and habitat quality. Due to the serious and widespread nature of the problem and lack of viable control strategies, APHIS approved in several western states the release of various species of leaf beetle (*Diorhabda*). In order to understand the long-term effects of this biological control effort, ARS scientists in Reno, NV conducted greenhouse experiments to understand the effects of the beetle on *Tamarix* physiology, particularly the proximate cause of early leaf fall that results from beetle herbivory, and a yearlong field study to understand the effects on early leaf fall on litter quality and decomposition rates. Results demonstrated that early leaf fall from herbivory renders saltcedar unable to control water loss during the day and night, and results in water stress. The field study demonstrated that early leaf fall produces litter of higher quality because plants were unable to retranslocate both nitrogen and phosphorus and the litter decomposed more rapidly. Physiological responses to herbivory were shown to have potential impacts on carbon, water and nutrient cycling and improved understanding of the restoration potential of the highly valued riparian system. (Snyder et al., 2010; Uselman et al., 2010)

**Prescribed burning reduces emergence of invasive weeds.** In the Northern Great Plains, common weed control methods rarely prevent weed seeds already on the ground from reestablishing. ARS scientists at Miles City, MT evaluated fire management to reduce seed viability. Seeds of Japanese brome, spotted knapweed, Russian knapweed, and leafy spurge were deposited on the soil surface, subjected to fire at six fuel loads common to grasslands, and results compared to a non-burned control. Seedling emergence was reduced 79-88%; with fuel loads common to the north-central Great Plains, emergence was reduced by at least 97%; with fuel loads typical of the northeastern Great Plains, emergence probabilities were less than 1% for all species except spotted knapweed. There is high potential for using fire to disrupt the life cycle of invasive species across the northern Great Plains. (Vermeire and Rinella, 2009)

**Growth regulator herbicides prevent invasive annual grass seed production.** Growth regulator herbicides are commonly used to control broadleaf weeds in rangelands and croplands. The herbicides often reduce cereal seed production if applied to cereals while the grasses are developing reproductive parts. ARS scientists at Miles City, MT tested whether 2,4-D, dicamba, and picloram at typical field use rates would reduce Japanese brome seed production. Picloram reduced seed production nearly 100% when applied at the internode elongation, boot, or heading stages of growth, whereas dicamba appeared to be slightly less effective and 2,4-D was much less effective. Results were corroborated in the field, reducing Japanese brome seed production by more than 95% by applying aminopyralid or picloram at three different plant growth stages. Our results indicate it may be possible to control Japanese brome by using growth regulator herbicides to reduce its seed production, thereby depleting its short-lived seed bank. (Rinella et al., 2010a and 2010b)

**Economical method to reduce cheatgrass and produce new agricultural products.** Between 2000 and 2009, 16.2 million acres—an area about the size of West Virginia—burned in the Great Basin in part because of invasive annual weeds. ARS scientists in Reno, NV

conducted research to evaluate the impact of mechanically removing the invasive annual cheatgrass seed source as a technique to reduce its seedbank, reduce wildfire frequency, and restore degraded rangelands. The possibility of using cheatgrass seeds collected as part of the restoration efforts were evaluated as a new grain for use in producing food and beverages as a means to offset the cost of revegetating Great Basin rangelands. The research successfully demonstrated that cheatgrass seeds are a viable source of grain for use in agricultural products and that commercial vendors would purchase the seed if available. No further work is planned at this time as it has been successfully demonstrated that cheatgrass seeds are a viable protein source in producing agricultural food products. (Clements et al., 2009)

**Selection of weed control methods is facilitated by cost-benefit calculation tool.** Selection of weed control methods is complicated by the inability to determine weed impacts on forage production. ARS scientists at Miles City, MT used data from field experiments throughout the range of leafy spurge and spotted knapweed to develop models that predict weed and forage responses to management actions. An internet-available model (<http://199.133.173.229/WeedImpact/>) was developed that uses weed density to calculate impacts on desired species, forage production and livestock carrying capacity.

**Quantifying invasive weeds.** Spotted knapweed rapidly invades and destroys native rangelands. Scientists at the U.S. Sheep Experiment Station have established that aerial digital imagery is an effective tool for monitoring the frequency and density of spotted knapweed plants in various sagebrush steppe terrains. This research has resulted in monitoring tools that are better suited for landscape-scale monitoring of extensive rangelands. A manuscript describing this research was submitted to a peer-reviewed scientific journal. (Firincioglu et al., 2009; Taylor et al., 2009; Mitchell et al., 2007; Seefeldt et al., 2007; Taylor et al., 2005)

**An Assessment of Saltcedar and Russian Olive Control Science.** A comprehensive assessment of the state of the science on saltcedar and Russian olive control was produced from a multi-agency meeting in collaboration with ARS scientists in Reno, NV. The report contains a comprehensive review on the extent of the problem, effects on water cycling and wildlife, the efficacy of control strategies, and subsequent restoration efforts that is useful for managers and policy makers. Information contained within this document was used to provide congress with an assessment of *The Saltcedar and Russian Olive Demonstration Act of 2006* (Public Law 109-320), and provided applicants with a common background for prospective demonstration projects. The produced document was peer-reviewed and published by the U.S. Geological Survey. (Nagler et al., 2010)

### **Improving Rangeland Reseeding Success**

Millions of acres of rangelands are incapable of self-repair and are in need of restoration to reinstate ecosystem function and services. Reseeding is an important tool that managers use to meet this objective with tens of millions of dollars spent each year in applying seed to disturbed sites; however, most efforts fail to successfully establish vegetation. Consequently, it is imperative that we understand why reseeded efforts are failing and use this information to develop new techniques to improve reseeded success.

**Seeding following fire or other disturbance is prone to failure.** ARS scientists at Burns, OR examined the demographic processes causing seedlings to fail over three years and in four large fire sites seeded by the BLM. A major recruitment bottleneck (i.e. loss of seedlings) was found across years and sites at the transition between seed germination and emergence. For example, germination across species, years and sites was high, averaging 72%; however, up to 85% of the seeds that germinated failed to emerge. Of the seeds that did germinate from the four fires sown, 98% failed to emerge. This research is the first to identify a reoccurring bottleneck to seedling recruitment. (Article submitted to *Ecological Applications* Sept. 2010)

**Seeding rangelands following fire often fails.** ARS scientists at Burns, OR evaluated the degree to which water availability, invasive plant abundance and seeding technology influenced post-fire seedling establishment. Results indicate that planting depth was the single most important factor limiting plant establishment. This research suggests that moderate improvements in seeding technology may yield large benefits to post-fire reseeding success. (James and Svejcar, 2010)

**Forecasting Grass Seedling Germination based on Soil Temperature and Moisture Conditions.** Semi-arid rangeland systems exhibit high variability in seedbed temperature and moisture during fall, winter and spring establishment periods making it difficult to predict potential performance of alternative plant materials in the field. ARS scientists in Boise, ID developed a new geo-statistical model for evaluating population-level seedlot performance under an unlimited variety of field-variable temperature regimes. This model was demonstrated to be more accurate than other available models in predicting seedlot performance. Models of this type can be used in conjunction with historical data, weather forecasting and seedbed models, and climate change predictions to evaluate potential seedlot performance under historical, current and potential future climate and microclimatic field scenarios. These models are currently being used to evaluate BLM fire-rehabilitation and restoration practices to optimize resource expenditures by identifying seeding-treatment options that have the highest probability of success. (Roundy et al., 2007; Hardegee, 2006a and 2006b; Hardegee and Winstral, 2006)

**Managing nitrogen availability to control invasive annual grasses is questionable.** Managing soils for low N availability has been suggested as a strategy to reduce competition from invasive annual grasses and improve establishment of desirable plants. As a result, some rangeland restoration projects have included expensive treatments to reduce soil N. Growth rate, biomass allocation and physiology of annual and perennial grasses were measured, by ARS scientists at Burns, OR, under a wide range of soil nitrogen levels. No evidence was found to suggest that low soil nitrogen levels favored the growth of perennial over annual species. These results suggest the cost of nitrogen reduction efforts is not warranted in post-fire strategies. (James et al., 2010)

**Managing plant density for optimum forage production.** In restoring degraded cropland and pastures in the southern Great Plains, land managers need information on the optimum density of plants to seek during seeding operations. At Woodward, OK, ARS researchers evaluated the effects of plant density on two native bluestem grasses (*Andropogon* species). They found that the optimum plant density for forage production was between 6.0 and 8.0 plants per square meter. However, the optimum density for plant crude protein was at low density (1.2) and at a much higher density (10.8). They also found that the greatest leaf area occurred at 10.8 plants per square meters. Therefore the most palatable and highest quality forage could be obtained with only a small decrease in total dry matter when there averaged 10.8 plants per square meter. This is equivalent to 43,560 plants per acre. Assuming good germination, this density can be achieved by planting approximately 4 lbs of pure live seed per acre for sand bluestem and slightly less for big bluestem because its smaller seed size. (Springer et al., 2007)

**Selecting Plant Species for Post-Fire Rehabilitation and Rangeland Restoration Treatments.** Species used for rangeland restoration must compete with aggressive introduced annual weeds that establish and grow in the late winter and early spring. ARS scientists in Boise, ID conducted a broad comparison of relative field-germination and establishment response of cheatgrass, a principal invasive annual weed, and a large number of native perennial bunchgrass species including bluebunch wheatgrass, Sandberg bluegrass, big squirreltail, bottlebrush squirreltail, Basin wildrye, Idaho fescue, and thickspike wheatgrass. They found that cheatgrass germination in the field is approximately twice as fast as native perennial species, but that this effect is probably not as important as relative seeding rates given the weed produces up to 10 times the number of seeds typically planted in a rangeland seeding. A germination-response index developed from these data is being used to evaluate long-term

probabilities of favorable weather for plant establishment, and to evaluate potential climate change effects on native and non-native/weed establishment. Regional differences in species used for rangeland restoration may be an important factor in plant-materials selection for rangeland rehabilitation projects. (Hardegreer et al., 2010 and 2008a)

### **Managing Kentucky bluegrass and smooth brome on the northern Great Plains.**

Kentucky bluegrass and smooth brome are common invasive species on the cool-season grasslands of the northern Great Plains. ARS scientists in Mandan, ND and North Dakota State University evaluated different management strategies including combinations of burning, herbicides and defoliation on rangelands near Mandan, ND and on the Cheyenne National Grasslands near Lisbon, ND. The initial research at Mandan suggested that plant community and target species impacted the effectiveness of restoration strategies. The success of burning followed by an application of glyphosate in warm-season grass communities at Mandan resulted in a US Forest Service funded project to evaluate time of burning and glyphosate applications on the tall-grass prairie near Lisbon, ND. This research suggests that a fall burn followed by a spring application of glyphosate could rapidly increase the amount of warm-season native grasses in the species composition. (Hendrickson and Lund, 2010; Lund et al., 2007)

**Small mammals do heavy lifting in restoring Great Basin rangelands.** Approximately 26 million acres of the Great Basin Sagebrush ecosystem, nearly 40 percent, is seriously degraded and has minimal capacity to produce forage for wildlife and domestic livestock. ARS scientists in Reno, NV have found that desert rodents can have profound impacts on specific plant species and on species composition of arid plant communities. It was demonstrated that superficially buried, scattered seed caches made by rodents accounted for 95% of seedling production of the native Indian ricegrass, which is a critical forage resource for livestock and wildlife. It is possible to enhance the beneficial effects of a seed-caching rodent species of native plant species by providing inexpensive commercially available, “diversionary” seeds that are highly desirable to the rodents. Rodents would then cache both seed types but preferentially recover the preferred diversionary seeds before beginning to consume the less desirable native plant species seeds. This approach can be a cost-effective means of revegetating rangelands, as more native plant seeds are available for emergence as seedlings using this passive and inexpensive restoration scheme. (Murray et al., 2006)

**Reducing the cost in restoring Great Basin Rangelands.** Over 2 billion dollars has been spent fighting wildfires in the western United States and millions more in trying to restore these devastated landscapes over the last decade. ARS scientists in Reno, NV have been working on cost-effective methods for revegetating these landscapes and reducing the frequency of burning. The scientific team has developed techniques that increase the success of restoration and/or revegetation practices on burned sagebrush plant communities and cheatgrass invaded rangelands. These techniques on average cost \$119.75 per acre during the first year. However, if treatments are delayed until the second year costs goes up to \$207.95 per acre and success is significantly less. Federal and state agencies are now using these techniques in the battle to restore native rangelands in the western portion of the Great Basin. (Clements et al., 2009)

**Objective C.2:** Develop and evaluate plant materials for rangeland repair, revegetation, and restoration.

### **Molecular Research of Desirable Plant Traits**

With increased emphasis on establishing a biologically diverse ecosystem of native and introduced plant materials, there is a need for improved grasses, legumes, and forbs that can compete with invasive weeds (BLM, 1999; 2000). This section documents how ARS scientists are using plant genetic diversity and gene discovery research to identify, select, and develop improved plant materials for sustainable stewardship of rangelands, pastures, and other growing environments in the Western U.S.

**Genetic mechanisms behind complex traits.** The understanding and utility of genetic mechanisms behind complex traits (forage yield, poor forage quality, salinity tolerance, winter hardiness, seed shattering, and rhizomatous growth habit etc.) in perennial grasses enhances our ability to identify markers and improves the efficiency with which we can develop better plant materials. Comparative mapping has demonstrated conservation of quantitative trait loci (QTL) and genes across divergent grass species and facilitated identification of genes controlling these traits. (Cogan et al., 2007; Bellgard et al., 2004; La Rota and Sorrels, 2004)

**Development of expressed gene libraries of perennial forage grasses.** A fundamental requirement for genetic improvement of crop plants is DNA sequence information. A collection of sequenced genes of an organism, called an expressed sequence tag (EST) library, provides a source of DNA markers for fingerprinting and genetic mapping, phylogenetic studies, and often a blueprint of genes found to be expressed under different environments. ARS scientists in Logan, UT have developed EST libraries for *Leymus cinereus* (Basin wildrye), *Leymus triticoides* (beardless wildrye), *Elymus wawawiensis* (Snake River wheatgrass), *Elymus lanceolatus* (thickspike wheatgrass), *Pseudoroegneria spicata* (bluebunch wheatgrass), and *Dactylis glomerata* (orchardgrass). The first five species are dryland Triticeae, are found throughout the western U.S.A., and are the major grazing native grasses in dryland areas. Orchardgrass seed sales are over \$14 million USD each year in the U.S.A (Oregon Agriculture Information Network). These libraries will help in the development and improvement of these grasses, and have been used for genetic mapping, elucidation of chromosome regions controlling rhizome development, and studies of diversity among germplasm, and comparative genomics. Several international researchers have requested sequences or data concerning the EST libraries, and functional genetic research on these organisms has accelerated. (Larson et al., 2009; Bushman et al., 2008; Kaur et al., 2008a and 2008b)

**Development of molecular markers for differentiating genomes, species, chromosomes and cultivars in Triticeae grasses.** Cheap and easy-to-use molecular markers are needed to identify various genomes, species, and chromosomes in Triticeae for studies of taxonomy, genomics, genetics, and breeding. Scientists at Logan, UT developed RAPD-derived CAPS markers to differentiate two closely related E genomes from other genomes of perennial Triticeae including R, the genome of rye. A Y-genome STS marker was developed to reliably detect its presence in any *Elymus* species containing this unique genome. They also identified a battery of EST-SSR markers for each individual E-genome chromosomes, many of which carry desirable traits for plant improvement. This technology has been adopted by the industry to differentiate seed lots containing potentially noxious weeds from wheatgrass seed lots that would otherwise have been released as non-certifiable by the seed certification agencies. (Wang et al., 2010; Okito et al., 2009; Li et al., 2007)

**Mapping of genes and molecular markers in *Leymus* and other grasses.** Molecular research of perennial grasses is lagging behind work on annual crops. Scientists at Logan, UT mapped molecular markers and genes for 6-SFT involved in fructan biosynthesis, anthocyanins, carbohydrates, and growth traits, and characterized their gene expression in several grasses. These studies represent the first mapping effort of molecular markers and genes of perennial grasses. Results from these works laid the foundation for further molecular research. Because of these studies, additional funds were obtained to develop EST and BAC libraries for the *Leymus* and other grasses through Specific Cooperative Agreements with University of Illinois and Texas A & M University, respectively. DNA sequence information derived from these molecular tools is now available to the whole scientific community to accelerate the gene discovery and help plant improvement through marker-assisted selection. (Larson et al., 2009; Bushman et al., 2008; Kaur et al., 2008a and 2008b; Hu et al., 2001; Wei et al., 2000)

**Genetic analysis of perennial grass rhizomes.** Tiller and rhizome development plays an important role in establishment, persistence and productivity of perennial grasses. The ARS

scientists in Logan, UT have studied the genetic control of rhizome development in two perennial grass species, creeping wildrye and Snake River wheatgrass. A genetic linkage map has been developed for *Leymus* that has been used to detect major effect quantitative trait loci (QTL) controlling growth habit. Microarrays were used to assess gene expression in tiller and rhizome buds of these perennial wildryes. This work identified expression polymorphisms associated with gravitropism in subterranean branch meristems of *Leymus* wildryes. A genetic map for the *Elymus* species thickspike wheatgrass x Snake River wheatgrass was developed to further study rhizome development. (Kaur et al., 2008b; Larson et. al., 2006; Wu et al., 2003)

**Identification and genetic mapping of the seed shattering genes in native grasses.** In many grass species native and introduced seed shattering significantly reduces seed yields and quality, which increases seed costs. Often in wildland collected materials, seeds mature throughout the year, causing seed harvesting to be difficult. ARS researchers in Logan, UT discovered a gene that causes seed-shattering in *Leymus* wildryes and showed that it maps to the same chromosome location as genes causing seed shattering in American wildrice (*Zizania palustris*). Plant breeders in Logan, UT are using this gene to develop *Leymus* wildrye cultivars with improved seed retention, which in turn allows seed to properly mature before it is harvested. (Larson and Kellogg, 2009)

**Molecular characterization and release of salt-tolerant germplasms.** Salinity tolerance in Triticeae grasses is a quantitative trait controlled by many genes on different chromosomes. Furthermore, there are different mechanisms conferring salt tolerance. Scientists at Logan, UT in collaboration with scientists at U.S. Salinity Laboratory (Riverside, CA) had combined genes for salt tolerance from two sources into one so that the resultant germplasm lines had a higher tolerance than either parent. Being the most salt-tolerant germplasm lines of wheat in the world, W4909 and W4910 (PI 631164 and PI 631165) have been distributed to and used by 21 wheat breeders in 8 countries to pyramid salt tolerance genes into wheat cultivars. Material Transfer Agreements have been signed by 12 geneticists in 8 countries to receive seeds of Yecora Rojo x W4909 and Yecora Rojo x W4910 to study the inheritance of salt tolerance. The molecular data on mechanisms and genes conferring salt tolerance in W4909 and W4910 had been used by four groups of scientists elsewhere. (Mott and Wang, 2007; Wang et al., 2003b)

**Genetic mapping of grass forage quality traits in wildryes.** Basin wildrye and many other cool-season grasses have elevated K/(Ca, Mg) ratios (KRAT), which increases grass tetany potential in livestock. Moreover, protein and fiber properties of basin wildrye and other native grasses are relatively poor compared to cultivated forage grasses. Researchers at Logan, UT identified chromosome regions controlling fiber, protein, mineral content and grass tetany potential in two creeping x basin wildrye mapping families and, in particular, identified regions of *Leymus* chromosomes LG7a and LG7b that implicated in the grass tetany response. ARS researchers isolated and genetically mapped the 6-SFT gene in cool-season grasses. Identification and cross-species comparisons of chromosome regions and the 6-SFT gene locus elucidate key genetic factors involved in uptake of minerals, synthesis of soluble carbohydrates, forage quality, and reclamation of phytotoxic solid contaminated by heavy metals and acid forming compounds. (Larson and Mayland, 2007; Hu et al., 2005)

### **Release and Evaluation of Newly Developed Plant Materials**

Vast areas of semiarid rangelands in the western U.S. are severely disturbed, frequently burned, increasingly eroded, and subsequently infested with troublesome weeds such as cheatgrass (*Bromus tectorum*), medusahead rye (*Taeniatherum asperum*) and others. Reseeding these disturbed rangelands with genetically improved plant materials that are competitive enough (seedling establishment and persistence) to replace existing undesirable vegetation is often the most plausible and economically feasible way to reclaim such sites.

