1. PROJECT NUMBER: W-173

2. **PROJECT TITLE:** Stress Factors of Farm Animals and Their Effects on Performance

3. REQUESTED PROJECT DURATION: October 1, 2001 through September 30, 2006

4. STATEMENT OF THE ISSUES AND JUSTIFICATION: Environmental and management stressors erode efficiency and cost livestock production enterprises billions of dollars annually in lost potential profitability. For example, summer heat stress results in annual losses to the dairy industry that total $5-6 billion, due to reduced milk production and reproductive potential (Ray et al., 1992). Additionally, stressor-associated neonatal mortality in the swine industry adds to a staggering sum annually. Under average market conditions, improving survival by one piglet per litter would be worth approximately $400 million annually to the U.S. swine industry. In 1995, economic losses to Iowa cattle feeders that could be associated specifically with severe heat stress were estimated to be $28 million (Dorr, 1995). Additionally, animal well-being research is needed to provide information about how animals interact with the production environment and respond to animal management practices. Animal well-being is a significant societal concern which is difficult to evaluate monetarily. However, research ensuring animal well-being throughout the food production cycle should help decrease animal health-care costs and provide the public with greater assurance related to animal well-being concerns.

Livestock stress and well-being are complex biological phenomena, cutting across both intensively and extensively managed production sectors, covering all species of livestock. The W-173 regional research project has enjoyed a long and productive history and the group has made measurable progress since the last project revision (detailed in section 5.1 below). Although progress has been made toward understanding and dealing with environmental and management stressors, and in continuing to improve farm animal well-being, much remains to be accomplished. The multidisciplinary, collaborative team approach to investigating objectives set forth in this proposal is the most efficient for investigating, addressing, and solving these broad issues. To this end, the current regional research project draws on the strengths of interactions of scientists trained in a variety of relevant disciplines (endocrinology, behavior, immunology, nutrition, livestock management, engineering, statistics, veterinary medicine, etc.) and having expertise in a broad range of livestock species.

5. RELATED CURRENT AND PREVIOUS WORK:

5.1.CRIS Search. W-173 and S-299 are the only multi state projects that have objectives related to environmental and physiological stress of domestic animals. S-299 is a new project that focuses almost exclusively on reproductive performance of heat stressed dairy cattle under hot, humid environments. One component of the proposed new W-173 project will also focus on heat stress in dairy cows but under hot, dry conditions, a significantly different situation.

5.2. Collaborative Accomplishments Achieved Under the Previously Approved W-173 Project. During the course of the previous project, the group has collectively expanded both the basic and applied knowledge base in the areas of farm animal stress and well-being, and livestock environmental management. In total, since the last project revision, participating stations reported 97 full-length, refereed scientific publications and 185 abstracts of research presented at national or international meetings, together with numerous book chapters, nontechnical reports, dissertations, etc. W-173 scientists continue to serve in leadership roles at national and international scientific conferences as organizers and invited presenters on panels and symposia devoted to livestock stress and environmental management. Six very recent examples are cited. First, W-173 co-sponsored an international symposium in August 1998 titled "The Biology of Animal Stress". The symposium was funded by a USDA-NRI grant co-authored by Dr. Joy Mench and the late Dr. Gary Moberg. The publication resulting from that symposium, The Biology of Animal Stress: Basic Principles and Implications for Animal Welfare, is now published in hard back form. Secondly, W-173 scientists organized and participated in a symposium at the 2000 joint meeting of the Midwest Branch of the American Dairy Science Association-Midwestern Section American Society of Animal Science titled "Describing and Understanding the Relationships Between Stress, Behavior, and Performance". Third, and most recently, W-173 participants were fundamentally involved in the organization and in presentations at a symposium held at the 2000 joint annual meeting of the American Society of Animal Science and American Dairy Science Association in Baltimore, MD. This day-long symposium titled "Livestock Transport: Industry Issues and Research Challenges" touched on a multitude of relevant contemporary issues associated with livestock transport, including both American and European perspectives on topics ranging from the basic biology of stress to welfare issues impacted by livestock transportation. Members of W-173 also were involved in organization of a symposium titled "Recent Advances in the Assessment and Management of Heat Stress in Domestic Animals". This symposium was held at the 1998 joint annual meeting of the American Society of Animal Science and American Dairy Science Association in Denver, CO. Finally, W-173 personnel were (1997), and are currently (2001) involved in the planning of recent International Livestock Environment Symposium meetings. Clearly, W-173 serves not only as a mechanism for attacking broad regional research problems, but additionally serves as a vehicle for sharing and disseminating the most current information on livestock stress, health, welfare and environmental management on a national and international scale.