**ARS has a long history of developing and releasing native and non-native plant materials.**

As indicated in the following accomplishments, the ARS continues to release improved varieties for rangeland management. However the potential impacts of these releases are not immediately obvious but history shows that impacts can be significant. Data on the economic impact of ARS releases, particularly for rangeland grasses, are summarized in Appendix Three.

**Promoting squirreltail biodiversity.** The squirreltail complex encompasses five *Elymus* bunchgrass taxa that are valuable for restoration of weed-infested wildfire-prone lands. This important early-seral plant has been successfully adopted by the seed industry as a “new crop”, and it is in high demand by the BLM, owing to its ability to compete with invasive annual grasses that fuel wildfire. Six pre-variety germplasms have been released between 2003 and present, which include two high seed-yielding germplasms for rangeland restoration. A total of 242 acres of Sand Hollow, Fish Creek, and Toe Jam Creek germplasms were certified in 2009. During 2006 and 2007, the most recent major fire years, the BLM purchased 11,207 lbs. of Sand Hollow seed at a cost of \$336,572 and 8,963 lbs. of Toe Jam Creek seed at a cost of \$277,226 for rangeland rehabilitation projects. (Jones et al., 2004a and 2004b)

**Providing affordable Indian ricegrass seeds.** Indian ricegrass is a native rangeland grass important for wildlife, livestock and as a historical food for humans. In nature this grass has adequate seed-yielding attributes, but in agronomic settings seed production is limited by its indeterminate flowering and susceptibility to shattering, that limits mechanical seed harvest. ARS researchers in Logan, UT discovered that population PI 478833 possesses high seed retention following storm events and NRCS, ARS, and two universities released this population jointly as ‘Rimrock.’ Rimrock now dominates the seed trade for this species (59% of acreage from 2005-2009). Twenty-five years ago, Indian ricegrass seed sold for more than \$30 per pound. Although Rimrock seed initially sold for \$15.50 per lb. in 1999, it now retails for \$4.00 due to high harvestable seed yields. A total of 4,575 lbs. of Rimrock foundation seed has been distributed overall, and over the last five years, annual certified seed acreage has averaged over 600 acres. During 2006 and 2007, the most recent major fire years, the BLM purchased 256,074 lbs. of Rimrock seed at a cost of \$2,755,240. Lower seed prices have allowed Indian ricegrass to be used to make Montina™, a 25%-ricegrass gluten-free, affordable flour-blend product for sufferers of celiac disease. ARS researchers also found that Indian ricegrass is self-pollinating. As a result, seed-field isolation standards set by the national U.S. seed organization (AOSCA) have been changed to reflect these findings. (Jones et al., 2007)

**Recovery – a new grass cultivar to improve rangeland restoration.** Western wheatgrass is an important native grass, but its low rate of seed production and poor seedling vigor have limited its use when quick establishment is needed to restore degraded rangelands. ARS scientists at Logan, UT developed novel selection protocols and seeding practices for western wheatgrass, and worked with the U.S. Army Corps of Engineers and the NRCS to develop and jointly release ‘Recovery,’ a superior western wheatgrass. Developed and tested over a period of 10 years, Recovery was released in 2009 and designed for reseeding rangelands following severe disturbance, frequent wildfires, and soil erosion. With 20% increase in rate of successful establishment, Recovery allows land managers to use a native grass species to help limit weed infestation and soil erosion where reestablishment of wheatgrass is inhibited by frequent disturbance. Recovery is being recommended by the NRCS and the U.S. Army Corps of Engineers for reseeding private, public, and military training lands throughout the northern Plains and Intermountain West. Foundation seed sales have been strong with over 250 pounds already sold. (Waldron et al., 2011b, 2006a and 2005; Palazzo et al., 2005)

**Improving thickspike wheatgrass.** A native perennial grass, thickspike wheatgrass is widely used in rangeland revegetation projects. However, seed production and stand establishment are poor. ARS scientists at Logan, UT using plant evaluations and selection have developed improved populations that exhibit improved seed production and seed quality when compared to currently available varieties and native populations of thickspike wheatgrass. The seed trade and

restoration projects will benefit from more reliable seed production and stand establishment.  
(Robins and Jensen, 2010 and 2008)

## **Component 2: Pasture Management Systems to Improve Economic Viability and Enhance the Environment**

There is a need for sustainable technologies, germplasm, management practices and integrative strategies to improve the conservation and use of pasture agro-ecosystems to support livestock production and other natural resource uses.

### **Problem Statement D:**

Need for appropriate plant materials to improve the economic viability and enhance the environment in pasture-based livestock systems.

**Objective D.1:** Collect or develop and evaluate new plant cultivars that are biologically diverse, tolerant of biotic and abiotic stresses, more competitive, improved quality, and are easier to establish and maintain in pastures.

### **Enhancing Forage Grasses**

**Distinct lineages of reed canarygrass.** DNA marker and sequence analysis has identified three distinct European lineages of reed canarygrass: Scandinavian, Continental European, and Iberian. All bred cultivars in the USA and Canada trace to one or more of these lineages introduced to North America between 1880 and 1940. Initial results from samples collected across central and northeastern USA suggest that most common reed canarygrass along roadsides, riparian zones, and wetlands, are derived these European lineages and are not native varieties. These results are significant to conservationists concerned over the invasiveness of reed canarygrass, a high-yielding grass being considered as a biofuel feedstock. (Casler et al., 2009)

**Native grass persistence and performance in riparian areas.** Conservation Reserve Enhancement Program (CREP) requires more information on the suitability of locally adapted native grasses. ARS scientists at University Park, PA with NRCS plant materials specialists, evaluated five warm-season grasses and locally collected accessions of Virginia wildrye, a grass native to New England, for persistence and vigor under riparian conditions. The grasses were planted in Pennsylvania, Maryland, and New York on sites that would qualify for inclusion in the CREP program. The warm-season grasses prairie cordgrass, eastern gamagrass and switchgrass were most suited for riparian zones, whereas, Indiangrass and big bluestem performed relatively poorly. Virginia wildrye tolerated wet soils at all sites and seasonal flooding at some sites during three years. The locally adapted accessions performed as well as commercially available sources. This research was incorporated into NRCS recommendations for the use of native grasses in riparian zones. (Skinner et al 2009; Sanderson et al., 2010a).

**A new pasture species for humid, cool-season regions of the USA.** ARS scientists with the U. of Wisconsin documented naturalized meadow fescue populations that originate from early immigrants to the Upper Mississippi River Basin. Meadow fescue has been documented on hundreds of farms in the region and agronomic trials found very high forage quality and pasture suitability relative to other species. A new cultivar has been developed from this germplasm and others are currently in the development pipeline. Improved knowledge to optimize management of meadow fescue was also developed. (Casler et al., 2007; Brink et al., 2009; Brink et al, 2010a)

**New dallisgrass cultivar offers improved persistence and production.** ARS scientists at College Station, TX in collaboration with Louisiana State and Texas A&M evaluated the nutritive value, forage yield, and grazing tolerance of several dallisgrass biotypes. One biotype, Uruguayan, produced more forage and was more persistent under grazing than the common biotype. An accession of Uruguayan was recently released as the cultivar 'Sabine'. Producers in

the southeastern USA. have expressed interest in this new cultivar and seed are being increased for distribution. (Venuto et al., 2003; Burson et al., 2009)

**Improved low input grass licensed for southern pastures.** Bahiagrass is a productive forage grass that requires low inputs but one limitation is in newly seeded pastures where an establishment period of as much as 5 months is required before the grass can be grazed or cut for hay. ARS researchers at Tifton, GA have released TifQuik bahiagrass that has faster germination, fewer weeds during establishment, and can be cut for hay 45 weeks earlier than other varieties. It can also be use by growers interested in sod-based rotation systems. Three licenses have been granted for seed production and sales of the new variety and TifQuik was on the market for 2008 fall seeding. (Anderson et al., 2009)

**Development and release of new cultivars of indiangrass, a warm-season native grass and a native cool-season grass.** Three new improved indiangrass (*Sorghastrum nutans*) cultivars, 'Chief', 'Scout', and 'Warrior' were released for the Great Plains and the Midwest USA in forage-livestock production systems. 'Chief' is adapted to USDA Plant Hardiness Zone 4 (HZ 4) and the upper half of HZ 5. It produces significantly greater forage yields than the other available HZ 4 cultivars. 'Scout' is adapted to HZ 5 in the Great Plains and Midwest, USA and potentially other regions where it has not been tested to date. It produces significantly greater forage yields than other adapted indiangrass cultivars when grown for hay in the western part of its adaptation region. 'Warrior' is adapted to HZ 5 and the upper part of HZ 6 in the Great Plains and Midwest. It produces forage with high *in vitro* dry matter digestibility (IVDMD) that results in improved animal gains when utilized by beef cattle in well managed grazing systems in regions where it is adapted. In the regions where they are adapted, these cultivars also could be used in mixtures with other grasses in multi-species mixture to produce biomass for bioenergy. A Canada wildrye (*Elymus canadensis*) cultivar 'Homestead' was also released for use in conservation planting in the Midwest. (Vogel et al, 2010a; Vogel et al, 2010b)

**Switchgrass and intermediate wheatgrass – preferred warm season natives.** Limited statistics are available on acreages of forage grass cultivars. Based on foundation and certified seed records, the switchgrass and intermediate wheatgrass cultivars developed or co-developed by ARS scientists at Lincoln, NE are the most widely utilized cultivars of these species in the Great Plains and Midwest over the last two decades. The demand for cultivars for which grazing data are available greatly exceeds that of other cultivars. Seeded acreage of switchgrass and wheatgrass cultivars is estimated at one million acres and their use conservatively adds over \$20 million annually to the economy of the mid-continental USA. Seed demand for the two big bluestems, Goldmine and Bonanza, has exceeded supply since they were released. (Vogel et al, 2005a,b; Vogel et al., 2006a,b)

### Enhancing Forage Legumes

**Determining heritability of frost-seeded red clover establishment success.** In late winter after disappearance of snow cover, red clover is often broadcast seeded into forage legume-depleted grass pastures. This method of establishment is referred to as frost-seeding. An estimated 30-40% of frost-seeded pastures in Wisconsin, USA fail to establish. Breeding for increased frost-seedability has not been attempted by plant breeders. This study by ARS scientists at Madison, WI measured the genetic basis of frost-seedability. Inheritance of frost-seeded seedling establishment traits is very low, suggesting that developing new varieties for this trait will be difficult. (Riday, 2007)

**Yellow x purple flowered alfalfa hybrid seed production using leafcutter bees.** Yellow flowered by purple flowered alfalfa hybrids have been shown to produce increased hay yields. The alfalfa leafcutter bee is an important pollinator species used to produce alfalfa seed. This ARS study at Madison, WI examined leafcutter bee preference for purple flowered alfalfa versus

yellow flowered alfalfa. Higher amounts of yellow in the flowers of alfalfa plants were shown to produce less hybrid seed than would be expected without pollinator preference. (Riday, 2008)

### **Improving Resistance to Biotic & Abiotic Stress**

**Development and utilization of a salt screening protocol.** Improvements in plant materials for salt tolerance have been largely unsuccessful due to inadequate selection protocols. ARS scientists at Logan, UT developed a protocol to screen a large number of plant genotypes based on their ability to remain green under saline conditions. This procedure has resulted in the development of salt tolerant germplasm in alfalfa, grasses, and wheat. (Jensen, et al. 2005; Peel et al. 2004; Wang et al. 2003a,b)

**Incorporating genetic resistance to diseases in grasses.** Stem rust is the most damaging disease in grass seed production in the Pacific Northwest where over 400,000 pounds of fungicide at a cost of \$10 million are used annually. ARS scientists at Corvallis, OR developed the first stem rust resistant tall fescue germplasms. Four years of testing showed that during years with heavy stem rust infection, genetic resistant germplasm provided seed yields higher than commercial cultivars without use of fungicides. This new disease resistant germplasm is now utilized by most major tall fescue breeding programs in the US and Europe. (Barker, 2006)

**Objective D.2:** Provide greater efficiency in developing improved germplasm through collecting and characterizing germplasm; improving physiological, biochemical, and genomic techniques to describe and identify useful genetic traits; and improving tools and methods for developing improved forages.

### **Tools and Techniques for Improving Forages**

**Development of genomic tools for forage grasses.** Molecular tools for forage grasses are limited. To improve these tools, a large scale sequencing effort has been conducted by ARS researchers at Lexington, KY in collaboration with University of Kentucky and Mid-South Area Genomics Facility scientists. Roughly 140,000 cDNA clones from normalized meadow fescue/*Epichloe festucae* libraries and tall fescue libraries have been sequenced. Presently 128,189 EST sequences have been submitted to the National Center for Biotechnology Information database. Assembly of the plant specific cDNAs resulted in 42,154 unigenes, or unique genes. (Tavva et al., 2006; Tavva et al., 2007a,b; Tavva et al., 2008; Tavva et al., 2009; Florea et al., 2009; Dinkins et al., 2010)

**Genetic linkage maps of buffelgrass and Texas bluegrass now available.** ARS scientists at College Station, TX in cooperation with Texas A&M University scientists constructed genetic linkage maps of both buffelgrass (*Cenchrus ciliaris*) and Texas bluegrass (*Poa arachnifera*). The buffelgrass map included nine markers linked to apomixis (asexual reproduction). This linkage map provides a bridge for comparative genomic resources of sorghum and major cereal crops. Two markers on the Texas bluegrass map were linked to dioecy (separate male and female plants). These markers make it possible to identify the sexuality of Texas bluegrass plants at the seedling stage and saves plant breeders at least a year in determining which plants to use in a crossing block. A cDNA library was made available from buffelgrass through the publicly-accessible Genbank database <http://www.ncbi.nlm.nih.gov/Genbank/index.html>). Seventy EST-SSRs were identified and characterized from these buffelgrass cDNAs. (Jessup et al., 2003; Renganayaki et al., 2005)

**Molecular standards characterized for real-time PCR analysis.** There is very little information available on genes that might serve as internal standards during analyses of gene expression in grasses. Specific genes were identified at Corvallis, OR that are expressed at relatively constant amounts in different tissues throughout the life cycle of perennial ryegrass (*Lolium perenne*).

This accomplishment improved the quality, sensitivity and accuracy of gene expression analyses that quantify changes in the amount of gene expression that occur during the growth and development of forage and turf grasses. (Martin et al., 2008; Dombrowski and Martin, 2009)

**Storage of valuable grass germplasm.** ARS scientists at Corvallis, OR developed cryopreservation methods using meristem tissues for long-term genetic stock storage. This approach will preserve genetic diversity without maintaining isolated large field populations and will drastically reduce storage costs of these germplasm collections. (Reed et al., 2006; Clement et al., 2008)

**Improved detection of endophyte in grass germplasm.** An improved method to detect endophytes in seed and plant tissues from tall fescue, perennial and annual ryegrass was developed by ARS scientists at Corvallis, OR. DNA primers specific for the endophyte were developed and validated in a PCR approach where the endophyte was detected in a variety of grass species and tissues. This PCR method provided an accurate, sensitive approach to detect the presence of the endophyte in a diverse set of important forage and turf grasses, and will help plant breeders and seed testing laboratories in their assessment of germplasm quality and forage safety. (Dombrowski et al., 2006)

**Generation of tall fescue clone pairs with and without endophyte.** Tall fescue physiological and gene expression studies are best conducted on clones generated by propagation of vegetative ramets. To test effects of the seed-transmissible endophyte, *Neotyphodium coenophialum*, clone pairs are needed where one clone possesses the endophyte and the other lacks it. Such clone pairs were not widely available. ARS scientists at Lexington, KY used tall fescue cultivar Kentucky 31 plants infected with *N. coenophialum*, and tall fescue plants with novel endophytes to generate clone pairs. Each plant represented a distinct genotype. Plants were divided into ramets, and fungicide (propaconazole) treatment of some ramets of each plant generated endophyte-free clones. A total of 29 endophyte-infected and endophyte-free tall fescue clone pairs were generated. The clone pairs aid in designing experiments to test endophyte effects on tall fescue physiology and gene expression under various conditions such as water deficit stress, grazing, nematode parasitism or insect feeding. (Phillips and Dinkins, 2010)

**A new approach to improve plant stress tolerance.** ARS scientists at Corvallis, OR and Columbus, OH with university researchers found that tolerance to flooding was enhanced by expression of a gene construct that delayed plant senescence by causing the production of cytokinins. The impact and feasibility of delayed senescence on improving stress tolerance was previously unknown. These scientists created transgenic *Arabidopsis* plants and quantified survival, recovery, and cytokinin production in wild type and experimental plants subjected to flooding. This new approach will improve stress-tolerance improvements. (Chang et al., 2003; Huynh et al., 2005)

**Developing Tools to Assist in Plant Improvement.** ARS scientists at Lincoln, NE developed the Plant Adaptation Region concept to provide an ecological and climatic method of classifying plant germplasm for geographic regions. The Plant Adaptation Region concept, although recently published, has already been validated as a mechanism for defining adaptation regions for grasses including switchgrass and has been used in Pecan and urban ecological restoration research. It was described in a recent review (Current Opinion in Biotech.19:202-209, 2008) as an outstanding accomplishment. The scientists also developed the frequency grid for the rapid and reliable quantification of plant stands in swards. The Frequency Grid procedure is currently being used at over 25 research stations in the USA, and there are over 40 citations to this work. (Vogel and Masters, 2001; Vogel et al., 2005c, Schmer et al., 2006)

**An improved GC/MS method for quantization of n-alkanes in plant and fecal material.** Estimation of forage intake and dietary composition of grazing animals has been successfully

accomplished using the naturally occurring “fingerprint” of *n*-Alkanes present in the cuticular waxes of forage species. Total analysis time of current techniques is usually 16 to 18 hours per sample and solvent requirements are estimated at 35 mL per sample, making large studies with multiple samples costly and logistically difficult. ARS scientists at Lexington, KY developed a gas chromatography-mass spectrometry (GC/MS) method for the quantization of *n*-alkanes in forage and fecal samples. Extraction times were minimized to 30 minutes per sample as compared to more than 24 hours for the traditional gas chromatograph–flame ionization detection methods using saponification and liquid–liquid extraction. Method recoveries, including extraction efficiency, were greater than 91%. The linear dynamic range was 5 to 100 nmol injected onto the column, with limits of quantization less than or equal to 2.5 nmol. Intra-assay coefficients of variation for the analysis of annual ryegrass, subterranean clover, and bovine feces ranged from 0.1 – 12.9%, where lower concentrations of *n*-alkane produced a higher degree of imprecision. This method permits larger forage grazing experiments to be conducted. (Smith and Strickland, 2007)

**Forage fatty acids can be quantified using absorbance of infrared light.** ARS scientists at Beaver, WV and Akron, CO compared results obtained with three types of infrared instruments with data obtained with conventional wet chemistry methods. Scanning monochromator-grating-based infrared spectroscopy (SMNIRS) and diffuse reflectance Fourier transform mid-infrared (FTMIRS) and near-infrared spectroscopy (FTNIRS) performed similarly for estimation of concentrations of myristic, palmitic, stearic, palmitoleic, oleic, linolenic, and alpha-linolenic acids in 11 different forage species. Although calibration and validation statistics were higher for most fatty acids with SMNIRS than FTMIRS and FTNIRS, it is not clear if any of the infrared methods is distinctly better. Agreement between infrared and wet chemistry results show that the infrared technology can be used successfully for quantification of fatty acids in forage grasses, legumes, and forbs. (Foster et al., 2006a; Caldron et al., 2007)

**Comparison of detergent fiber analysis methods for forages high in pectin.** Pectin and tannin interfere with fiber analysis of broadleaf forages. Accurate determination of nutritive value is essential for efficient incorporation of such alternative forages into production systems for small ruminants. ARS scientists at Beaver, WV compared fiber analysis methods for grass, legume, tannin-containing legume, and brassica forage groups. Performing analyses in sequence on the same sample improved estimation accuracy of fiber concentrations in high-pectin brassicas and legumes compared to conducting analysis on a new sample for each step, but had no effect on results with low-pectin grasses. Neither method produced acceptable accuracy for tannin-containing legumes. (Cassida et al., 2007)

**Improved real-time assay for plant O-methyltransferases.** Plant O-methyltransferases are key enzymes in plant metabolism and play a crucial role in the generation of intermediates during lignin biosynthesis. Earlier assays for these enzymes, and ones specifically involved in lignin biosynthesis were cumbersome and/or involved the use of radioactive substrates. A new method developed by ARS scientists at Lincoln, NE relies on fluorescence as a means to detect and quantify the activity of these enzymes. This new assay can be used in genetic studies to modify lignin composition of biomass, which can affect its conversion to liquid fuels. (Palmer et al., 2008)

## **Understanding and Improving Plant Stress Mechanisms**

**High temperature and stress tolerance.** Although high temperature provides severe cropping risks, development of wheat with improved heat tolerance is limited by lack of knowledge of how anti-oxidative enzyme activity can reduce the impact on seed yield. ARS scientists at Corvallis, OR demonstrated that wheat superoxide dismutase activity decreases when the plants are exposed to a single heat shock and that this decrease in enzyme activity is accompanied by a significant increase in chelating activity. ARS scientists compared SOD and chelation activity in

plants exposed to a 24 hour exposure at 45 degrees Celsius and found significant heat-associated reduction in SOD activity. These results on basic mechanisms provides lines of research for improving heat tolerance in wheat and other cool-season grasses. (Banowetz et al., 2007)

**Plant adaptations to high temperature identified.** Changes in plant structures that permit certain grasses to withstand high soil and air temperatures in Yellowstone Park geothermal areas were identified. Microscopic analyses conducted by ARS scientists at Corvallis, OR compared plants adapted to geothermal areas to other isolates that were not adapted. These analyses showed that structures called trichomes were more numerous and thicker in adapted plants plus other adaptations at the cellular level also helped the plant maintain water in the hot environment. This research will help plant breeders develop new varieties of grass and wheat that have greater tolerance to hot weather. (Banowetz et al., 2008)

**Genes involved in post harvest stress tolerance identified.** At Corvallis, OR, 598 unique gene sequences associated with post harvest stress in grasses were identified. PCR based subtractive suppression hybridization was used in the model grass species *Lolium temulentum* to identify genes associated with post harvest stress. During grass seed production, grasses in the field are cut while still green and physiologically active, very little is known about the molecular processes at this stage of development. The isolation and identification of these genes provide valuable molecular tools to develop new genetic approaches to improve utility of grasses as forage and biofuel feedstock. (Baldwin and Dombrowski, 2006; Baldwin et al., 2007)

**Early signaling utilized by forage and turf grasses to sense their environment when exposed to wounding conditions.** Grasses are continually cut for hay and grazed by livestock, however very little is known concerning the molecular events that occur in grass plants as a result of wounding. ARS scientists in Corvallis Oregon identified the activation of a signaling protein in response to wounding in various grass plants. It is not known if the activation of this signaling protein in response to wounding induces defense compounds and genes or if it stimulates growth pathways to replenish lost tissues. This research provides the first step towards our understanding of these molecular signaling events and networks, which in the long term have the potential to improve and increase the yield, sustainability and quality of grass feed stocks utilized for livestock and biofuel production. (Brown et al., 2005; Dombrowski and Bergey, 2007)

**Identification of three genes involved in salinity stress tolerance.** Salinity is one of the major abiotic stresses responsible for reduced persistence, yield and biomass accumulation in many crops including forage grass. A gene encoding for a small protein involved in stress responses and a stress tolerance gene involved in production of protective compounds were found that apparently play a role in dehydration stress tolerance. The isolation and identification of these genes by ARS scientists at Corvallis, OR provide valuable tools for develop genetic approaches to improve stress tolerance in forage and turf grasses. (Dombrowski et al., 2008)

**Stem rust resistance source in perennial ryegrass discovered.** ARS Scientists at Corvallis, OR have identified a source of genetic resistance to the stem rust pathogen in perennial ryegrass. Stem rust is the most important disease problem in producing seed of cool-season grasses in the Northwest USA. Currently, no commercial cultivar of perennial ryegrass has a level of rust resistance sufficient to avoid use of fungicides for disease control. By repeated testing under controlled conditions, several individuals from a New Zealand cultivar of perennial ryegrass were found to have high levels of stem rust resistance. Identification of this host resistance source enables new research to characterize genetics of resistance, which will lead to the development of improved genetic material and strategies to incorporate stem rust resistance into perennial ryegrass. (Jo et al., 2009)

**Variants of the stem rust pathogen in ryegrass discovered.** Stem rust is the most important disease problem in producing seed of cool-season grasses in the Northwest USA, and no current commercial cultivar of perennial ryegrass has a level of rust resistance sufficient to avoid use of fungicides for disease control. Genetically purified cultures of the pathogen were isolated by ARS scientists at Corvallis, OR and tested against host plants with variable resistance to rust, it was determined that there is genetic variation for pathogenicity for these different isolates of the pathogen in the different host genotypes. This information will enable scientists and plant breeders to more effectively define, discover and develop disease-resistant lines of this important seed crop. (Pfender, 2009)

**Intermediate wheatgrass confirmed as a host plant for the wheat-stem maggot in Oklahoma.** ARS scientists in Woodward, OK observed damage consistent with that of the wheat-stem maggot in a field plot of intermediate wheatgrass. Adult wheat-stem maggot flies were reared from aborted reproductive culms of intermediate wheatgrass collected in western Oklahoma. Percentage of aborted inflorescences, determined by randomly sampling ranged from 10 to 28%. Intermediate wheatgrass was confirmed as a new host for this species of stem-maggot in Oklahoma. This information will help develop strategies for managing the pest. (Springer and Arnold, 2008)

**Improving Understanding and Management of Tall Fescue/Endophyte Symbiosis Regulation of loline alkaloid production.** ARS scientists at Lexington, KY with the University of Kentucky investigated the regulation of loline alkaloid levels in meadow fescue *with Neotyphodium uncinatum* and *N. siegelii*. After harvesting leaf blades, loline alkaloids in regrowth reached two- to five-fold higher levels than in the initial harvest. This effect was attributable to tissue age, because young leaf blades had much higher levels of lolines than did older leaf blades. The fungal loline alkaloid biosynthesis genes were not expressed more highly in regrowth over the initial harvest. However, the amino-acid precursors of loline alkaloids were much higher in younger than older leaf blades of endophyte-free plants. It was concluded that substrate availability rather than gene expression determined the loline-alkaloid levels. Understanding the relationship between free amino acids and alkaloids will facilitate plant breeding and management to control the alkaloid levels. (Spiering et al., 2008)

**Incidence of arthropod pests and natural enemies in tall fescue pastures expressing modified alkaloid profiles.** Grass breeders are introducing novel endophytes with attenuated alkaloid production into new or existing cultivars, but their use could alter the balance between arthropod pests and their natural enemies. ARS scientists at Lexington, KY with University of Kentucky researchers, collected vacuum samples in 2008 and 2009 and sorted samples for herbivores (leaf- and planthoppers, caterpillars, wireworms, grasshoppers, and crickets, and their natural enemies including spiders, predatory beetles, and parasitic wasps). There were few differences between pastures planted in endophyte infected or endophyte-free grasses. These results suggest that tall fescue cultivars harboring novel endophytes may be planted for improved livestock performance without increased risk of loss of natural enemies. (Potter et al., 2008)

**Drought stress tolerance in tall fescue.** ARS scientists in Lexington, KY and Athens, GA, in collaboration with University of Kentucky researchers, used clonal pairs of tall fescue plants with and without the endophyte in time courses of withholding water for zero to five days to simulate drought. Recovery from drought and changes in levels of free sugars, free sugar alcohols, and free amino acids were measured. In two experiments with one plant genotype, recovery upon re-watering was significantly greater for endophyte-infected than endophyte-free plants after two or three days of withholding water. Differences in metabolites were evident one day before establishing the difference in recovery, when shoot and root levels of glucose, fructose and proline were two- to three-fold higher in endophyte-infected than endophyte-free clones. In an experiment with a second plant genotype, similar but less dramatic effects of endophyte on recovery were observed, and the endophyte effects on metabolites were more evident in roots

than in shoots. Roots of endophyte-infected clones exhibited higher levels of free fructose (56  $\mu\text{mol/g dw}$ ) compared to endophyte-free clones (37  $\mu\text{mol/g dw}$ ). Levels of free glutamine and asparagine were three-fold higher in endophyte-infected roots than endophyte-free roots. These results demonstrate that the endophyte affects levels of free osmolytes that help protect plants against drought stress, thus providing managers with the additional management tool of using endophyte containing plants in drought prone areas. (Zhang, 2009)

### **Using Medicinal Plants to Improve Animal Health**

**More effective screening of medicinal plants for active ingredients.** Artemisinin, an antiparasitic chemical found in *Artemisia annua*, has been difficult and expensive to analyze. ARS scientists at Beaver, WV working with Chinese colleagues developed a simplified analysis of plant extracts for artemisinin using gas chromatography with electron capture detection. The method is sensitive to less than 9  $\mu\text{g/ml}$ , allowing dry samples as small as 100 mg to be accurately quantified. The method can be used to screen plant populations for high artemisinin production and to assess *A. annua* quality of plants and extracts destined for the pharmaceutical and medicinal markets. This more accurate and affordable test will improve quality control of artemisinin products here and abroad. (Liu, et al. 2008; Peng, et al. 2006; Ferreira and Gonzalez, 2009)

**Sweet wormwood is a nutritional medicinal plant.** Sweet wormwood contains compounds that can kill some parasites affecting small ruminants. ARS scientists at Beaver, WV working with international collaborators demonstrated that sweet wormwood is also a rich source of antioxidants, crude protein, vitamins, calcium, and potassium while the amounts of anti-nutritional compounds were negligible. This information will help in integrating sweet wormwood into diets of small ruminants to support nutritional and health needs. (Ferreira 2007; Ferreira and Gonzalez, 2008; Brisibe et al., 2009; Ferreira and Luthria, 2010; Squires et al., 2010a)

### **Enhancing Switchgrass for a Forage & Bioenergy**

**Processes regulating seed dormancy in switchgrass characterized.** Seed dormancy of switchgrass adversely affects both seed availability and stand establishment. ARS scientists determined the roles of nitric oxide and reactive oxygen species during switchgrass seed germination and results suggest at least two different, but uncharacterized, sensors for nitric oxide in switchgrass seeds. Discovery of these sensors opens the door to further study of seed dormancy and germination that may lead to improved stand establishment and seed availability. (Sarath and Mitchell, 2008)

**Results of large-scale switchgrass sequencing effort will lead to improved use of bioenergy crop.** In cooperative research, ARS geneticists and molecular biologists at Lincoln, NE and Albany, CA and cooperating university scientists developed approximately 425,000 expressed sequence tags (ESTs) of switchgrass containing a total of 265 million bases of sequence. This work will enable efficient selection in switchgrass based on inexpensive DNA-based markers and help translate research findings in other plants to help improve the performance of switchgrass as a bioenergy crop. The ESTs are being utilized in switchgrass genetic studies. (Tobias et al., 2008)

## **Problem Statement E**

Need for economically viable pasture-livestock systems for the Mid-South that enhance the environment.

**Objective E.1:** Provide improved forage-based cattle, small ruminant and horse production systems that meet economic and environmental objectives for the diverse environments found in the Mid-South.

## **Tall Fescue Management**

**Organic carbon in soil fractions differ in tall fescue systems.** ARS scientists at Lexington, KY, in collaboration with U. of Kentucky examined how endophyte-infected and endophyte-free tall fescue growth affected soil carbon fractions four years after establishment. Significant effects between endophyte-infected and endophyte-free fescue were observed for microbial biomass carbon, mineralizable carbon, carbon in micro-aggregates, and aggregate distribution, but not for total carbon, and particulate organic matter carbon. At zero to 15 cm, microbial biomass carbon (endophyte-infected 26% greater than endophyte-free), mineralizable carbon (endophyte-infected 43% lower than endophyte-free), carbon associated with micro-aggregates (endophyte-infected 15% lower than endophyte-free), and micro-aggregates (46% more micro-aggregates in endophyte-infected than endophyte-free) were affected by endophyte infection, confirming the hypotheses that early changes in soil properties are reflected in labile carbon fractions and soil structure. Endophyte status in tall fescue has quantifiable effects on carbon sequestration and soil structure achievable in a relatively short period that can be used to monitor conservation efforts including pasture renovation. (Handayani et al., 2010a,b)

**Approaches for managing recovery from fescue toxicosis.** ARS scientists at Lexington, KY with the U. of Kentucky conducted a two-year grazing experiment to evaluate weight gain and rates of recovery of steers exhibiting fescue toxicosis following feeding soy hulls during pasture grazing and post-graze anabolic steroid implantation. Rectal temperatures declined to normal in healthy animal at 5 days following removal from toxic tall fescue pastures. In both years, serum prolactin (low levels are a measure of intoxication) increased and stabilized at approximately 20 days following removal from the toxic tall fescue. At the conclusion of grazing, body weights were greater for the soy hull fed group (768 lb) than those receiving no hulls (737 lb). Further, there was an additive effect of steroid implantation on body weights (implantation = 1110 pounds; without implantation = 963 pounds). Results demonstrate that heat stress can be alleviated in 4 days in cattle grazing toxic tall fescue when removed from the pastures in late summer and that prolactin can increase and stabilize in 20 days. Feeding soy hulls at approximately 1% of body weight to yearling steers on fescue can improve weight gain, and when combined with anabolic steroid implantation can improve the efficiency of gain in the feedyard. (Aiken et al., 2008a,b)

**A hair cut and steroid implants improves heat tolerance of steers grazing toxic endophyte-infected tall fescue.** Cattle consuming toxic endophyte-infected tall fescue are prone to severe heat stress during warmer months, leading to decreased animal performance and occasional mortality. ARS scientists at Lexington, KY investigated the effects of hair coat clipping and steroidal implants on rectal temperatures, rates of sweating, and hair growth of beef steers grazing toxic endophyte-infected tall fescue. Rectal temperatures for clipped steers were lower than for unclipped steers at 84 days when the highest mean ambient temperature (33° C) was recorded (clipped = 39.3° C, unclipped = 39.5° C). Sweating rates declined as ambient temperatures increased and the rates tended to be higher with the estradiol than the progesterone-estradiol implant. Hair growth rates averaged 0.28 mm/d and were unaffected by the treatments. These results indicated that toxic tall fescue induced retention of rough hair coats, continuous growth of hair, and a reduction in sweating rate at higher ambient temperatures are factors that contribute to the vulnerability of cattle to heat stress during the summer. Estradiol implants may offer some benefit in reducing heat stress in cattle eliciting fescue toxicosis. (Aiken et al., 2006)

**Endophyte-infected tall fescue may be a tool for enhancing soil fertility and carbon dioxide sequestration.** Localized research from Georgia found that endophyte-infected tall fescue pastures supported larger soil nutrient pools, suggesting that the plant-fungal endophyte symbiosis plays a role in carbon sequestration and promoting soil fertility. ARS scientists at

Lexington, KY in collaboration with University of Kentucky scientists sampled more than 13 tall fescue pastures, with paired, adjacent endophyte-infected and endophyte-free plots, located across the southeastern United States. Surface soils were analyzed for organic carbon and total nitrogen and microbial community composition and biomass. The researchers showed that the Georgia findings are robust in that endophyte-infection appears to enhance the ability of tall fescue pastures to store carbon and enhance soil fertility across the southeastern USA, although effects on the microbial communities are less pronounced. These results highlight that the tall fescue–endophyte symbiosis can have significant environmental impacts in addition to well known animal health issues, and suggest that more research is needed to understand the mechanisms producing these results and potential use as an environmental mediator. (Siegrist et al., 2010)

**Evaluating horse pastures in Central Kentucky.** ARS scientists at Lexington, KY in collaboration with U. of Kentucky scientists, initiated research regarding microscopic analysis of horse manure to determine diet composition because pregnant mares grazing pastures containing tall fescue are vulnerable to severe fescue toxicosis (including death of foal and mare) and a validated method of readily assessing tall fescue consumption in production mares was not available for risk assessment. Results from this experiment showed that microscopic analysis of herbage fragments from horse manure can determine the percentage of individual cool-season grasses that horses consumed. Potentially, this technology can be a useful risk analysis tool to predict the potential for fescue toxicosis or diagnose extent of exposure in horses grazing tall fescue pastures in the central and eastern USA. (Morrison et al., 2009)

**Red clover produces beneficial antimicrobials.** Most bacteria in the gastrointestinal tracts of cattle are beneficial, but some rob the animals of nutrients. One group of detrimental bacteria, called the hyper ammonia-producing bacteria, *Clostridium sticklandii*, convert valuable amino acids into ammonia. The excess ammonia is converted to urea, and is lost in the urine and becomes an environmental pollutant. The hyper ammonia-producing bacteria can be controlled with antibiotics, but non-clinical use of antibiotics has contributed to antibiotic resistance, and recently led to a change in the FDA's stance on agricultural use of antibiotics. Fortunately, plants also make antimicrobials that can be used instead of antibiotics. ARS scientists in Lexington, KY determined if red clover produced beneficial antimicrobials capable of improving nitrogen utilization in the rumen. High performance liquid chromatography analysis revealed that clover tissues are rich in the isoflavonoids such as biochanin A. Biochanin A and crude clover extracts inhibited the growth of *C. sticklandii*. These results indicate that clover compounds may have a role in preventing amino acid degradation and provide a natural alternative to non-clinical antibiotics for improved ruminant nutrition and environmental protection. This work was recently mentioned in an ARS press release (<http://www.ars.usda.gov/is/pr/2010/100420.htm>). (Flythe and Kagan, 2010)

#### **Reducing input costs and improving viability in livestock production systems.**

Appalachian silvopastoral systems can increase livestock carrying capacity and contribute to a reliable supply of high quality herbage. ARS researchers in Beaver, West Virginia found that forage grown under moderate tree canopies had higher crude protein (CP) than similar forages grown in open pastures. Understory forage was lower in energy than open pasture herbage so managers need to ensure there are adequate sources of energy available to utilize the CP. Soil in woodlots converted to silvopasture quickly takes on characteristics similar to improved pasture, and herbage yield is approximately 60% that of traditional pastures. Grazing livestock in woodlots can provide an additional low-cost source of CP, extend the grazing season, and reduce need for supplemental feed. An agroforestry approach to woodlot management supplies both forages and wood products, and turns land that is often considered marginal into a more viable part of the farm enterprise. (Neel et al., 2008; Staley et al., 2008; Feldhake et al., 2010)

#### **Parasite Control in Small Ruminants**

Niche markets for small ruminants offer limited resource farmers opportunities to engage in profitable livestock operation. However in the southeast, developing this market opportunity has been limited by a serious parasite problem. Because of resistance to commercially available dewormers, alternative methods of control are needed.