Fundamentally, W-173 and other regional research projects exist to enhance and expand the capabilities of individual research stations beyond what could be accomplished if each worked independently. This enhanced productivity occurs through formal sharing of animal tissues and reagents, sharing ideas, techniques and data sets, and on-site visits and exchanges between scientists at different stations nationwide. Since the previous project renewal, several specific examples of regionally directed accomplishments can be cited.

Relative to the first objective of the previous project, scientists at ARS-MO, ARS-IN, TN, IA and KS evaluated a variety of management and immune stressors in prenatal, neonatal and weaned pigs focusing on a different biological responses in their respective stress models. In each case, the endpoints measured at a single study location were increased, notably without additional animal usage, by exchange of animal tissues and reagents, and by technique sharing from collaborating visiting scientists. The endpoints evaluated in these studies ranged from cellular responses at the transcriptional level to evaluation of live animal performance and behavior. Results of these collaborations have been summarized and published (Lay et al., 1999; Ramanathan et al., 1999; Balaji et al., 2000a; Haussmann et al., 2000; Heo et al., 2000; Hohenshell et al., 2000). Collectively, these studies have established that prenatal stressors have the potential for permanently altering the endocrine stress axis of swine; that immune challenges associated with bacterial cell wall products or with live bacteria produce differing profiles of changes in systemic homeostatic mediators in pigs; and that disease and environmental stressors applied to young pigs produce lasting alterations in cellular and systemic regulators of factors affecting appetite, growth, and health. Another area of collaboration was established between scientists at TN and ARS-TX on studies dealing with the effects of split marketing on the physiology, behavior and performance of finishing pigs. These studies were accomplished through sharing of video equipment for behavioral analysis (Scroggs et al., 2000). Collectively, these studies have furthered the fundamental understanding of the integrated biological responses to management stressors. Scientists at ARS-NE, MO and NE worked together to refine body temperature and respiratory rate measurement techniques in support of basic thermal stress response measures (Gaughan et al., 2000; Parkhurst and Mader, 2000).

Relative to the second objective of the previous project, considerable productive interactions have occurred between scientists at NY, MO, NE, ARS-NE, ARS-TX, CO, MI, HI, and AZ. Much effort has been directed toward the study of effects of heat waves on feedlot cattle. These studies have led to research that is ongoing (ARS-NE, NE, MO, and ARS-TX.) to studies of the effects of heat waves on feedlot cattle (Parkhurst and Mader, 2000). Experiments, completed in environmental chambers and supplemented with field observations, to evaluate responses of cattle to conditions that have caused severe losses of feedlot cattle during heat challenges, have provided the basis for improved management guidelines (Hahn, 1995, Hahn et al., 1998, Hahn et al., 1999; Hubbard et al., 1999; Nienaber et al., 1999; Hahn and Mader, 1997; Mader et al., 2000). Producers can be better prepared for hot weather if they have advanced warning of weather challenges, can recognize early signs of distress (accelerated respiratory rates), and have tools available to ameliorate environmental conditions (sprinklers, shade, or unobstructed airflow). The group also has collaborated extensively to understand how best to alleviate production problems associated with heat stress in dairy cattle. A unique, portable calorimeter was designed and constructed at the NY station to measure, in real time, evaporative heat loss, convective heat loss, and radiant heat exchange from the hair coat of livestock under naturally occurring conditions (Hillman and Gebremedhin 1997, Hillman and Gebremedhin 1999). The portable calorimeter has the ability to vary air velocity across the hair coat to match prevailing air velocities and to adjust to different hair-coat thickness. During the summers of 1996, 1997, and 1999 it was used to measure heat loss from dairy cows under shade in a hot, dry environment at the AZ station. During the summer of 1998, it was used with Simmental heifers at the MO station within environmental chambers and outdoors. In addition it was used to measure heat transfer of Holsteins in a hot, humid environments at the MS station under shade in July 2000 and at the HI station under direct sunlight in August 2000. Finally, the impact of wetting the dorsal skin surface and blowing air over heat-stressed Holsteins was investigated by the NY and AZ stations in the summer of 1999 in a hot, dry environment. NY conducted similar experiments in the summer of 2000 for a hot, humid environment at the MS station. Comparing a hot, humid environment with a hot, dry environment revealed differences. Maximum wetting at an air velocity of 5 mph lowers body temperature .8/C per hour and respiratory rate by 22 breaths/min in the hot, humid environment. In the hot, dry environment body temperature was lowered 1.3/C per hour and respiratory rate was lowered by 35 breaths/min. Both wetting the hair coat and blowing air over the cows are effective in cooling cows, although wetting the hair alone is more effective than only blowing air over the cow. Mathematical models have also been developed for evaporation at the hair coat and air flow around cows (Gebremedhin and Wu 2000, Wu and Gebremedhin 2000). Collectively, these collaborations are contributing significantly to the development of management tools that are useful for effective environmental decision making processes.

5.3. **Stressor Effects on Growth, Behavior, and Immunity**. Activation of the hypothalamic-pituitary-adrenal axis is a well-established response to models of endotoxemia (Matteri et al., 1998) and to acute disease challenge (Balaji et al., 2000). Recent reviews by Johnson (1997) and

Spurlock (1997) suggest that pro-inflammatory cytokines released in response to a pathogenic challenge reduce feed intake and growth. Indeed, both endotoxemia (Wright et al., 2000) and enteric disease (Balaji et al., 2000) are associated with reduced circulating IGF-I in pigs.

Birth is a stressful event and is associated with a prepartum rise in circulating cortisol. In many species, a prepartum rise in glucocorticoids is associated with maturational changes of various tissues that are essential for survival outside the uterus (Thornburn and Challis, 1979; Liggins, 1994). Prenatal and early postnatal events can have effects on the hypothalamus-pituitary-adrenal (HPA) axis that are reflected in the animal's response to stress later in life. Lay et al. (1997a) reported that transportation stress during gestation resulted in an enlarged pituitary gland in the fetal calf. Prenatal stress-induced alterations in the HPA also are maintained in the rat for at least 90 d (Henry et al., 1994), in the calf for 150 d (Lay et al., 1997b) and in the pig for at least 80 days (Haussmann et al., 2000). Piglets delivered by C-section grow poorly and express altered measures of growth- and stress- related endocrine function in the subsequent neonatal period (Daniel et al, 1999; Carroll et al, 2000b).

The natural drive to eat is determined by complex interactions between biological mechanisms of appetite control and responses to challenges from the physical environment. Suppressed intake and performance associated with environmental, social, and disease stresses are well recognized (Forbes, 1995; van Houtert and Sykes, 1996; Gonyou and Stricklin, 1998; Johnson, 1998; Wenk, 1998; Matteri et al, 2000a).

Preliminary evidence suggests that orexin expression is positively correlated with piglet body weight in normally feeding piglets (Dyer et al, 1999). In rodents, hypothalamic orexin gene expression is enhanced by fasting (Yamamoto et al, 2000) and insulin-induced hypoglycemia (Griffond et al, 1999). Orexin pathways may be involved in the response to environmental stressors. The level of hypothalamic orexin mRNA can be decreased by immunological challenge in neonatal pigs (Matteri et al, 2000b). The expression of orexin in the lateral hypothalamus of rats can be acutely stimulated by immobilization or cold stress (Ida et al, 2000).

A series of studies to investigate peripheral and central mechanisms of stress effects on behavior and immunophysiology of the pig has been completed (Salak et al., 1993; Salak-Johnson et al., 1996; 1997). In these studies we began to dissect potential mechanisms of stress-induced behavioral and immune events. Central administration of corticotropin-releasing-hormone (CRH) had profound effects on behavior. Pigs vocalized and were very active for several hours following CRH treatment. CRH induced repetitive behavioral sequences and anxiety-like behavior. CRH briefly reduced NK cytotoxicity and neutrophil chemotaxis. Peripheral administration of cortisol, ACTH and CRH had minor effects on various immune parameters but had no effect on behavior. It is hypothesized that CRH controls behavior responses via the amygdala and other neuropeptides may be responsible for immunological changes induced by stress.