**Alternative methods for controlling parasites in goats and sheep.** *Haemonchus contortus*, an abomasal species of nematode, is a major problem for sheep and goats in warm, humid climates worldwide. Copper oxide wire particles (COWP) were developed to treat copper deficiency and reduce infection of *H. contortus* in sheep and goats. ARS scientists at Booneville, AR working with Fort Valley State University and Louisiana State determined that *H. contortus* could be controlled in goats and sheep when COWP was administered in a bolus or in the feed and may be more effective when combined with sericea lespedeza. The COWP was much more effective when *H. contortus* is the predominant parasite. COWP costs less than 10 cents per animal to administer compared to 70 cents for the more commonly used chemical dewormers, which are often ineffective. Because of resistance, producers are being encouraged to use more than one chemical dewormer and this practice can cost more than a dollar per animal. COWP offers an effective and affordable alternative to help in controlling parasites in both goats and sheep. (Burke and Miller, 2006; Burke et al., 2007a,b, 2010a,b; Soli et al., 2010)

**Controlling gastrointestinal parasites in sheep and goats in the southeast.** Rotational grazing of pastures as a means to control internal parasites has been suggested by the U.S. National Organic Program, but has never been examined without the use of chemical dewormers. ARS scientists at Booneville, AR, working with Louisiana State and Fort Valley State University, determined that rotational grazing of lambs on bermudagrass led to fewer deworming treatments than animals not rotated. Along with scientists at Auburn University, they also determined that diets containing 50 to 75% of dried sericea lespedeza can reduce worm problems in small ruminants while providing nutrients for the animals when compared to diets lacking the lespedeza component. A patent was issued on this technology and a small family farm business will commercialize a pelleted form of the dried sericea lespedeza to control worms in small ruminants. (Burke et al., 2009; Lange et al., 2006; Shaik et al., 2006; Moore et al., 2008; Terrill et al., 2007, 2009)

**FAMACHA necessary to maintain dewormers for sheep and goats in the southeastern US.** The FAMACHA system is a tool used to determine which animals need dewormer to control *Haemonchus contortus* or barber pole worm. This selective treatment will minimize dewormer used and extend its efficacy. Scientists at USDA, ARS in Booneville, AR in cooperation with Fort Valley State University, University of Georgia, Louisiana State University and others determined that the FAMACHA system can easily be used by producers affected by *H. contortus*. Scientists also determined that the FAMACHA system can be used to identify superior sires for parasite resilience/resistance, thus increasing flock resilience, and perhaps resistance. More than 5,000 U.S. farmers have been trained to use FAMACHA and more than 16,000 cards have been distributed in the USA. A survey of producers attending workshops on worm control indicated a high percentage of producers were satisfied with the FAMACHA system and it saved them money through reduced use of dewormer and even more importantly, increasing the lifespan of dewormers (Burke et al., 2007c; Burke and Miller, 2008; Whitley et al., 2010)

**Orange oils have potential for controlling barber pole worms in small ruminants.** ARS scientists at Beaver, WV showed that orange oils inhibit embryonation and hatching of barber pole worm (*H. contortus*) eggs and inhibit the motility of the infective worm larvae, but do not adversely affect metabolism of rumen microorganisms. With their collaborators at Virginia Tech, they showed that orange oils administered to sheep reduced the number of parasite eggs excreted in the feces by 90% and the number of worms in the gastrointestinal tract by 50%. Thus, orange oils have promise as a natural dewormer. (Foster et al., 2009b; Rosскопff et al., 2008; Squires et al., 2010b.)

**Anthelmintic potential of chicory forage is influenced by sesquiterpene lactone composition.** Chicory is used in small ruminant pastures to help control gastrointestinal parasites like *H. contortus*. Anthelmintic activity is attributed to the presence of natural compounds called sesquiterpene lactones. ARS scientists at Beaver, WV showed that one of the three principal sesquiterpene lactones, 8-deoxylactucin (DOL), is the primary constituent responsible for the anthelmintic activity of chicory. They analyzed forage from three cultivars during the growing season and showed that the cultivars Lacerta and Puna have consistently high DOL concentrations and, therefore, were useful in integrated programs for gastrointestinal parasite control in small ruminants. With ARS and university collaborators in Pennsylvania, Ohio, and Mississippi, they showed that most of the other forage chicory cultivars available to US farmers (Choice, Oasis, Six Point, and Laniña) are similar to Puna and Lacerta in sesquiterpene lactone composition. The cultivar Forage Feast has much less DOL. (Foster et al., 2010; Foster et al., 2009a; Foster et al., 2006b)

**Meat goats will eat chicory containing beneficial anthelmintic compounds.** Forage chicory contains compounds that may help goats tolerate worms, but the bitter taste of the forage has reduced intake in other livestock species. Scientists at the ARS lab in Beaver, WV determined that goats could detect bitterness differences among chicory forages, but this did not reduce their willingness to eat it. Therefore, chicory with greater amounts of beneficial compounds can be fed to goats in order to provide an alternative to ineffective commercial dewormers. (Cassida et al., 2010)

**Objective E.2:** Provide improved pasture management practices to improve and maintain forage production and quality cost effectively while enhancing the environment.

### **Need for Better Adapted Forages**

Climatic variation and higher costs for inputs such as nitrogen fertilizer have generated a need for forages better adapted to local conditions. The availability of tall fescue with novel endophytes has created interest in how these can be used effectively.

**Gamagrass as an alternative warm-season forage in the Mid-Atlantic Region.** ARS scientists at Raleigh, NC evaluated eastern gamagrass production and quality to determine if this grass was suitable for the Mid-Atlantic Region for pasture, hay and silage, and bioenergy. Eastern gamagrass, a native grass, requires lower inputs of nitrogen per unit of production compared to many introduced pasture grasses. Eastern gamagrass pastures were grazed to obtain estimates of steer daily gain and potential pasture production, and cut and preserved as both hay and silage to determine its nutritive value and quality. Eastern gamagrass was found to be a viable lower-input alternative warm-season. In wet soils, eastern gamagrass yields are better than switchgrass. Eastern gamagrass was found to be particularly suitable on poorly drained soils. (Burns and Fisher, 2010a,b; Sauve et al., 2010)

**Caucasian bluestem is an appropriate forage for the Mid-Atlantic Region.** Experiments were conducted by ARS Researchers at Raleigh, NC to determine if Caucasian bluestem was sufficiently pliable to be used as a pasture and hay source in the Mid-Atlantic Region. Caucasian bluestem was grazed to obtain an estimate of steer performance and potential pasture production, and cut and preserved as hay to determine its nutritive value and quality. Caucasian bluestem, also a possible candidate for cellulosic production for biofuel, is sufficiently pliable to be an integral part of ruminant production systems in the Mid-Atlantic Region. (Burns and Fisher, 2010d)

**Cool-season grasses with drought tolerance.** Cool-season grasses are an essential component of animal production systems across the north-south transition zone of the USA. Hays made with a grass with improved drought tolerant, 'Persist' orchardgrass, was compared with two improved tall fescue cultivars ('MaxQ' and 'Ark+', both with nontoxic novel-endophyte).

All were found by a feed trial with sheep to be similar in quality, whereas steers consumed more of the 'Persist' orchardgrass but digestible dry matter intakes were similar. Improved tall fescue selections with a nontoxic novel endophyte or orchardgrass can be used in production systems depending on agronomic characteristics desired. (Burns and Fisher, 2010c)

**Impact of grazing novel endophyte infected tall fescue on beef heifers.** Ruminants grazing toxic tall fescue have reduced feed intake, average daily gain, and poor conception rates costing livestock producers in excess of \$600 million annually. Tall fescue cultivars have been developed that contain a novel endophyte that does not produce the toxic ergot alkaloids that cause fescue toxicosis, but comparisons of breed types of growing heifers grazing different tall fescue cultivars are limited. ARS scientists from Booneville, AR with cooperation from the U. of Arkansas evaluated the effects of toxic and novel endophyte infected tall fescue on growth and grazing behavior of pregnant Brangus and Gelbvieh x Angus heifers. Average daily gain was not different between breed types. However, heifers grazing novel endophyte infected tall fescue were heavier than heifers grazing endemic endophyte infected tall fescue. Fewer heifers grazing tall fescue infected with the endemic endophyte were observed grazing during the hot time of the day than heifers grazing novel endophyte infected tall fescue. Using novel endophyte infected tall fescue in a grazing program can increase the performance of pregnant beef heifers compared to heifers consuming endemic endophyte infected tall fescue and this is partially due to more time spent grazing. (Bailey et al., 2009)

### **Managing Environmental Impacts**

**Effect of tall fescue containing an endemic endophyte on bluebird reproduction.** ARS scientists at Booneville, AR, with Arkansas Tech University scientists, determined that Eastern bluebirds (*Sialia sialis*) laid smaller eggs, nestlings tended to be in poorer condition, and weight of nestlings at the time of first flight was lower in endemic endophyte infected tall fescue pastures compared with endophyte-free tall fescue pastures even though endemic infected tall fescue pastures were greater in arthropod numbers so food was more available. While the specific factors adversely affecting bluebird performance are not clear, preservation efforts for the birds should avoid endemic endophyte-infected tall fescue pastures. (Arkansas Tech University Thesis: Effects of Endophyte-Infected Tall Fescue (*Festuca arundinacea*) on Eastern Bluebird Reproduction by Dean Sedgwick (ATU), Chris Kellner (ATU), and David Burner (ARS Booneville). Successfully defended June 26, 2009.)

**Reducing undesirable environmental impacts when applying dry poultry litter to pastures.** Poultry litter provides nutrients for perennial forages, but the current practice of spreading litter on the surface of pastures allows significant nutrient losses that decrease nutrient use efficiency and often result in serious air- and water-quality problems. ARS scientists at Booneville, AR found that placing poultry litter applications below the pasture surface can reduce these negative environmental impacts by more than 90% (Sistani et al., 2009). A prototype machine known as the ARS Poultry Litter Subsurfer was developed in cooperation with the National Soil Dynamics Laboratory to make injection a practical option for producers. A provisional patent application entitled "System for Distributing Poultry Litter below the Soil Surface" was filed for this technology by USDA-ARS in July, 2008 and converted to a standard, utility-type application for U.S. patent rights in July, 2009. The USDA-ARS also filed an application for international patent rights. This technology has the potential to improve pasture productivity and help solve both air- and water-quality problems for thousands of producers on millions of acres. (Pote et al., 2009)

**Reducing the prevalence of *E. coli* and *Salmonella* in ruminant livestock.** Reducing the shedding of *Escherichia coli* 0157:H7 and *Salmonella* in beef cattle feces reduces risks to human health. ARS scientists at Booneville, AR working with the U. of Arkansas and ARS scientists at College Station, TX evaluated management practices to reduce pathogen shedding. They found that feeding hay to grazing cattle reduced *E. coli* shedding and feeding the

dewormer, fenbendazole, reduced *Salmonella* shedding (Looper et al., 2006a). The Booneville scientists working with scientists from the ARS station in Lexington, KY and U. of Arkansas found that reducing the presence of muddy conditions around cattle watering tanks decreased the prevalence of *E. coli*. Also, fecal shedding of *E. coli* tended to be greater in sheep fed diets of endophyte-infected tall fescue compared with low endophyte, but feed intake was lower. There were no differences in shedding of bacteria in beef cows grazing endophyte-free or infected tall fescue. (Looper et al., 2007; Looper et al., 2009a,b)

## **Problem Statement F**

Need for economically viable forage-livestock systems for the Great Plains.

**Objective F.1: Develop forage-based livestock production systems that will lower production costs, reduce the need for harvested feedstuffs, increase marketable yields of animal products, and enhance the environment.**

### **Improving Performance of Forage-Livestock Systems**

**Assessing different management of grazing systems that increase production and forage quality with lower inputs.** The Great Plains region uses grazing systems that are normally a combination of warm-season perennial grasses and wheat or perennial cool-season forages. ARS researchers at El Reno, Langston, and Woodward, OK have compared numerous grazing systems for production, quality and reduce inputs. Though wheat is an excellent forage capable of producing stocker weight gains of greater than 3 lb/day, it can accumulate high levels of nitrate that could pose a health risk to the animals. ARS researchers at El Reno, OK over a four year period compared the levels of crude protein and nitrate in fall forages clipped from winter wheat pasture and perennial cool-season grass pastures of tall wheatgrass, smooth bromegrass, and intermediate wheatgrass. In two out of four years, winter wheat forage contained nitrate levels that could pose a risk to cattle. In contrast, the level of nitrate in the perennial cool-season forages measured each of the four years did not pose a risk to cattle. Levels of crude protein in the perennial cool-season forages were as much as 37% less than that of wheat but were still sufficient for growth of stocker calves. ARS scientists at Langston have demonstrated that cool-season grasses can be established by no-till seeding into dormant warm-season pasture, without the need and cost of herbicide suppression of the existing vegetation. Because no-till seeding avoids destruction of the existing warm-season pasture, the year-long forage yield was increased by no-till compared with conventional planting of cool-season grasses. This no-till over-seeding system has been most effective with annual ryegrass, which has allowed an average year-long forage yield increase of 19% above unimproved warm-season pasture and increases of 37% are observed with the addition of Korean lespedeza. Farmers who have sown annual ryegrass for forage have reported that they have been able to reduce their purchases of hay for feeding during the cool-season. ARS scientists at Woodward, OK, in collaboration with Kansas State University, evaluated the costly practice of burning or spraying herbicides to control weedy species on yellow bluestem pastures to increase pasture productivity. They found that these practices are not necessary for the optimal management of Yellow bluestem pastures and do not increase livestock performance. Adequate fertilization and proper grazing management seem to be the most effective annual treatments for the management of Yellow bluestem pastures. Eliminating ineffective practices results in a substantial savings to the producers and reduces adverse health and environmental impacts. (Bartholomew, 2005; Bartholomew & Williams, 2008; MacKown and Northup, 2010; Gunter and Gillen, 2010).

**Reduction of nitrate toxicity through managed grazing for the Great Plains.** Winter wheat is the primary cool-season forage of the southern Great Plains but, like other cool-season grasses, winter wheat can accumulate high levels of nitrate that are a threat to ruminant health. ARS researchers at El Reno, OK found that nitrates do not collect evenly throughout the above-

ground biomass of the winter wheat plant. Nitrate concentration in the leaves is substantially lower and thus the leaves are safe, particularly when compared to the stem-like tissue of the juvenile wheat plants that is very high in nitrates. Extensive grazing systems with light stocking rates are much safer than rotational grazing with intensive stocking rates that force the livestock to graze higher nitrate biomass. (MacKown et al. 2007)

**Improving pasture persistence, forage quantity and quality of intermediate wheatgrass and alfalfa in the northern Great Plains.** ARS scientists at Mandan, ND evaluated the impact of grazing management on plant productivity, persistence and soil quality to improve profitability for cattle producers. Intermediate wheatgrass is a high quality, high yielding perennial grass that is limited by a lack of grazing persistence. The researchers found that grazing intermediate wheatgrass prior to late boot enhanced persistence. Demographic information gathered during the study resulted in the release of 'Manifest' intermediate wheatgrass, which has improved persistence under grazing. The impact of grazing on soil bulk density, pH and organic carbon was negligible implying grazing did not negatively impact critical soil function. Intermediate wheatgrass pasture can be part of sustainable crop rotation systems because the grass persists and grazing does not result in soil compaction that adversely affects follow-on crop production. Using alfalfa in grasslands can enhance quantity and quality of the overall pasture but more knowledge is needed on how defoliation can impact persistence and productivity. The researchers found that including alfalfa increased total biomass yield from 38 to 185% without decreasing grass yield. A biomass alfalfa entry (SCMF 3713), released as AC Yellowhead, produced the most biomass. AC Yellowhead and one of the hay-type alfalfa lines 'Vernal' produced the most biomass when clipping was delayed until the flowering stage. A concurrent soil evaluation suggested that both hay and grazing types of alfalfa could increase stock of soil C and N in grasslands. This research suggests that incorporating alfalfa into grassland can increase total biomass without a decrease in associated grass yield while at the same time enhancing soil C and N. (Hendrickson et al., 2005; Liebig et al., 2008a; Hendrickson et al., 2008; Liebig et al., 2010b)

### **Greenhouse Gases and Grazing Management**

#### **Assessment and mitigation of greenhouse gas emissions from grazing practices.**

Greenhouse gas emissions (GHGs) produced from cattle grazing has been cited as contributing to global warming but little is known of the role of grazing lands as net sinks or sources of GHGs. ARS scientists at Mandan, ND have estimated current GHGs and possible methods of reducing methane emissions from cattle. The scientists estimated net global warming potential (GWP) for three grazing management systems located in central North Dakota. The team measured soil organic carbon change and nitrous oxide and methane flux in the three grazing treatments, and combined their findings with estimates for methane emission from cattle (via enteric fermentation) and carbon dioxide emissions associated with producing and applying nitrogen fertilizer. Summing across factors, net GWP was negative for native vegetation pastures, implying net removal of GHGs from the atmosphere during the years of the study. This finding underscored the value of grazed, mixed-grass prairie as a viable agroecosystem serving as a net GHG sink. Intake of some types of condensed tannins by sheep, cattle, and goats can reduce their emissions of methane by as much as 30% and their urine urea excretion by as much as 20%. Though many commonly grazed or fed forages do not contain condensed tannins, grazing livestock may be supplemented with condensed tannin in their drinking water and studies concluded that ruminant livestock will ingest small amounts of condensed tannin via their drinking water. The ARS scientists also quantified the effects of tannin-affected cattle urine and normal cattle urine on carbon dioxide, methane, and nitrous oxide flux over a six-week period in a mixed grass prairie. Though the tannin urine treatment possessed 34% less N than normal cattle urine, cumulative nitrous oxide emission between the treatments did not differ. Furthermore, methane uptake from the tannin urine treatment was 40% less than the normal urine treatment. Results from these studies suggest the use of condensed quebracho tannin as

a dietary amendment for livestock does not yield direct GHG mitigation benefits in the short-term. However, short-term emissions ammonia was not evaluated and should be in the future to fully assess this approach for GHG mitigation. (Krongberg, 2008; Krongberg, 2010; Liebig et al., 2008b; Liebig et al., 2010a)

**Assessment of effects of soil compaction on variation among Italian ryegrass cultivars.**

There is a lack of information on how no-till systems affect variety performance of Italian ryegrass. ARS scientists at Langston OK demonstrated that increased soil compaction under no-till generally slowed seedling growth and development. However, the relative productivity of a range of commercially-available cultivars of Italian ryegrass and tall fescue forage grasses was unchanged by increased compaction. The work suggests that cultivar rankings determined under conventional cultivation and sowing are valid for no-till compacted soils. (Bartholomew & Williams, 2010)

**Objective F.2:** Develop cost effective management practices that improve pasture establishment, persistence, and forage quality while enhancing the environment.

**Assessment of current and proposed management practices that can assure pasture and cattle productivity.** Researchers at Langston and Woodward, OK have determined methods to insure sustainable pastures and the appropriate use of mineral supplements for improvements in overall cattle production systems and to increase net profitability. In four years of studies ARS scientists at Langston, OK found that although annual ryegrass could reseed itself for up to two years after initial sowing, it was very inconsistent (over 40% of failed to reestablish). In concurrent studies of reduced-input methods of planting they found that ryegrass can be satisfactorily established by broadcast sowing methods that minimize time and equipment needs. For resource-limited farmers annual planting of ryegrass is likely to prove more productive and more reliable than dependence on re-establishment by self-seeding. ARS scientists at Woodward, OK found that forage intake rate of wheat increased as sward surface height increased, resulting in steers that grazed the tallest pastures being the most efficient since cattle were able to reach the same forage intake level in less time. Cattle rotation schedules in the Southern Great Plains should allow swards to grow taller before turning in livestock and pasture size and the time animals are allowed adjusted to favor grazing that balances nutritional needs with grass height. Other Woodward scientists found that supplementing cattle with available free-choice mineral mixture (Ca, 16% and P, 4%) in a wheat-grazing production system improved body weight gains of 30 to 45% faster than cattle not offered minerals. Supplemented cattle weighed 6 to 8% more after grazing than non-supplemented cattle. Mineral intakes averaged 73 to 168 g/d, resulting in a cost of supplement to kilogram of added body weight gain conversions of \$0.26 to \$0.64 assuming a mineral cost of \$0.88/kg. Producers can improve the ADG of cattle grazing winter-wheat pasture on the Southern Plains and the supplement to added body weight gain conversion appear profitable (Gregorini et al., 2009b; Bartholomew & Williams, 2009; Gunter and Combs, 2010)

**Improved cattle gain with improved management system for the Great Plains.** The primary two-forage system that includes a warm-season forage with a cool-season forage has land-use and seasonal gaps in forage availability and quality that limits the flexibility and profitability of the stocker enterprise. ARS scientists at El Reno, OK and Langston, OK found that no till seeding of cool-season grasses into dormant warm-season pasture can increase cool-season forage availability without adversely affecting the productivity of the existing warm-season pasture. The research also showed that broadcast seeding was less effective than drill sowing in stand establishment, but there was no difference in harvested yield in the spring between sowing methods. The reduced cost of broadcast seeding rather than drill seeding, and the limited equipment required for broadcast seeding makes this an attractive option to limited resource small farmers seeking to extend their grazing season and increase profitability. They also found that replacing a portion of the winter wheat fields with endophyte-free tall fescue could significantly increase animal gains. With minimal management, the addition of the tall

fescue extended the grazing season by 30 days and produced comparable gains as using winter wheat alone but at lower cost. By increasing management and doing intensive grazing (2 or 3 times normal stocking rates) for 35 days in the fall and spring when winter wheat and warm-season grasses were not available, an additional 145 pounds of gain per head were obtained. Using this information, producers can increase cost-effective gains and have greater flexibility in marketing livestock. (Bartholomew and Williams, 2008)

**Alternative annual summer legumes.** ARS scientists at El Reno, OK explored grazing annual summer grain legumes to fill gaps in pasture forage quality and quantity in the late summer and early fall. Pigeon pea, cowpea, mung bean, and guar were found to have the potential to provide high quality forage with mung bean, cowpea and guar using less water than pigeon pea or soybean. Greater water-use efficiency could make these legumes more effective in winter wheat-summer legume rotation systems. ARS research included importing pigeon pea germplasm from south Asia and has enabled Texas AgriLife and Tennessee State University to begin research on developing this legume as alternative legume grain crops for specialty markets. (Rao and Northup, 2009a.)

**Alternative cool-season grain legumes.** ARS scientists at El Reno, OK have also explored using cool-season grain legumes such as grass pea (*Lathyrus sativum*) and lentil (*Lens culinaris*) to provide high quality forage during the late spring and early summer forage gap. Among the cool-season grain legumes, grass pea has the potential to capture greater amounts of nitrogen than other cool-season grain legumes. Publishing these research findings in popular farm magazines resulted in a 434% increase in sales of grass pea seed within the Great Plains region. These research findings were also published in the trade journal, European Association for Grain Legume Research Magazine. This research was recognized and awarded as a “Notable Technology Development” for the mid-Continent Region by the Federal Laboratory Consortium. (Rao and Northup, 2008; Rao and Northup, 2009b)

## **Problem Statement G**

Need for economically viable pasture-livestock systems for the Northeast and North Central States that enhance the environment.

**Objective G.1:** Develop new management and supplementation practices to optimize the utilization of mixed-species cool-season pastures by grazing dairy cattle to produce products tailored to meet traditional and niche markets of the Northeast and North Central states.

## **Forage-Livestock Interactions**

**Assess grazing behavior of cattle in biodiverse pastures.** ARS scientists at University Park, PA have evaluated planting dairy pastures in a mix of grasses and other forage species. They found such mixtures increase forage productivity and maintain production more consistently during seasonal and annual climatic variations. These scientists then determine if this greater diversity of plants changes the grazing behavior of cattle and affects animal performance. They offered four grass species to cattle in short-term intake trials and found that plant height had the largest and most consistent effect on cattle grazing behavior. They also found that grazing was more efficient on taller plants because the cattle were able to take more and larger bites. Producers can influence grazing efficiency by managing forage height by regulating grazing schedules and intensity in established pastures. In establishing and rehabilitating pastures, they can select forage mixtures that favor taller plants that still meet nutritional and seasonal requirements and are adapted to local conditions. (Soder et al., 2009)

**Pasture canopy structure influences potential stocking capacity and animal performance.** Dry matter yield is a primary criterion determining the value of a temperate grass for pasture.

When intensive rotational grazing is employed, however, animals frequently have high nutritional requirements and are not forced to consume all the dry matter produced by the grass. The dry matter yield contained in different horizontal layers of the pasture canopy, and its quality, thus have an influence on potential stocking capacity and nutrient intake. Research at Madison, WI on meadow fescue, orchardgrass, quackgrass, and reed canarygrass has identified differences in canopy structure of these grasses. Meadow fescue possesses a canopy structure that supports fewer animals during portions of the growing season but has greater quality throughout the canopy. The results provide guidelines by which producers may select and utilize different temperate grasses in rotational grazing systems. (Brink et al., 2008)

#### **Precision feeding of grazing dairy cows improves performance and environment.**

Precision feeding of dairy cows is a concept often used in confinement operations but not with dairies using grazing. ARS scientists at University Park, PA investigated new techniques of precision feeding energy (corn silage) and protein supplements to grazing dairy cattle. Two key findings were: 1) Supplementing a pasture-based diet with increasing levels of supplemental protein changed ruminal fermentation patterns, reduced nutrient digestibility, and altered ruminal pH and nitrogen metabolism. These nutritional imbalances result in increased feed costs and decreased nutrient digestibility by 8%, which in turn would increase nitrogen losses to the environment. 2) Providing supplemental energy in the form of corn silage to grazing cows 9 hours rather than 1 hour before a grazing period improves ruminal fermentation and digestibility and may reduce environmental impact of nitrogen losses in grazing cattle. Precision feeding of dairy cows on pasture can save farmers money, increase milk production, and reduce environmental pollution. (Gregorini et al., 2010; Soder and Gregorini, 2010)

**Molasses as a feeding supplement on organic dairy farms.** Escalating organic grain prices and changes in milk contracts have forced organic dairy farmers to seek alternative energy sources for organic dairy cows. Sugar cane molasses, a rich source of sugars, may be a cheaper source of supplemental energy and minerals than organic grain. However, milk production responses to molasses supplements have been variable. ARS scientists at University Park, PA compared pasture supplementation with molasses or corn meal on ruminal fermentation in laboratory simulations of ruminal metabolism. Molasses supplementation increased protein digestibility and decreased ruminal ammonia concentrations by 18% (compared to a pasture-only diet) when fed in combination with corn meal, resulting in improved nitrogen utilization. However, there was no effect on dry matter or fiber digestibility, ruminal pH, volatile fatty acids, or microbial protein production. At low levels of supplementation, molasses showed similar results to corn meal in improving ruminal fermentation and nitrogen utilization with both supplements only showing minimal improvement compared with a pasture-only diet. (Soder et al., 2010)

**Pasture is "easier" to chew in the evening.** Cud chewing is an important part of the digestion process in cattle and things that make forage difficult to chew and digest can reduce cattle performance. ARS scientists at University Park, PA evaluated changes in the "toughness" of pasture forage in relation to its diurnal variation in chemical composition. Using an indirect measure of how hard forage is to chew, toughness decreased later in the day and this corresponds to a decrease in fiber and protein with an increase in sugar content. These results help explain why cattle graze longer and more intensely in the evening. (Gregorini et al., 2009a)

**Pasture finishing sheep and goats can be acceptable for niche markets.** The rapid growth in Halal markets for small ruminants is an economic opportunity for small producers. At Beaver, WV, ARS scientists evaluated production systems that rely on forages to reduce the need for supplementation. Three breeds of hair sheep (Barbados Blackbelly, Katahdin, and St. Croix) were put on alfalfa hay diets and evaluated for forage intake and utilization. Growth without energy supplementation was moderate and did not allow the Katahdin, an improved breed, to express its higher growth potential when compared to the St. Croix. Weight gain and carcass quality was also compared between traditional lambs (Suffolk), hair sheep lambs (Katahdin) and

Boer x Kiko meat goats finished on mixed pasture of orchardgrass, red clover, and white clover with and without whole cottonseed supplementation. In all cases, daily gains, body weights and desirable carcass qualities were greater with the supplemented animals. However, all animals finished on pasture or hay alone diets were of acceptable quality for the Halal market. In another experiment, goats were finished on alfalfa, red clover, or orchardgrass pastures. All pasture types produced marketable carcasses. Research efforts have resulted in numerous speaking invitations, and findings are being applied both regionally and nationally. (Turner et al., 2006; Turner et al., 2007; Wildeus et al., 2007a,b)

**Increasing profitability of sheep production on small Appalachian farms with prairie bromegrass.** Productivity of most pasture species used in Appalachia decreases in autumn at the time when nutrient needs of grazing lambs are greatest. As a result, costly supplements are often used. ARS scientists at Beaver, WV showed that prairie bromegrass, a productive and nutritious perennial grass that grows well during cooler periods, is an excellent alternative resource for forage-based lamb finishing systems in Appalachia. They also found that using a mixture of prairiegrass and forage turnips in pastures provided high quality forage throughout the grazing season and greatly reduced the need for supplementation. Lambs finished on prairiegrass-turnip pastures nearly doubled their rate of gain over lambs finished on a traditional grass-clover pasture. (Cassida et al., 2009)

### **Pasture Management to Improve and Maintain Productivity**

**Managing seed banks to control invasive weeds.** Buried seeds in pasture soils are a seed bank for both desirable forage species and weeds. For three years, ARS scientists at University Park, PA evaluated management practices to reduce weeds in the soil seed bank by evaluating the relationship between four different above-ground-plant mixtures and below-ground seeds. Annual weed species were less abundant in the seed banks of pastures containing a greater variety of forage plant species than the pastures containing two forage-species. Therefore, pasture managers may be able to reduce annual weeds by planting more complex forage mixtures than the more traditional grass-clover systems. Follow up research explored the concept of “pasture fallowing” where the seed bank is augmented by natural reseeding. The fallowing produced more grass and weed seeds, but the seeds did not persist in the seed bank or lead to longer-term differences in pasture composition. Fallowing does not appear to be a viable strategy for pasture regeneration in the Northeast. (Sanderson et al. 2007c; Goslee et al., 2009)

**Renovating chicory pastures.** Chicory is a high yielding, drought resistance, nutritious forage, but its usefulness is limited by poor persistence. ARS scientists at University Park, PA evaluated three seeding methods, 1) frost seeding in February, 2) no-till seeding in March, or 3) broadcast seeding combined with hoof incorporation in May, to reestablish chicory into a productive cool-season pasture. Seedling emergence was adequate, averaging 5 to 6 seedlings/ft<sup>2</sup>, and did not differ among methods, but seedling mortality was high so that chicory only contributed 5% of total biomass by the fall of the establishment year. Maintaining chicory in cool-season grazing system can be successful if steps, such as close grazing, are taken to promote seedling survival. (Skinner and Dell, 2010)

**Species-rich pastures maintain nutritive value.** Limited-resource farmers who manage pastures with complex mixtures of forage species are concerned that large changes in the botanical composition of these species-rich pastures may cause unstable and lower herbage nutritive value and compromise livestock production. USDA-ARS scientists at University Park, PA measured the variability in nutritive value of pastures planted to complex (6 or 9 species) mixtures of forage species or a simple grass-legume mixture. The results showed that the number of forage species in the mixture did not control herbage nutritive value. Rather, functional group proportions (i.e., grasses, legumes, and forbs) controlled the nutritive value of

mixed-species pastures. Legume proportion controlled crude protein in forage, whereas the grass component controlled fiber levels. Farmers with highly diverse pastures can realize greater yields with lower inputs without compromising nutritive value. (Deak et al., 2007; Sanderson, 2010b)

**Forage mixtures reduce risk and improve grazing economics.** Using forage mixtures in pastures can increase forage yields but mixtures may be more difficult to manage. Farmers will only adopt mixtures with potential for greater net profit. ARS scientists at University Park, PA in cooperation with Penn State University used a computer model simulating an entire dairy farm along with 100 years of weather data to evaluate the short-term (2 years) and long-term (25 years) economics of altering forage mixtures and grazing strategies. Results demonstrated that grazing management based on forage height rather than plant morphology criteria (e.g., first bloom in alfalfa) increased net economic returns by 8% in the short-term to 18% in the long-term. Using complex mixtures of up to 7 grasses and legumes increased net return by 15% in the short-term and 32% over the long-term compared with nitrogen-fertilized grass. Forage mixtures had 30% smaller production risk. For dairy pastures, managing complex mixtures of forages is a way to reduce production variability and increase profitability. (Deak et al., 2009; Deak et al. 2010)

**Alfalfa harvest yield and quality is affected by season.** ARS scientists determined how rapidly alfalfa yield changes relative to quality in the spring, early summer, late summer, and fall in ID, PA, and WI. They found average yields were always greatest in the spring at all locations. In the eastern United States, alfalfa yield increased most rapidly in the spring while in the Midwest and West, alfalfa yield increased most rapidly in the early summer. At all locations forage quality declined most rapidly in the spring and most slowly in late summer and fall. Thus, decisions regarding the appropriate cutting times for alfalfa are more important in the spring when its highest proportion of annual yield declines more rapidly in quality. (Coblentz et al., 2008; Brink et al., 2010b)

**Endophyte in tall fescue is unnecessary in cold climates.** Tall fescue is becoming more importance for pasture and hay production systems in the northern USA. A multi-location experiment evaluated four diverse tall fescue cultivars with and without endophyte infection. Following three harsh winters, endophyte infection had no impact on tall fescue survival. These results demonstrate that tall fescue endophytes do not protect tall fescue against severe cold or freezing stresses. (Casler and van Santen, 2008)

### **Improving Pasture Management Tools**

**Choice of vegetation sampling methods depends on patchiness of pastures.** Accurate affordable pasture sampling methods are vital to evaluate pasture condition, set livestock stocking rates, and assess management impacts. Environmental variations and grazing can result in patchy patterns across a pasture. At University Park, PA, ARS scientists evaluated four vegetation sampling methods (random, transect and two multi-scale methods) to determine how variations in patchiness affect the accuracy in estimating species frequency and cover. The most used methods (random and transect) were not the most accurate with the transect method being very inaccurate in pastures with considerable patchiness. Appropriate statistical analyses must be used with these multivariate and spatially-complex sampling methods. These results are being used to develop protocols for the USDA-NRCS pastureland National Resource Inventory. (Goslee, 2006; Goslee and Urban, 2007; Goslee, 2010)

**Surrounding land use strongly affects pasture plant communities.** The soils, climate, and topography of a site determine what plant species thrive, but surrounding land uses may also influence the plant community by providing seeds or hosting predators or pathogens. ARS scientists at University Park, PA analyzed pastures across the northeastern United States and

discovered that forage species were little affected by surrounding land use or landscape pattern, but composition of non-forage species was strongly related to local land use and landscape pattern, as well as to soils and climate. Biodiversity depends not only on site characteristics and management, but on the structure of the entire landscape. (Goslee and Sanderson, 2010)

**High-resolution elevation data matters for Best Management Practice placement.** Best Management Practices (BMPs) recommend placing buffers and filter strips downslope from livestock heavy use areas, but do not explain how to do so. ARS scientists at University Park, PA with researchers from Penn State used geographic information systems to compare two methods of siting BMPs: placing them at the closest point on the stream, or using elevation maps to trace topographic effects on water movement from the heavy use area to the stream. Fine (1 m resolution), medium (10 m) and coarse (30 m) elevation maps all showed that water from heavy use areas can enter the stream a long distance from the closest point, most commonly 85 m away. Water travels 91% farther from the heavy use area to the stream than measuring the closest stream point would suggest. The medium-resolution elevation maps were most useful for planning the locations of new BMPs. Visual estimation is not sufficient to identify the best location for streambank BMPs. (Piechnik et al., 2010)

### **Pasture Management to Enhance the Environment**

**Carbon sequestration in mature pastures.** Converting annual cropland to perennial pasture may increase soil carbon sequestration but more information is needed on the carbon sequestration of land that has been in pasture for several years. Monitoring daily carbon fluxes from two central Pennsylvania fields that had been managed as perennial pastures for at least 35 years revealed that these pastures were small carbon sinks when only the physiological processes of photosynthesis and respiration were considered. However, when a complete carbon budget was calculated including the removal of harvested forage and application of manure, the pastures became net carbon sources to the atmosphere. Mature pastures in the Northeast may not help slow the increase in atmospheric carbon dioxide and may actually contribute to an increase. (Skinner, 2008b)

**Winter carbon dioxide Fluxes of Northeastern pastures.** Scientists at University Park, PA used eddy covariance to quantify the magnitude of winter fluxes for two humid-temperate pastures in the northeastern USA over a three year period. Carbon losses during winter months (1 Dec to 31 Mar), averaged 2.89 g CO<sub>2</sub> m<sup>-2</sup> d<sup>-1</sup>. Even though photosynthetic carbon dioxide uptake occurred at temperatures as low as -4 C, wintertime carbon losses were large and must be taken into account when constructing annual carbon budgets. Timing of the transition from carbon sink to source in the autumn was related to standing biomass and available solar radiation but not to temperature. Increasing temperature in future climates may not extend the carbon accumulation period in the fall. (Skinner, 2007)

**Liming marginal soils lowers risk of ground water pollution.** Pasture-based sheep and meat goat production are increasing on small farms in Appalachia on soils that are often shallow, acidic, and infertile. A few repeated urinations by goats on these soils will kill most of the vegetation and seriously pollute groundwater with nitrates. ARS scientists demonstrated that liming the soil ameliorates the nitrogen "burn" and plant scorching effects of urine's concentrated salts and organic acids and reduces nitrate-nitrogen in drainage water after two goat urine applications. (Ritchy and Ferreira, 2006)

**Composted poultry manure helps sustain hill pasture water quality.** West Virginia poultry operations annually produce more than 75,000 metric tons of litter. The sustainability of repeated applications of composted turkey litter to grazed, mixed-forage pastures was investigated by ARS scientists at Beaver WV. Biennial applications of 3 metric tons per hectare of composted

turkey litter to pastures that included chicory was found to be sustainable with no nitrate leaching. (Boyer et al., 2007.)