Previous research has involved understanding the effects of stress (i.e., shipping and social stress) on behavior and natural killer cell cytotoxicity in growing pigs using *in vivo* and *in vitro* techniques (McGlone et al., 1993; Morrow-Tesch et al., 1994; Hicks et al., 1998). In these studies, a three-pig model was used to identify the social status of each animal within a group prior to exposure to a particular stressor (e.g., shipping). These studies investigated the interaction between social status and immunological responses of animals exposed to commons stressors. The social status of the animal influenced the animals' immunological response to a stressor.

5.4. Thermal Stress and Meat Quality. Prolonged (> 5d) exposure to heat and high humidity produced a numeric decline in marbling score (Berg, Spiers, and Leonard, unpublished) and finishing cattle, when exposed to a prolonged heat wave, experience loss of intra-muscular fat (marbling) due to the increased energy demand of cooling and lowered energy intake (Leonard, unpublished). Also, the likelihood of dark cutting beef is increased due to the depletion of intramuscular glycogen stores that are necessary for normal muscle pH decline and development of acceptable beef color during the conversion of muscle into meat. The effect of chronic heat in a swine production environment has also been recently evaluated for its influence on fresh pork quality. Spenser (unpublished data) found that pigs raised in total confinement at high ambient temperatures possessed paler pork and higher drip loss. Berg (1998) identified critical points in swine production that may introduce stress on the pig that can ultimately devalue meat quality. These include production environment, pre-transport handling, transport (stocking density, temperature, air flow, length of transit), lairage at the packing plant (presence of water sprays for cooling, feed restriction, length of lairage), and stunning (method of movement to stunner, method of rendering the animal insensible).

5.5. Genetic Effects on Responses to Environmental Stress. Response to environmental stress may be genetically determined, with the heat stress response being associated with a single "thermosensitivity" locus (Hamet et al., 1994). The KIT locus (receptor tyrosine kinase) is an important candidate gene for degree of spotting as an indicator of resistance to solar radiation exposure (Reinsch et al., 1999). Many studies have concentrated on heat stress/shock proteins (HSP) that are induced by numerous stressors, including elevated temperature, pressure, hypoxia, and ischemia (Gray et al., 1999). HSP70 and 72 in cardiac muscle of rats has been used to identify HSP metabolism and phase of heat acclimation (Maloyan et al., 1999). Terajima et al. (2000) has shown that whole-body hyperthermic preconditioning in rats is associated with an increase in hepatic HSP70 and hemeoxygenase/HSP32 activity. Other proteins have also been identified as respondents to heat stress.

5.6. Management of Thermal Stress. Animals respond to a changing thermal environment with both acute and chronic responses, which represent both transient and stable activities, respectively. It is the understanding of the dynamics of these responses that is essential for the development of an effective environmental management program for livestock. From the heat stress side, the continuous buildup of heat load is important, in that one to two days exposure to emergency THI conditions can have significant impact on mortality rate (Hahn and Mader, 1997).

An understanding of the dynamic responses of animals to an environmental challenge is a basis for rational environmental management. Associations between thermoregulation and behavior (Hahn, et al., 1990; Hahn and Nienaber, 1993; Hahn, 1995), between immune parameters and heat stress (Morrow-Tesch and Hahn, 1994), and between the acclimation process and thermal challenges (Korthals et al., 1995), are examples of efforts needed to increase the understanding of dynamic responses to heat stress and development of practical management guidelines. Data from thermal stress studies of feeder cattle established thresholds for respiratory rates (~21 C) and body temperature (~24 C) increases as effective ambient temperature increased (Hahn, 1999). It is also well documented that immune status is altered by exposure to heat stress (Sharma, 1997), with specific effects on antibody titer (Firouzi and Motamedifar, 1999).

A study was conducted to assess adaptation to repeated sinusoidal heat waves. The results show that adaptation does occur with continuous heat challenge, but there is no clear indication that adaptation carries over following a 14-day recovery period at thermoneutrality (Leonard et al., 2001).