**Pasture type impacts groundwater quality in Appalachia.** Silvopasture management is being studied by ARS researchers in Beaver, WV to balance the seasonal distribution and persistence of high quality herbage with environmental goals. Water quality was monitored at the soil to bedrock interface under conventional pasture, silvopasture, and in hardwood forest on a central Appalachian landscape. Silvopasture significantly reduced the concentration of nitrate leaching to shallow groundwater, but macropores contributed to significant increases in fecal bacteria reaching shallow groundwater. (Boyer and Neel, 2010)

**Transport and redistribution of fecal pathogens by rain splash in forage canopies can have consequences for livestock and human health.** Transport of fecal bacteria by rain splash was studied by ARS scientists at Beaver, WV. Three forage species were maintained under short, moderate, and tall managements on various slopes to determine management and site effects on fecal bacteria movement. In order to reduce the opportunity of infection from contaminated forage, the use of broad-leaved forages should be managed taller and young or stressed animals should be kept off of the forage during rainy periods. (Boyer and Belesky, 2009.)

**Good canopy management may reduce Cryptosporidium, a pathogen organism that infects human and animal.** ARS scientists at Beaver, WV analyzed surface soil and forage samples from pastures and winter-grazed hay fields for the presence and concentrations of Cryptosporidium oocysts. More than 40 percent of soil and vegetation samples in pasture and hay fields contained Cryptosporidium oocysts. Cattle and wild animals were sources of the Cryptosporidium oocysts. Canopy management, short-cycle rotational grazing, and control of wildlife were recommended as strategies for reduction of cryptosporidium oocysts in pasture and lessening the risk of contamination of water supplies. (Boyer and Kuczynska, 2010)

## **Component 3: Harvested Forage Systems for Livestock & Bioenergy**

Bioenergy production may increase farm profits and utilize crop surpluses while conserving soil and water resources and reducing our dependence on foreign oil. However, improvements in germplasm, management practices and production systems are needed to improve economic viability and environmental sustainability of using harvested forages

**Objective H.1.** Provide the scientific knowledge, technologies and germplasm needed to develop plant materials that can be produced economically and efficiently converted to high-value products while enhancing the environment.

### **Cell Wall Structure and Abiotic Stress**

The understanding of cell wall structure and biochemistry provides crucial information for improving utilization of alfalfa and forage legumes. Altering plant developmental characteristics will provide opportunities to balance improved feed characteristics and resistance to environmental stresses.

**Alfalfa expressing polyphenol oxidase activity can decrease protein degradation in the silage and decreased the need for protein supplements in animal diets.** A major problem with ensiling is excessive protein degradation due plant proteases released during the early stages of harvest, wilting and ensiling. Red clover has high protein levels similar to alfalfa, but requires little effort to preserve protein (>85%) during ensiling because of the presence of polyphenol oxidase (PPO) and *o*-diphenols such as chlorogenic acid . Alfalfa does not naturally produce PPO or the appropriate *o*-diphenol substrates that together inhibit protein degradation by proteases. Red clover PPO genes have been identified in and used to produce alfalfa transformants expressing PPO. Extracts from PPO-alfalfa contain significant levels of PPO activity combined with chlorogenic acid results in protein degradation decreasing by 80-90%. Ensiling experiments with PPO-alfalfa and added chlorogenic acid resulted in decreased protein degradation. Chlorogenic acid is produced in many common plants including coffee, dried plums, and forages such as tall fescue and timothy. These may serve as economical sources of *o*-diphenol to be used with ensiled PPO-alfalfa to improve protein preservation and better animal performance. (Sullivan et al., 2004; Sullivan and Hatfield, 2006; Schmitz et al., 2007)

**Identification of a novel red clover hydroxycinnamoyl transferase capable of creating hydroxycinnamate malic acid esters.** Red clover accumulates large amounts of the *o*-diphenol phasalic acid, a caffeic acid ester of malic acid that is a powerful antioxidant that protects red clover protein following harvest. ARS scientists have identified a novel red clover enzyme with an activity capable of creating phaselic acid or its immediate precursor (*p*-coumaroyl malate). This enzyme may be responsible for a key step in phaselic acid biosynthesis. If the phaselic acid biosynthetic pathway could be recreated in alfalfa and other forages, the polyphenol oxidase (PPO)/*o*-diphenol protein protection system could be exploited with the potential of saving farmers \$100 million annually and preventing release of excess nitrogen into the environment. This finding also provides important basic information to researchers investigating plant secondary metabolism. (Sullivan, 2009)

**Impact of stem length on fiber digestibility in alfalfa.** Particle size of forages fed to dairy cows impacts on milk production. However, little is known about how forage cell wall lignification interacts with plant anatomy in large particles to affect forage fiber digestibility. To test if internode length could predict degradation of stem tissues, ARS scientists at St. Paul, MN selected alfalfa genotypes for rate of fiber digestibility and examined them for internode length. The rapidly digested genotypes were found to have shorter internodes whereas those genotypes with slower fiber digestion had longer internodes. These results suggest that plant breeders could select alfalfa plants that are more rapidly digestible by simply determining average length of stem internodes rather than using costly laboratory methods. (Engels and Jung, 2005).

**Alfalfa germplasm for remediation of excess nitrogen:** Alfalfa cultivars with greater capacity to remove nitrate would be useful for environmental protection and remediation, whereas alfalfa that is less competitive for nitrate may be useful in mixed plantings with grasses. ARS scientists in St. Paul, MN, developed a new, affordable field-based method to identify alfalfa germplasm that differed in capacity to take up soil nitrate. This new method was used to create alfalfa populations with either high or low amounts of nitrate uptake in the harvested hay. This approach is being used by commercial plant breeders to create new alfalfa cultivars with improved capacity to remove excess nitrate from over-fertilized fields or fertilizer spill sites to protect ground water from nitrogen contamination, or with decreased capacity for nitrate-N uptake to improve the nutrient quality of mixed species pastures for animal production systems. (Lamb et al., 2008.)

**Aluminum tolerance in alfalfa assessed with model legume:** Acidic soils and aluminum toxicity reduce yields of alfalfa. In defining genetic mechanisms regulating acclimation to aluminum toxicity in alfalfa, ARS scientists in St Paul MN used the model plant *Medicago truncatula*. They found that the root tip was the target of aluminum toxicity. After 12 hours of aluminum treatment, many genes involved in plant cell wall synthesis and modification were turned on. Genes involved in reducing the toxic effects of oxygen free radicals were also turned on. Production of these toxic compounds is due to the damaging effect of aluminum on plant cell membranes and other cellular components. Additionally, aluminum treatment turned on genes involved in plant cell death, presumably to remove cells damaged by aluminum. Understanding the molecular underpinnings of a plant's response to aluminum is key to developing new alfalfa varieties that can tolerate aluminum in acidic soil. Aluminum tolerant crops are needed to expand production in new areas and to sustain production in current crop production areas where soils are becoming acidic. (Chandran et al., 2008a,b)

### **Improving Resistance to Biotic Stress**

Productivity in alfalfa and forage legumes is often limited by the effects of biotic stressors such as fungal and bacterial pathogens, and feeding damage caused by nematodes. ARS researchers at Beltsville, MD, Prosser, WA, and St. Paul, MN have developed new methods for detecting the pathogens, and utilizing those methods for identifying and quantifying levels of resistance in existing alfalfa varieties and elite germplasm.

**Reducing brown root rot of alfalfa.** Brown root rot is a fungal disease that causes winter-kill and significantly reduces stand life and forage yield. Using a robust assay developed previously by ARS scientists (Larsen et al., 2002) that utilizes a molecular marker to rapidly detect the highly destructive pathogen, the distribution of the fungus was surveyed in Minnesota and Wisconsin alfalfa fields and in commercial wheat fields in Minnesota. The fungus was found widely distributed in both States and was associated with winter injury and winterkill of plants (Larsen et al., 2007). Because chemical control is not possible, management strategies are needed to reduce damage. Collaborative research between ARS scientists in St. Paul, MN and the University of Minnesota, found that corn and soybean residues increased pathogen populations while spring wheat, oat, and canola residues decreased pathogen populations. Multi-year field trials also identified alfalfa varieties that persist in locations with high amounts of the brown root rot fungus and have high forage yield potential. Losses from brown root rot can be reduced by rotating crops with a spring-seeded small grain crop and by planting brown root rot tolerant varieties. (Samac et al., 2008)

**Characterization of leaf spot disease in alfalfa.** In leaf spot diseases in alfalfa a high degree of genetic variability exists within species and strains that attack different crops worldwide. ARS scientists at Beltsville, MD developed methods to characterize molecular variation and the genetic structure of fungal populations that cause the leaf spot disease. This resulted in identifying several new fungal species that cause leaf spot on alfalfa. The research will help scientists, breeders, and managers identify specific species and populations causing leaf spot and lead to improved control strategies. (Liu et al. 2007; Wolf et al., 2010)

**Characterizing fungal pathogens to identify risks to alfalfa production.** Accurate tools are needed to detect and identify domestic and foreign fungal strains that can cause disease outbreaks that threaten the nation's \$8 billion alfalfa crop. ARS researchers at Beltsville, MD applied recent advances in DNA analyses to foreign and domestic collections of fungi to identify those causing disease, to determine their virulence, and to understand their pathogenicity. They discovered new species, branches in family trees and host associations that revealed strains not found in the U.S. that are potential sources of new diseases. This information will help diagnose new disease outbreaks more rapidly and develop appropriate control strategies. (Castell-Miller et al., 2008)

**Quantification of Root Rot Pathogens.** Alfalfa is subject to significant yield losses due to stand decline when infected with root rotting fungal pathogens. ARS scientists at Prosser, WA and collaborators identified DNA markers to detect pathogens and then develop a highly precise assay using fluorescent real time PCR to quantify the pathogen DNA present in infected tissue. The assay is being used to more accurately select the most resistant plants for use in breeding programs and accelerate the development of extremely resistant alfalfa cultivars (Larsen et al., 2007). Alfalfa seedlings can often be co-infected with *Phytophthora* and *Aphanomyces* but because the pathogens are so closely related, previous technologies could not distinguish the pathogens in infected plants. A real-time PCR assay for quantifying *Phytophthora* was developed along with a real-time PCR assay to simultaneously quantify alfalfa plants in mixed infections with *Phytophthora* and *Aphanomyces*. These methods provide an accurate measure of population dynamics involving multiple pathogens in a plant host. An additional multiplex real-time assay for quantification of both *Phoma medicaginis* and *Aphanomyces euteiches* has provided a more precise method for rapid identification of alfalfa plants highly resistant alfalfa plants to these pathogens than was previously possible (Vandemark and Ariss, 2007; Vandemark et al, 2010).

**Finding resistance to Verticillium wilt of alfalfa.** Inheritance of resistance to Verticillium wilt is not well understood. Scientists at Prosser, WA developed two specialized alfalfa populations and subjected them to repeated disease evaluations. They found resistance was highly heritable and controlled by additive gene action. This information will directly assist breeders to optimize strategies to most rapidly enhance levels of disease resistance. (Vandemark et al., 2006a; Vandemark et al., 2006b; Ariss and Vandemark, 2007)

**Reducing lodging in alfalfa.** Lodging resistance and spring vigor are important for maximizing biomass yield. ARS scientists at Prosser, WA identified two markers that showed significant association with lodging resistance, one of which is a candidate for use in marker-assisted selection for this trait. One unlinked marker demonstrated significant association with spring vigor. (Saha et al., 2010)

**Identification of nematodes that affect alfalfa, forages, and grasses.** Cyst nematodes are capable of damaging the roots of many different plant species including alfalfa, clover, grasses, and crops grown in rotation with alfalfa. ARS scientists in Beltsville, MD described the anatomical and molecular characteristics of a cyst nematode from soil associated with potato fields in eastern Idaho, providing the first detailed description of this nematode in the U.S. (Skantar, et al. 2007) They also developed a highly sensitive molecular method for detection and identification of these nematodes, including species that are regulated as quarantine pests in many countries. Scientists, regulators, and extension agencies will use the results of this research to more precisely identify and prevent the spread of these nematodes in the U.S. (Nakhla, et al., 2010).

**Root knot and sting nematodes are destructive to the roots of plants including alfalfa, grasses, and other forage crops.** ARS scientists at Beltsville, MD also described a wide range

of morphological and molecular variation in new occurrences of root knot and sting nematode populations. Because root-knot and sting nematodes can infect a number of economically important crops and grasses, scientists, regulators, and extension agencies will use this research to accurately identify this nematode and recommend appropriate management practices (Handoo, et al., 2009; Skantar, et al., 2008; Yan, et al., 2008).

**Objective H.2:** Develop improved grass and forage legume germplasm and varieties that can be more efficiently converted into livestock, bioenergy, and bioproducts that can be produced in a variety of environments.

### **Improved Forage Legume Germplasm**

Forage legumes are a key component for production of livestock and provide environmental services including enhancing soil fertility and protecting water quality. Current research focuses on alfalfa, the most widely grown and highest producing forage legume.

**Use of genetics and genomics to develop and improve alfalfa germplasm for bioenergy disease resistance and nutrient use.** Because alfalfa is an outcrossing tetraploid species, identifying genes controlling important traits such as cell-wall composition is exceptionally difficult. To overcome this limitation ARS scientists at St. Paul used genomics to identify genes involved in these processes. Some 50,000 genes were sequenced from the model legume *Medicago truncatula*. Methods were developed to utilize the Medicago GeneChip for measuring gene expression in stems of alfalfa plants selected for cell wall polymers important in efficient conversion of lignocellulosic biomass to ethanol. Numerous genes that regulate polymer biosynthesis in alfalfa were identified for the first time. A unique computer algorithm was developed to improve identification of expressed genes when using this GeneChip for analyzing alfalfa. Genes that contribute to disease resistance, forage quality, and nutrient use were identified through gene sequencing and GeneChip studies. DNA markers were identified that can be used to accelerate alfalfa breeding for improving disease resistance, environmental stress tolerance, animal nutrition, and bioenergy. Alfalfa geneticists and agronomist throughout the world are using the Medicago gene sequences identified and the algorithms developed by ARS scientists. (Tesfaye et al., 2006; Foster-Hartnett et al., 2007; Schnurr et al., 2007; Ameline-Torregrosa et al., 2008; Tesfaye et al., 2009; Yang et al., 2009; Yang et al., 2010)

### **For Improved Grass Germplasm and Varieties**

Grasses, including corn stover, have high biomass potential and many can be grown in a range of soil types. However, many species have not been assessed for biofuel characteristics and are unimproved for cultivation.

**The sfe corn mutant reduces ferulate cross linking, improves fiber digestibility, and increases milk production.** Corn silage is a major feedstuff for dairy production, but its dietary fiber fraction is poorly digested. Previous ARS research indicated that if cross linking of fiber polysaccharides to lignin (an inhibitor of fiber digestion) by ferulate molecules could be reduced, fiber digestibility should be increased. A seedling ferulate ester (sfe) mutant corn line with 50% reduction in seedling ferulate concentration was identified by ARS scientists at St. Paul and Madison in collaboration with the U. of Minnesota. A two-year field trial demonstrated that sfe lines had increased in vitro (laboratory) digestibility of silage fiber compared to the parental corn line. When fed to lambs, sfe corn silage resulted in less selectivity in what the lambs ate and the lambs digested more fiber. Inclusion of the sfe corn silage in a complex mixed diet for dairy cows resulted in greater feed intake and increased milk production. Research is underway to identify the mutated gene. Because all grasses contain these ferulate cross-links, the sfe gene may allow improvement of fiber digestibility of all grasses fed to livestock. (Casler et al., 2008; Jung and Phillips, 2010)

**Potential for improving corn stover quality to use in cellulosic ethanol production.** In collaboration with a University of Minnesota scientist, ARS scientists at St. Paul examined the correlations among grain yield, other agronomic traits, and cellulosic ethanol traits. All the cellulosic ethanol traits (cellulose, lignin, and glucose release) had moderate to high heritability and were not negatively correlated with grain yield or other agronomic traits. Quantitative trait loci (QTL) were found for all cell wall traits, but these QTL were small in magnitude. The results indicate that it should be possible to breed corn for improved stover cellulosic ethanol traits while continuing to select for increased grain yield. Such simultaneous improvements in corn yield and stover quality will allow significant increases in overall ethanol yield and net energy efficiency of biofuel production using corn. (Lewis et al., 2010; Lorenzana et al., 2010)

**Leadership in switchgrass improvement.** Switchgrass breeding and genetics research by ARS scientists at Lincoln and Madison forms the foundation for all switchgrass breeding and genetics research now underway worldwide. This research provided the basic genetic information for using self-incompatibility for producing switchgrass hybrid cultivars and hybrid cultivars of other grasses to improve yield over the past decade. (Casler et al., 2002, 2004; Martinez-Reyna et al., 2001, 2002; Tobias et al., 2005) In collaboration with the University of Georgia, the Great Lakes Bioenergy Research Center, the Bioenergy Sciences Center, and China Agricultural University, ARS scientists identified five distinct lineages of switchgrass: Gulf Coast lowland, Southern Plains lowland, Southern Plains upland, Central Plains upland, and Eastern upland. All populations are derived from three glacial refugia that served as tallgrass prairie reserves during the Pleistocene. This information is useful for conservation of the tallgrass prairie and oak-savanna or pine-barrens ecosystems as well as in switchgrass genetics and breeding. (Casler et al., 2007; Zalapa et al., 2010) Based on foundation and certified seed records, the switchgrass and intermediate wheatgrass cultivars developed by ARS scientists at Lincoln, NE and their collaborators are the most widely utilized cultivars of these species in the Great Plains and Midwest over the last two decades. The seeded acreage of the switchgrass and wheatgrass cultivars is estimated at one million acres and their use conservatively adds over \$20 million annually to the economy. Also seed demand for the two big bluestems, Goldmine and Bonanza, has exceeded supply since they were released (Vogel et al 2005a,b; Vogel et al., 2006a,b) ARS scientists at Raleigh, NC collaborating with North Carolina State University have been improving and evaluating switchgrass as a multipurpose crop for the Southeast that can be graze, harvested as hay or silage, or used as biofuel feedstock. Selecting switchgrass germplasm using criteria of yield potential, in vitro dry matter disappearance, and crude protein in a weighted index has resulted in the release of three cultivars, 'BoMaster' for yield, 'Colony' for improved cellulose concentration and yield, and 'Performer' for improved forage quality. Releases of the 3 cultivars have been made and seed will be soon available commercially. (Burns et al., 2008a, 2008b, 2010).

**When cool-season grasses are the better option.** ARS scientists at Logan compared the biomass potential of several warm-season perennial grass species, including switchgrass, to that of common cool-season perennial grasses grown under irrigated conditions of the Intermountain U.S. Although the warm-season grasses established as well as the cool-season grasses, they were more damaged coming out of winter and produced significantly less biomass than did the cool-season grass species across the entire growing season. Bioenergy production in these areas should focus on cool-season grasses or species other than warm-season grasses. (Robins et al., 2009; Robins, 2010)

**Modifying smooth brome grass to alter the linkage between forage yield and fiber concentration.** Profitability of livestock production is increased when forages have high yield per acre and low non-digestible fiber content. Low fiber content increases forage consumption and digestible energy content, resulting in improved animal performance. Unfortunately, some key forage grasses have a link between higher yield and higher fiber concentration, so opportunities to breed higher yielding, lower fiber varieties have appeared to be limited. ARS scientists at Madison found that breeding to improve smooth brome grass, an important

harvested forage, resulted in a 1% decrease in non-digestible fiber but was accompanied by a 5% decrease in yield. However, from studying hybrids of smooth brome grass they have been able to partially break this association and identified a few genes that improve yield but are not linked to fiber content. Based on this knowledge, work is proceeding to develop forages with both increased yield and lower fiber to increase livestock performance. (Casler et al., 2005.)