During the summer in most regions of the United States dairy cows are heat-stressed. This stress depresses feed intake, milk production, and reproductive efficiency. Heat-stressed cows rely more on sweating to keep cool than from panting. Unfortunately, they do not sweat enough to keep themselves from being heat-stressed (Hillman and Armstrong 2000). To reduce the impact of heat on dairy cows, many dairymen use fans in their barns and many will provide water spray in the feed alley (Armstrong et al. 1999).

A new cooling system has been designed to cool individual dairy cows as they lie down in their stalls (Hillman et al., 2000). When the system detects a cow in a stall, a metered amount of water is sprayed on the cow's back with minimal overspray so as not to wet the bedding. Wetting the bedding must be avoided because it can enhance the pathogenic environment and promote the spread of mastitis. For this system, an ultrasonic transducer is used to detect the presence of a cow. The time between wetting events is controlled by a master unit, which integrates information on air temperature, relative humidity and time of day. A cooled cow will remain lying down in a stall. Milk production should increase with the new system because heat stress will be greatly reduced as compared to systems in current use in freestall barns. In these cooling systems, cows that lie down in the freestall only do so for a short period of time because of the lack of water spray and will get up to stand under water spray. Constant standing or movement also decreases milk production.

Dry matter intake (DMI) by dairy cows is directly affected by heat stress. Reductions begin after 8 days of exposure to 25.6 C (McGuire et al., 1989) and continue to decline to 60-67% of normal intake at 40 C (Johnson et al., 1963, McGuire et al., 1989). This behavior lends support to strategic cooling during nighttime hours, and the potential for an increase or maintenance of DMI. Mid-lactation, Holstein cows were used in experiments to compare use of fan cooling during night-time (low Ta) to day cooling (high Ta). Fan cooling at Ta 24C was 3.3 times more effective in increasing heat loss than at 33.3C. The primary benefit of night-cooling was to shift Tcore and respiration rate below the levels seen in day-cooled cows. These differences in daily thermal balance were associated with significant changes in feed intake, milk production, and reproduction. Decline in feed intake and milk production was less in NC compared to DC cows. In addition, NC cows, compared to DC cows, had an increase in number of large ovarian follicles. These results showed that fan cooling during the night is superior to day cooling in reducing heat load of dairy cows at elevated air temperatures (Spain et al., 1998; Spiers et al., 1998).

Extreme cold temperatures also provide environmental challenges for cattle. During winter cattle in feedlots in Colorado can lose 30 watts per square meter more heat when exposed to clear cold skies than when exposed to cloudy skies (Hillman et al., 1996). The NY station measured heat transfer through pelts samples provided by CO, while CO collected meteorological data. Although this heat loss is modest, it can account for a loss of 0.17 kg of potential weight gain on a cold clear night as compared to a cloudy night. Depending on how many cold clear nights occur during the winter, this additional heat loss contributes to the observed 0.15 kg lower daily weight gain of winter cattle as compared to summer cattle.

5.7. Impact of Social Stress. Maternal separation exposing a mammalian dam to stress during gestation has been associated with profound behavioral and physiological alterations in her resultant offspring (termed "prenatal stress", Haussman et al., 2000). However, controversy exists as to whether these alterations are induced by maternal glucocorticoids or opioids. The inability of researchers to control the maternal compounds that may affect the developing fetus have hindered the elucidation of the mechanism responsible for prenatal stress. Thus, experiments were designed to determine if the chicken could be developed as a model for prenatal stress due to the developmental autonomy of the chick embryo. Development of this model would allow research to be conducted on prenatal stress without having to control for passage of maternally derived compounds. As a test to develop this model, on d 16 of incubation, eggs were given 60 ng of corticosterone, elevated incubation temperature (40.6 C) for 24 h, or no treatment. Administration of exogenous corticosterone to chicks during incubation replicated some, but not all, of the effects seen in prenatal stress in mammals (Lay et al., 2000). Further development of this model to study prenatal stress may prove invaluable in furthering our knowledge of the mechanism for prenatal stress.