**Plant Adaptation Regions concept for plant classification.** ARS scientists at Lincoln, NE developed new research tools that are being widely used in research and management. Developed the Plant Adaptation Region concept to provide an ecological and climatic method of classifying plant germplasm for geographic regions (Rangeland Ecol. & Mgmt. 58:315-319). The Plant Adaptation Region concept, although recently published, has already been validated as a mechanism for defining adaptation regions for grasses including switchgrass and has been used in Pecan and urban ecological restoration research. It was described in a recent review (Current Opinion in Biotech. 19:202-209, 2008) as an outstanding accomplishment. Developed the frequency grid for the rapid and reliable quantification of plant stands in swards. Based on 40 citations, the Frequency Grid procedure is currently being used at over 25 research stations in the USA. (Vogel and Masters, 2001; Vogel et al., 2005c, Schmer et al., 2006)

**Napiergrass for biomass production.** Napiergrass is a perennial grass used for forage and has considerable potential as a biofuel feedstock for the Southeast primarily because of its high biomass yield. Researchers at Tifton, GE assessed the genetic variation and genetic relatedness among 89 accessions. The accessions clustered into five groups including three groups from Kenya, a group from Puerto Rico, and accessions derived from the cultivar Merkeron. This research was the first molecular characterization of the Tifton accessions, which represent 30 years of plant collection and breeding. Furthermore, this work provides potential parents for napiergrass and pearl millet breeding improvement that is currently underway in collaboration with university personnel at Florida and with the bioenergy industry. (Harris et al., 2009)

## **Problem Statement J**

Need for economically viable, energy efficient and environmentally enhancing production systems for establishing, growing, maintaining, harvesting, treating, storing and transporting forages for livestock, bioenergy, bioproducts and conservation objectives.

**Objective J.1:** Provide improved management practices that enhance the environment and increase the economic viability of growing, harvesting, and storing grasses and legumes for bioenergy and byproduct systems.

## **Efficiency of Production Systems**

**Assessment of growing switchgrass as a bioenergy crop:** ARS scientists and their cooperators have focused on sustainable production and economic feasibility in growing switchgrass as a bioenergy crop in northern regions of the United States. To determine the economic feasibility of producing switchgrass as bioenergy crop in the eastern Great Plains on the farm ARS scientists at Lincoln, NE and Mandan, ND in cooperation with the University of Nebraska managed and assessed switchgrass production for five years on 10 farms spread across Nebraska and the Dakotas. Average yield was 3.4 tons/acre with average production cost of \$33/ton plus \$17/ton for land rent. Pro-rating the establishment costs over 9 years would reduce costs by \$6/ton. Two farmers experienced in switchgrass production were able to produce the biomass for less than \$40/ton including land costs. At this cost, the farm-gate feedstock cost per gallon of ethanol produced would be about \$0.50 per gallon (Perrin et al., 2008). Biomass yields and farmer inputs for the ten farms were used to determine the net energy balance of switchgrass as a biomass energy crops. All previous estimates have been based on small plot data or estimates. Net energy averaged 60 GJ ha<sup>-1</sup> y<sup>-1</sup>. Switchgrass

produced 540% more renewable energy than nonrenewable energy consumed. Farms on which the largest biomass yields were obtained also had the greatest net energy values. This large scale study clearly demonstrated that perennial herbaceous energy crops are net energy positive. Improvements in genetics and management will enhance both total and net energy yields. (Schmer et al., 2008) The effectiveness of indirect methods for estimating biomass yields and composition of switchgrass fields were evaluated. Visual obstruction was the best method for estimating yield on switchgrass fields with low to variable stand densities while elongated leaf height measurements would be recommended on switchgrass fields with high, uniform stand densities. Twenty to 30 elongated leaf height measurements in a field could predict switchgrass biomass yield within 10% with 95% confidence. (Schmer et al., 2010 a,b). (This research has received worldwide attention with one publication (Schmer et al., 2008) being downloaded 31,000 times and an online economics study (Perrin et al., 2008) downloaded over 300 times.) Related to optimal harvest times, ARS scientists at University Park, PA in collaboration with ARS scientists at Wyndmoor, PA, Madison, WI, and St. Paul, MN compared fall and spring harvested switchgrass yield and biofuel quality. Switchgrass yield decreased, but the biomass was much drier and nutrients lower in the spring. These findings have been used by policy makers (Pennsylvania State Senate), land managers and farmers (Ernst Biomass LLC has broken ground for the construction of a switchgrass pellet facility in Meadville PA), and agency personnel (USDA-NRCS Pocono Northeast RC&D Council has constructed a mobile switchgrass pellet system) to schedule switchgrass harvest time on dedicated biomass energy fields and conservation lands to optimize feedstock yield and quality for pelleted fuels. (Adler et al., 2006).

**Assessment of feedstocks for bioenergy conversion efficiency.** Feedstock species, management practices, environmental growing conditions and harvest dates can all affect conversion into various energy products so a fast and accurate method of determining feedstock quality is needed. Research by ARS researchers at Lincoln, NE, Peoria, IL, Wyndmoor, PA and their collaborators on the effect of plant maturity on the conversion efficiency were the first to clearly demonstrate that management practice based on specific maturities at harvest can affect conversion efficiency. As perennial grasses mature, lignin concentration increases and affects the yield of liquid fuels depending on method used and pre-treatments. Increased lignin concentration in general adversely affects ethanol yield from saccharification and fermentation conversion processes but can result in increased biocrude yields from pyrolysis. Production and conversion research used throughout the USA, Western Europe (Rothamsted Research, UK; Wageningen Univ., Univ. of Bologna) and in China are now based on these results. (Boateng et al., 2006, Dien et al., 2006, Numerous papers summarized in monograph chapter, Vogel 2004). A team of ARS scientists from Lincoln, NE, Peoria, IL, St. Paul, MN, and Madison, WI developed NIRS calibrations that, along with biomass yield data, enable the following switchgrass bioenergy traits to be rapidly and accurately determined: theoretical ethanol yield from hexose and pentose sugars, total actual and theoretical ethanol yield per Mg (ton), acre or ton from pre-treated biomass, and conversion ratio of actual to theoretical on a liter to liter basis. These calibrations are useful in breeding, genetics, and agronomic research and will aid biorefineries to estimating ethanol yield of switchgrass inputs. (Vogel et al., 2010c)

**Potential biomass production from Conservation Reserve Program (CRP) land.** Marginal croplands may have potential as a biomass source of bioenergy but there is limited information on the species composition on these lands and their suitability for ethanol production. Researchers at University Park, PA found that northeastern US. CRP grasslands with the highest number of species had the lowest potential ethanol yields per acre, however, sites dominated by a small number of native tall prairie grass species, such as switchgrass, big bluestem, and indiagrass, had the highest yields. The results from this study demonstrated that the species composition of plant mixtures used in low-input, high-diversity systems affects both biomass production and chemical composition of the resulting feedstock and that including a large number of species with undesirable fermentation characteristics could reduce ethanol yields. These findings have been used by NGOs (Chesapeake Bay Commission) to quantify the

biomass feedstock yield potential of abandoned farmlands in the Eastern US (Adler et al. 2009). ARS scientists at El Reno, OK, showed that Old World bluestem produced an average of 3380 lbs/acre, and a native mix produced 1710 lbs/acre of dry biomass feedstock on Oklahoma CRP land evaluated across all years, locations, and harvest dates. Maximum yields were in October for both Old World bluestem (3720 lbs/acre) and the native mixed species (1950 lbs/acre). At the native mixed species sites, three years of annual harvest did not alter species composition or soil characteristics, but biomass production consistently declined at all sites and for all harvest dates over the period. Maintaining sustainable production requires some form of nutrient replacement such as chemical fertilizers, manure, or conversion-process byproducts. Adding a legume component to the grasslands could also help. (Venuto and Daniel, 2010).

## **Sustainability of Biofeedstock Production**

**Assessment of potential adverse effects of corn stover removal on sustainability in comparison with switchgrass.** Many have assumed that crop residues such as corn stover are an abundant and inexpensive source of biomass that can be removed from fields to produce bioenergy and this resulted in less research emphasis on perennial biomass energy crops. Increased emphasis on crop residue was based on the assumption that with minimum or no-tillage farming methods, there will be no deleterious production or environmental effects. A long-term field study by ARS scientists from Lincoln, NE compared carbon sequestration of switchgrass managed as a biomass energy crop with non-irrigation, no-till corn production to evaluate the effects of stover harvested for biomass energy. In the first five years of the study, removal of half the stover significantly reduced corn yields. During the same time period, the potential ethanol yield for switchgrass was equal or greater than the potential total ethanol yield of corn grain and harvested stover. As a result of this and other research, DOE and USDA now recognize that sustainability is a major issue in the use of crop residues for biomass energy. New research initiatives on stover removal and sustainability are in progress. (Varvel et al., 2008)

### **Switchgrass grown for biomass energy results in significant soil C sequestration.**

Research by ARS scientists at Lincoln, NE, Ft Collins, CO, Mandan, ND, and University Park, PA documented carbon sequestration by growing switchgrass. The scientists at Lincoln, NE and Ft. Collins, CO conducted a long-term study that included two switchgrass cultivars, three N fertilizer rates and two harvest treatments. In the 9 year period from the spring of 1998 to the spring of 2007, soil C increased at rate of 2 Mg per hectare (0.9 U.S. tons/acre) per year in plots in which best management practices were used. Biomass yields and C sequestration was significantly greater in plots using N fertilizer. These results fully support switchgrass soil carbon sequestration data obtained previously in a five-year study on ten farms in NE, SD, and ND. The 10-farm study found that changes in available soil P were very low when used for switchgrass production (Follett et al., manuscript in preparation). Across these same ten sites, soil organic carbon (SOC) increased significantly at 30 cm and 120 cm, with accrual rates of 1.1 and 2.9 Mg C ha<sup>-1</sup> yr<sup>-1</sup>, respectively, demonstrating switchgrass's potential as a carbon negative bioenergy crop. The amount of sequestered soil carbon exceeded the levels used in previous switchgrass lifecycle assessments (LCAs) for greenhouse gases. However, change in SOC across sites varied considerably, ranging from 0.6 to 4.3 Mg C ha<sup>-1</sup> yr<sup>-1</sup> for the 30 cm depth. (Liebig et al., 2008). ARS scientists at University Park monitored biomass production and carbon dioxide fluxes over the first four years after switchgrass establishment. Averaged over the first four years of production, a net sink of 142 g CO<sub>2</sub> m<sup>-2</sup> yr<sup>-1</sup> (39 g C m<sup>-2</sup> yr<sup>-1</sup>) was observed. Photosynthetic C uptake, ecosystem respiration and evapotranspiration were all lower than results commonly observed in the Midwest, primarily due to lower growing-season temperature and lower available solar radiation (Skinner and Adler, 2010). Scientists at University Park, PA evaluated three switchgrass varieties under clipping and grazing management for seven years at two PA locations. Of the two varieties that persisted annual variation in weather and harvest management had larger effects on yield and nutritive value than between the switchgrass

cultivars. Soil carbon levels taken at 30 cm did not change after five years of growing and grazing switchgrass, but was 33% higher at 4.5 cm after seven years under clipping management to simulate harvesting for hay or bioenergy feedstocks. Data collected at multiple locations indicated a large variation in sequestered carbon and this needs to be considered in life cycle analyses and carbon trading policies. (Sanderson, 2008).

**Potential Impact of bioenergy crops on greenhouse gas emissions.** Bioenergy cropping systems have the potential to offset a portion of greenhouse emissions but quantifying the offsets is difficult because so many factors are involved. ARS scientists at University Park, PA and Ft. Collins, CO working with Colorado State University conducted life cycle analysis of net greenhouse gas emissions for several bioenergy cropping systems. This work has had major impacts on policy makers such as White House Office of Science and Technology Policy, U.S. Department of State, United States Senate, Pennsylvania State Senate, California Air Resources Board, and US EPA Renewable Fuels Standards. The life cycle analysis for bioenergy crops went from establishing and growing the crop through harvesting and conversion to utilization of the biofuel. Compared to fossil fuels, ethanol and biodiesel from corn rotations reduced gas emissions by 35-40%; reed canarygrass reduced emissions by 85%; and switchgrass and hybrid poplar reduced emissions by more than 115%. These findings have also been used by NGOs (Environmental Defense Fund, Council on Sustainable Biomass Production, Farm Foundation), and private companies (Monsanto, as part of a consortium with John Deere and Archer Daniels Midland Company) to select between alternative bioenergy production systems to help achieve national objectives including reducing gas emissions while increasing domestic energy supplies. (Adler et al. 2007).

### **Agronomic Practices to Improve Biomass Production**

**Improvements on feedstock crop establishment using herbicides.** Selected herbicides were tested on stand establishment and subsequent yields of adapted upland switchgrass cultivars in Nebraska, South Dakota, and North Dakota as well as lowland ecotypes in Nebraska by ARS scientists at Lincoln, NE and Mandan, ND. Applying quinclorac plus atrazine resulted in acceptable stands and high yields at all locations for all ecotypes. Quinclorac, which provides effective control of grassy weeds, and atrazine, which provides good broadleaf weed control, is an excellent combination for establishing switchgrass in the mid-continental USA. With good management, switchgrass can produce yields equivalent to half of full production in the establishment year and can be at full production the second year following planting. (Mitchell et al., 2010).

**Biomass alfalfa as a means of increasing biomass yields with fixed nitrogen.** Utilizing nitrogen fixing legumes in a bioenergy crop system could provide nitrogen for grass or tree growth plus additional bioenergy feedstocks. ARS researchers in St, Paul MN developed an alfalfa biomass germplasm and crop management scheme for bioenergy production. Alfalfa forage is separated into a leaf fraction and sold separately as livestock feed, while stems are used to produce liquid fuel (ethanol). A modified management regime with a reduction in the number of plants seeded per field and delayed, less frequent cutting schedule was effective for an alfalfa bioenergy production system to maximize both leaf and stem yield. The biomass-type alfalfa germplasm and management system had comparable leaf protein yields and 37% higher stem sugar yields and nearly double the potential ethanol production (99% increase) compared to the hay-type alfalfas under the hay management system. This research demonstrates that alfalfa can be a significant contributor to America's renewable energy future. (Lamb et al., 2007; Rock et al., 2009)

**Assessment of warm-season grasses under limited inputs for Southeast United States.** The first of two studies initiated in fall 2005 by ARS researchers at Tifton, GA, assessed the performance of perennial warm-season grasses under rainfed conditions with no fertilizer inputs.

Dry matter (DM) yield was highest in the second year for all species. When averaged over three years, the yields of energy cane, and napiergrass were significantly higher than switchgrass, but switchgrass had higher carbon to nitrogen uptake. (Knoll et al., 2010).

## Component 4: Turf Plant Materials

There is a need to develop and transfer technologies, germplasm, management practices and strategies to increase the sustainability of turf systems to meet economic, environmental and social objectives.

### Problem Statement K

Need for improved germplasm that is adapted to biotic and abiotic stresses and meets the economic and environmental objectives of turf producers and users.

**Objective K.1.** Identify, characterize, and develop grass germplasm suitable for turf uses with improved water-use efficiency, drought tolerance, and salt tolerance using conventional and molecular methods to improve the germplasm enhancement.

### Plant Genetics, Improvement and Evaluation

**Markers to improve St. Augustinegrass.** Because of the heterogeneous nature of St. Augustinegrass (*Stenotaphrum secundatum*), purported hybrids cannot be identified by their appearance. ARS scientists at College Station, TX in cooperation with Texas A&M University scientists used EST-SSR markers to identify and confirm interploid hybrids from crosses between polyploid and diploid cultivars (Genovesi et al., 2009). These markers were also used to assess genetic diversity among the cultivars. These findings are important in the breeding of cultivars with improved pest resistance and turf traits.

**Markers to improve zoysiagrass.** ARS scientists at College Station, TX in cooperation with Texas A&M University scientists constructed a genetic linkage map of zoysiagrass (*Zoysia matrella*) and identified the first molecular markers linked to fall armyworm resistance in turfgrasses (Jessup et al., 2011). The locus imparted high levels of resistance and was distinct from fall armyworm minor-effect loci in maize, indicating its potential as a novel source for host-plant resistance to fall armyworm in grasses.

**Genetic linkage map for the ryegrasses.** ARS scientists at Corvallis, OR working with scientists from Oregon State University, the U. of Wisconsin, Seed Research of Oregon, and the Noble Foundation developed a detailed genetic linkage map for ryegrass that confirmed close genetic relationship to Triticeae (wheat and barley). The linkage map was constructed from a cross between annual ryegrass and perennial ryegrass populations and populated with RAPD, RFLP, AFLP, SSR, and isozyme markers that showed seven linkage groups containing two major QTLs for vernalization and photoperiod requirement mapping on the same chromosomes as in Triticeae. Use of specific molecular markers to supplement the seedling root fluorescence (SRF) test was faster and more accurate at predicting ryegrass growth types than the grow-out test alone and improved marketing potential of high turf quality perennial ryegrass. Finding these linkages provides more genetic information for grasses grown as food crops as well as grasses for turf and forage. (Warnke et al., 2004; Brown et al., 2005)

**Genetic improvement of creeping bentgrass for improved biotic and abiotic stress resistance.** Creeping bentgrass (*Agrostis stolonifera* L.) is the premier turfgrass species used on golf course putting greens, fairways, and tees. One of the difficulties in determining the progenitors of the cultivated bentgrasses is the lack of accurate ploidy determinations for many of the *Agrostis* accessions maintained in the National Plant Germplasm System (NPGS). ARS scientists working in Beltsville, MD analyzed 75 *Agrostis* accessions representing 15 distinct species along with two *Apera* and four *Polypogon* accessions using flow cytometry and 1,309 DNA markers. Cluster analysis clearly separated the common turf-type *Agrostis* species into distinct groups. The inclusion of previously understudied species within these groups offers insights into the genomic origins of creeping bentgrass that will be useful in future breeding

efforts. In addition, the data suggest a narrowing of the genetic diversity within cultivated creeping bentgrasses. (Amundsen et al. 2010a and b)

**Improved disease resistance of Creeping Bentgrass.** The turfgrass diseases dollar spot and brown patch caused by the fungi *Sclerotinia homoeocarpa* and *Rhizoctonia solani* are the most widespread fungal diseases of highly managed turfgrass species such as Creeping bentgrass (*A. stolonifera*). More money is spent to manage these diseases than all other turfgrass diseases combined. Dollar spot and brown patch are widespread throughout the summer months in warm humid areas of the United States, and spray programs are generally scheduled at regular intervals to safeguard against disease establishment. However, these spray strategies can lead to the fungi developing fungicide resistance and this has become a major issue in certain regions of the county. ARS researchers at Beltsville, MD and Rutgers University are using new high throughput DNA sequencing technologies to determine the genes that are important in the interaction between turfgrass plants and their fungal pathogens. (Rotter et al. 2007b)

**Improved selection for stress resistance in Bentgrass.** ARS scientists collaborating with Rutgers University created genetic linkage maps from two experimental mapping populations to identify the location of genes affecting dollar spot resistance. The mapping populations were developed by crossing a dollar spot resistant creeping bentgrass plant to a susceptible creeping bentgrass plant. AFLP and SSR based genetic markers were then used to develop a linkage map and determine the location of genome regions influencing dollar spot resistance. A second experimental mapping population was developed at Rutgers using a cross between resistant colonial bentgrass and susceptible creeping bentgrass. AFLP markers were again used to develop a linkage map of genomic location of resistance genes. Using genetic markers for the selection of dollar spot resistance could dramatically speed up the development of dollar spot resistant creeping bentgrass and lead to a significant reduction in golf course fungicide applications. (Rotter et al. 2007a)

**Development of genetic tool for turf grass breeding.** ARS researchers at Tifton, GA developed 21 simple sequence repeat (SSR) markers specific to centipedegrass and used them to evaluate the centipedegrass collection for genetic diversity and for sting nematode resistance screening. Because many centipedegrass accessions are phenotypically very similar, the genetic diversity information is vital as the crossing of a resistant line (ex. sting nematode tolerance) to a more diverse line is needed for marker-assisted selection. For bermudagrass, a genetic map was created with the addition of 53 markers for desirable turf traits (bright green color, lack of seed head development, deep root systems) as well as sting nematode tolerance. These markers are currently being used in our lab to identify bermudagrass cultivars for golf course superintendents, sod farm businesses, and university researchers (University of Georgia). Harris et al., 2010)

**Biotechnology risk assessment of transgenic Kentucky bluegrass using the Round-Up (glyphosate) Ready trait as a selectable marker.** ARS scientists at Logan, UT with Utah State University characterized genome relationships, cross-hybridization, and gene flow within and among native, cultivated, and weedy *Poa* bluegrasses with particular reference to the polyploid Kentucky bluegrass complex (*P. pratensis*). Studies demonstrated evidence for existence of at least four distinct sub-genomes (Larson et al., 2001; Patterson et al., 2005) within *P. pratensis*, which are shared with 15 other *Poa* species but different from six other *Poa* species. Multi-year field research measured pollen-mediated gene flow in using ESPS-synthetase transgenes as selectable markers for glyphosate tolerance in *P. pratensis* (Johnson et al., 2006). Results indicated that *P. pratensis* shares genomes and hybridizes with native *Poa* species but not with weedy species. Kentucky bluegrass is an important turf grass world-wide with other important uses. Round-up ready traits have seen rapid adoption in other crops following approval. Regulatory approval of Round-Up Ready Kentucky bluegrass has large potential impact. These findings of genome relationships and inter-specific hybridization potential has identified

hybridization potential between transgenic Kentucky bluegrass, native *Poa* species, and weedy *Poa* species provide information useful in the regulatory process.

**Identification of peroxidase genes and proteins underpinning plant tolerance to insect herbivory.** Discovering specific peroxidase genes and proteins associated with plant response to insect feeding can help select for plants with improved resistance. Using polymerase chain reaction (PCR) strategies, some genotypic differences and relatedness of buffalograss peroxidases were documented. Additional research established an active role for specific peroxidases in the resistance response of turf buffalograsses to a major insect pest, chinch bugs. These results will help breed more resistant buffalograsses while the research method has potential application to other warm-season turf and bioenergy grasses. (Gulsen et al. 2007; Gulsen et al. 2010).

**Selecting best turfgrass varieties for different climates.** The National Turfgrass Evaluation Program (NTEP) housed in ARS facilities at Beltsville, Maryland, conducts a national program that evaluated the performance of 20 turfgrass species in trials conducted in forty states. While this is an evaluation project conducted mainly by university personnel it provides valuable information to those scientists seeking opportunities to improve plant varieties and the management of turf systems.

**Increasing access to genetic variation for plant improvement program.** Assembling diverse germplasm pools provides an improved genetic base for selection, manipulation, and genetic advancement of desirable characteristics. Scientists at Logan, UT traveled to remote parts of Russia during 2007, 2008, and 2010 to collect seed of grasses that hold potential for reduced-input turf. These collections are the basis of new grass cultivars with tolerance to drought, low fertility, low temperatures, mowing, and other challenging conditions. These grasses also hold potential for biotechnology applications to provide genes to improve insect and disease resistance in important crop species. Incorporating these collections into the National Plant Germplasm System will ensure preservation and access to these unique species.

### **Natural Products Associated with Turf Grasses**

**Naturally occurring bioherbicide discovered.** Scientists at ARS Corvallis, OR and Oregon State University discovered a naturally occurring bioherbicide produced by soil bacteria. Fluid from cultures of the bacterium arrested the germination of annual bluegrass, jointed goatgrass, and other grassy weeds that reduce the value of grass seed crops and the utility of recreational and professional turfs. Commercially producing sufficient quantities of herbicide from this soil bacteria is dependent on developing a chemical approach to synthesizing the herbicide, or culturing the bacteria to produce large quantities of the product. The team identified the genetic regions regulating the production of the herbicide by soil bacteria. With this information sufficient quantities of the herbicide can be isolated and the chemical structure identified. When the chemical structure is known, manufacturing processes can be developed to manufacture a new bioherbicide for weeds in grass and cereal crops. (Banowetz et al., 2008; Banowetz et al., 2009; Kimbrel et al., 2010)

**Discovered that GAF inhibits the growth of a plant pathogen.** The bioherbicide that arrests the germination of grassy weeds (GAF) also was found to inhibit the growth of a bacterium that causes fire blight in apples and pears. ARS scientists in Corvallis Oregon, along with cooperators from Oregon State University placed small amounts of GAF on plates containing cultures of the fire blight bacterium and found that GAF prevented growth of the bacterium. This discovery may provide a means to develop a biological approach to controlling

this disease in apples and pears. The biological control approach has the potential to reduce the amount of pesticide used for disease control in these food crops. (Armstrong et al., 2009)

## **Soils and Water Management**

**BMPs for management.** Most urban playing fields and parks are constructed based on best management practices (BMPs) developed for the construction industry, not the turf industry. This results in a disconnect in standards between building and operating parks and fields. ARS scientists at Beaver, WV have recently begun research in with Virginia Tech and the Raleigh County Solid Waste Authority to develop turf based BMPs. Basic research on the construction process and the maintenance program is now being carried out and reported. (Ibrahim et al., 2010a,b; Morris et al., 2009)

**Soils modeling tools for turf management.** Scientists at Beaver, WV have begun work on the response of turf and pasture grass root systems to soil compaction, acid (low pH), toxic (heavy metals or excess fertilizer), excess or insufficient water, and variable quality soils. They are gathering data and are developing model frameworks that will lead to decision-support tools on how to manipulate plant root systems to achieve optimal adjustment to variations in soils and environmental conditions. (Zobel, R. and Waisel, Y. 2010; Zobel, 2010.)

**Salt tolerance in Kentucky bluegrass.** Competition for limited water resources make it likely that turf irrigation will need to rely on lower quality water in the future. ARS scientists at Riverside, CA with collaborators from Rutgers University evaluated six cultivars of Kentucky bluegrass for salt tolerance based on cumulative biomass production, growth rates, leaf chloride concentration, and remote sensing canopy reflectance. Cultivars Baron, Brilliant, and Eagleton were rated more salt tolerant than Cabernet, Midnight and AO1-856.

## Appendix 1: Overview of ARS

### Overview of the ARS Mission, Structure and Research Planning

**The Role of the ARS.** The Agricultural Research Service (ARS) is the principal **in-house research agency** of the U.S. Department of Agriculture (USDA). It is one of the four component agencies of the Research, Education, and Economics (REE) mission area. Congress first authorized Federally-supported agricultural research in the Organic Act of 1862, which established what is now the USDA. That statute directed the Commissioner of Agriculture "... To acquire and preserve in his Department all information he can obtain by means of books and correspondence, and by practical and scientific experiments,..." The scope of USDA's agricultural research programs has been expanded and extended many times since the Department was first created.

**Interaction with National Institute of Food and Agriculture (NIFA).** NIFA is also in the REE mission area and has responsibility for advancing scientific knowledge to help sustain the U.S. agricultural sector through a research, education and outreach system **external to USDA** by working with eligible partner institutions and organizations. The interrelated and complementary programs of ARS and NIFA requires these agencies to be visionary and forward thinking in effectively utilizing limited public resources through effective collaborative partnerships. Therefore, the two agencies regularly seek opportunities to increase the efficiencies of cooperative strategies and activities involving partnerships with the land-grant colleges and universities, other non-land-grant universities, Federal and State agencies and customers.

**ARS size and budget.** Today, ARS's workforce is approximately 9,000 employees including 2,000 scientists representing a wide range of disciplines. ARS has approximately 900 research projects spread over 100 locations across the country and at 4 overseas laboratories. The National Agricultural Library and the National Arboretum are also part of ARS. The annual budget is just over one billion dollars.

**Mission objectives entering the 21<sup>st</sup> Century.** ARS conducts research to develop and transfer solutions to agricultural problems of high national priority and provides information access and dissemination to

- Ensure high-quality, safe food and other agricultural products,
- Assess the nutritional needs of Americans,
- Sustain a competitive agricultural economy,
- Enhance the natural resource base and the environment, and
- Provide economic opportunities for rural citizens, communities, and society as a whole.

**A problem-solving organization.** To achieve these broad objectives, ARS identifies critical problems affecting American agriculture; develops strategies to mobilize resources (both human and financial); writes and reviews research plans to solve these problems efficiently; performs multi-disciplinary research; and reports the results to the customers. Each step of the process involves communicating and interacting with the scientific community, customers, stakeholders, partners, and beneficiaries to ensure program relevancy, quality and impact.

**National programs.** ARS research currently organized into 18 National Programs. These programs are managed by the Office of National Programs (ONP) through 30 National Program Leaders (NPLs) to bring coordination, communication and empowerment to the more than 1200 research projects carried out by ARS. The National Programs focus on ensuring the relevance, impact, and quality of ARS research. The national programs as currently structured are:

### **Animal Production & Protection**

Food Animal Production  
Animal Health  
Veterinary, Medical, and Urban Entomology  
Aquaculture

### **Natural Resources & Sustainable Agricultural Systems**

Water Availability & Watershed Management  
Climate Change, Soils and Emissions

### **Pasture, Forage & Rangeland Systems**

Agricultural & Industrial Byproducts  
Agricultural System Competitiveness & Sustainability  
Bioenergy

### **Crop Production & Protection**

Plant, Microbial & Insect Germplasm Conservation & Development  
Plant Diseases  
Crop Protection & Quarantine  
Crop Production  
Methyl Bromide Alternatives

### **Nutrition and Food Safety**

Human Nutrition  
Food Safety (animal & plant products)  
New Uses, Quality & Marketability of Plant & Animal Products

**Teamwork between national programs.** Ideally, the entire ARS research portfolio would go through an integrated programming cycle, but this is not possible because of diversity, size and geographical distribution of agency's activities. The national program structure is an administrative construct dividing a complex research program into manageable parts. There are clearly overlaps between national programs and this is addressed by managing each national program with a National Program Team (NPT) of NPLs that provide the appropriate mix of skills and experience.

## Appendix 2: Overview of National Program 215

### **Overview: Pasture, Forage and Rangeland Systems National Program (NP215)**

**National Program Team.** The NP215 NPT consists of NPLs from Food Animal Production, Water Availability & Watershed Management; Climate Change, Soils and Emissions; Agricultural & Industrial Byproducts; Agricultural System Competitiveness & Sustainability; Bioenergy; Crop Protection & Quarantine; Plant, Microbial & Insect Germplasm Conservation & Development.

**National Program 215 Goals:** To contribute significantly to meeting national needs for food, fiber, fuel and ecosystem services by restoring, conserving and using the Nation's pasture, hay, turf, and range lands. These goals reflect the objectives of the U.S. Department of Agriculture, and the REE (Research, Education and Economics) Mission Area.

**National Program Approach:** Goals are met through basic and applied research that develops and transfers economically viable and environmentally sustainable conservation, production and monitoring practices and strategies that incorporate an improved understanding of ecological and agronomic processes, enhanced plant materials, advanced technologies, and user-friendly decision-support tools based on sound science.

A full spectrum of basic and applied research is conducted to meet the goals of the program. The scope of research extends from molecular genetics and toxicology to plant genetics and breeding to livestock production systems and landscape monitoring across a variety of ecosystems and agroecosystems.

Solving today's complex problems requires sustained research by interdisciplinary teams working at multiple scales. Creating such teams requires close and sustained collaboration between (1) ARS scientists and locations; (2) between ARS scientists and scientists in other federal, state, local and private research institutions; and (3) between scientists and those citizens who use the products of research.

**Importance of Pasture, Forage, Turf and Rangelands:** Our Nation's grass and shrub lands including pastures, hay, turf and range lands provide forages, open spaces and ecological services that contribute significantly to our agricultural, environmental, economic, and social well-being. Rangeland, pasture, and forages together comprise about 55% of the total land surface of the United States, about a billion acres. Privately owned lands comprise about 45% of this total. These lands represent the largest and most diverse land resources in the U.S. Rangelands and pastures include the annual grasslands of California, the tundra rangelands of Alaska, the hot arid deserts of the Southwest, the temperate deserts of the Pacific Northwest, the semiarid cold deserts of the Great Basin, the prairies of the Great Plains, the humid native grasslands of the South and East, and the pastures and hay lands found in all 50 states from Hawaii to Maine and Alaska to Florida.

These lands are the primary forage base for our livestock grazing industry in the U.S. and are utilized by more than 60 million cattle and millions of sheep and goats. Forage livestock systems are the foundation of an industry that contributes more than \$70 billion in farm sales annually to the U.S. economy. The estimated value of hay production alone is \$11 billion, our third most valuable crop to U.S. agriculture, behind only corn and soybeans. The publicly owned rangelands in the western U.S. are also important, providing forage on 260 million acres for 3 million beef cattle and sheep. Nearly 70% of dietary protein and 40% of dietary calories for the U.S. population are of animal origin, and forage resources are crucial for sustained production of our animal-based products. The functions of these lands are of increasing importance as watersheds and as habitat for a set of biologically diverse plants and animals. Maintaining adequate supplies of clean water for urban areas, irrigated agriculture, and environmental needs is a critical function of rangeland, pasture, and forage-producing ecosystems. Rangelands and

pastures also provide forage and habitat for numerous wildlife species, including 20 million deer, 500,000 pronghorn antelope, 400,000 elk, and 55,000 feral horses and burros. Associated with these functions is an array of additional demands placed on these natural resources, including camping, hiking, fishing, hunting, and other recreational activities. This multitude of uses--from grazing lands to watersheds, critical habitats, and recreational areas--require an improved understanding of basic ecological processes and the effect on these processes on grazing, livestock production, and management practices. Science-based solutions to these needs must be economically viable, socially acceptable and enhance the environment. The overall goal of this national program is to provide the appropriate technologies and management strategies to sustain our rangelands and pastures.

Harvested and conserved forages provide a dietary resource for continuity of livestock production. This is especially important during periods of cold or drought when nutrient-rich plants are not available. Harvested and conserved forages also provide an important source of roughage and nutrients for dairy cattle in confined animal feeding operations. To meet this demand, nearly 200 million tons of forage crops are harvested each year from 73 million acres in the U.S., which is 24% of the cropland. About one-half of these crops provide the forage requirements of dairy cattle. The remainder, along with rangeland and pasture, supplies the forage needs of beef cattle, sheep, goats, horses, and other livestock.

The Nation's 30-40 million acres of turf lands are found around our homes, schools, municipal and commercial buildings, in our parks, greenbelts and recreational areas, and along our roadsides, airports and right-of-ways. These lands contribute to our well-being in many ways including beautifying our towns and cities, enhancing property values, providing vital environmental services and contributing an estimated \$40 billion a year to the economy.

**ARS Locations with NP 215 Research Projects:** This program is national in scope with a nationwide network of locations:

Beaver, WV  
Beltsville, MD  
Boise, ID  
Booneville, AR  
Burns, OR  
Cheyenne, WY  
College Station, TX  
Corvallis, OR  
Dubois, ID  
El Reno, TX  
Ft. Collins, CO  
Langston, OK  
Las Cruces, NM  
Lexington, KY  
Lincoln, NE  
Logan, UT  
Madison, WI  
Mandan, ND  
Miles City, MT  
Prosser, WA  
Raleigh, NC  
Reno, NV  
St. Paul, MI  
Tifton, GA  
University Park, PA  
Woodward, OK

**Cooperation within ARS:** ARS's national programs are uniquely positioned to play an important national role in research to understand and manage the Nation's pasture, hay, turf and range lands. In addition to NP215, other ARS National Programs are making significant contributions to improving the productivity, profitability and environmental sustainability of the Nation's range, pasture, forage and turf lands. These programs include: Water Availability & Watershed Management; Bioenergy; Crop Protection & Quarantine; Crop Production; Climate Change, Soils & Emissions; Agricultural System Competitiveness & Sustainability; Animal Production Systems; Food Safety; and Plant, Microbial, and Insect Genetic Resources, Genomics, and Genetic Improvement.

Because of space limitations, the accomplishments of projects in other national programs related to the management and use of pasture, hay, turf and rangelands are not included in this report unless NP215 scientists were collaborators. An example of research results not included in this report is a significant portion of the water resources research done on pasture and rangelands at ARS locations at Coshocton, OH; Watkinsville, GE; Temple, TX; and Tucson, AZ. Another example is the research on ruminant genetics, physiology and health that is in the Food Animal Production national program.

A number of ARS locations house projects from several national programs that cooperate closely. Examples of where there are both NP215 and Food Animal Production projects include Beaver, WV; Dubois, ID; El Reno, OK; Lexington, KY; Madison, WI; and Miles City, MT. There are NP215 and water resources projects at Boise, ID; El Reno, OK; and Booneville, AR. There are similar examples involving projects from the invasive weeds, climate change, and soils national programs.

As research problems become more national and interdisciplinary in scope collaboration between projects and locations in different national programs is becoming stronger and cross-reporting between programs is increasing through annual reports and co-authored papers.

**Cooperation with Other Public and Private Institutions:** There has always been a need of cooperation between ARS, other federal, state, local agencies, universities and the private sector. As problems become more complex and research budgets become tighter, this need for cooperation is increasing. Greater cooperation is being fostered by expanding communications between Federal agencies at the Washington level and in the regions. Increased emphasis on larger, longer term, interdisciplinary research is increasing the number of joint ARS-University research proposal. ARS-University cooperation is furthered by thirteen of the NP215 locations being co-located on university campuses. Notable examples of NP215 participating in cooperative efforts on national priorities are given in the next section.

## Appendix 3: Impact of Plant Improvement Activities

During this reporting period there were 28 releases of native and introduced grasses, forage legumes and rangeland forbs. Details on these releases are discussed in the specific accomplishments in the four components of this report. The specific benefits of these releases often takes several years to measure as seed is multiplied, marketed and planted. However, the impacts of plant improvement can be significant over time as the following table indicates.

Table 1. Value of selected native and non-native cultivar releases by the ARS Forage and Range Research Laboratory, Logan, Utah for use in the western U.S.

Cultivar	Foundation Seed Sold*	Acres of Certified seed planted (4 lbs/Acre)	Seed Yield per Acre	Lbs of certified seed produced Annually	lbs to the Acre planted	Acres Planted Annually	***Price/lbs	Potential Revenue generated per yr from certified seed sales
Siberian Wheat grass								
Vavilov (95)	15757	3939.25	800	3151400	10	315140	3.4	10714760
**Vavilov II (08)	2273	568.3	800	454600	10	45460	3.4	1545640
Crested Wheatgrass								
Hycrest (85)	4909	1227.25	1000	1227250	10	122725	2.8	3436300
**Hycrest II (08)	3584	896	1000	896000	10	89600	2.8	2508800
Russian Wildrye								
Bozoisky (85)	7674	1918.5	350	671475	10	67147.5	9.5	6379012
**Bozoisky II (08)	2168	542	350	189700	10	18970	9.5	1802150
Meadow brome grass								
**Cache (04)	5667	1417	1000	1416750	15	94450	3.2	4533600
Slender Wheatgrass								
**FirstStrike	893	223.25	350	78137.5	10	7813.75	1.88	146898
					Total	761,306	Total	24,541,250

\* Foundation seed sales provided by Stan Young of the Utah Crop Improvement Association (2009)

\*\* Cultivars released since 2004

\*\*\*Prices taken from 'Utah Seed' Spring 2010 Catalog

## Appendix 4: References for Component 1, Rangelands

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## Appendix 5: References for Component 2, Pasturelands

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## Appendix 6: References for Component 3, Harvested Forages

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