Modern livestock production is potentially stressful to swine. When a pregnant sow is stressed, cortisol crosses the placenta to possibly affect the fetal hypothalamus and development; a process termed prenatal stress. A study examined the physiology and behavior of pigs whose dams were injected with adrenocorticotropic hormone (ACTH) during gestation, which replicates the effects of prenatal stress in other species. These data showed that exogenous ACTH during gestation causes a hyperactive HPA axis of the sow's offspring and during stressful situations later in life, growth, health, reproduction and welfare may be compromised (Haussman et al., 2000). A research collaboration between TX and TN investigated the relationship between plasma and salivary cortisol, and corticosteroid-binding globulin (CBG) in pigs during acute and chronic administration of ACTH. CGB increased during chronic administration of ACTH, making the relationship between bound and free concentrations of cortisol and salivary cortisol (free) rather complex.

5.8. Other Management Stressors. A number of management decisions can alter the stress response. Among them are dietary manipulations, dietary supplements for growth and health manipulations, dehorning, castrations, and transportation. Management decisions regarding these procedures can vary the degree of stress.

A combination of a yeast cell-wall component (beta-glucan) and a protected vitamin-C product has been used successfully to enhance growth and decrease response to an LPS challenge in piglets from birth through 28 d of age. Pigs receiving both products had enhanced growth and cortisol was decreased by vitamin C with and without beta-glucan. These data demonstrated that the inclusion of vitamin C, beta-glucan, or both altered piglet growth and the response to an endotoxic challenge (McKee et al., 2000).

First-calf dairy heifers experience a number of stressors at calving; new environment, metabolic and dietary changes, hormone changes, increased metabolic demand by milking, calving, and frequently a first contact with the milking parlor and milking machines. Possible solutions include habituation to the milking parlor or milking prior to calving. Greene et al. (1988) found no significant differences between prepartum milked heifers and their contemporaries milked postpartum for days to first recorded estrus, days to first breeding, days to conception, number of inseminations, or conception rates. However, there appeared to be a slight advantage for prepartum milked heifers in days to first recorded estrus and in days to first breeding, which suggests that prepartum milking is not detrimental to fertility, but further work is needed to elucidate any effect. This was further supported by a recent study by Kearney et al. (2000) who observed that premilked heifers had fewer services per conception and fewer days open than control heifers. Haptoglobin, an acute phase protein that follows tissue damage, returned to pre-calving concentrations sooner in heifers that had been pre-milked for 3 wk prior to parturition (Eicher et al., 1999). Therefore, we hypothesize that uterine involution may also occur more quickly in premilked heifers.

Transportation is a complex stressor, involving mixing of animals, feed and water removal, thermal conditions, and movement within a vehicle. The impact of this stressor has been shown to be different on high-lean compared to low-lean lines of pigs (Busse et al., 2000). The effect of transport stress on pigs showed the relationship between social status and lymphocyte proliferation and natural killer cell cytotoxicity disrupted during acute transport stress (Hicks et al., 1998). Isolation was determined to be a major cause of stress associated with transport at 7 to 10 days of age (Morrow-Tesch and Dailey, 2000).

5.9. Literature Cited: Refer to Appendix 1.

6. OBJECTIVES:

7. **Identify appropriate measures of animal stress and well-being and characterize factors affecting the biology of the stress response.**

8. **Evaluate management strategies that minimize the detrimental effects of animal stress.**

7. METHODS:

7.1 The following are collaborative research efforts to address the first objective (item 6.1 above).

IL will conduct studies designed to measure ex vivo functions of alveolar macrophages and blood monocytes, and to explore the potential physiological basis for the impaired functions of cells of the immune system. Briefly, pigs are exposed to multiple simultaneous stressors, at the end of the experimental period blood monocytes and lung macrophages are isolated. The functional aspects of blood monocytes and macrophages using various immune assays and the role chemokines play in mediating stress-related immunosuppression are determined.

MS, TN, TX, HI, and ARS-IN are collaborating to investigate the role of the adrenal gland during pregnancy, the estrous cycle and in response to various reproductive management techniques. These will include measures of pituitary and adrenal responses, and interrelationships with oxytocin release. Management strategies will include mimicking primitive procedures for milk let-down, induced anestrus (pseudo-pregnancy) and synchronization of estrus in parallel with modern strategies for reproductive management. Moreover, the effects of environmental stressors (e.g., shade vs. no shade) on adrenal responsiveness and reproductive function will be examined in MS and HI. MS and HI will conduct the animal trials and steroid hormone assays, while TX will conduct the oxytocin analyses. In addition, the role of the adrenal gland during pregnancy and different stages of the estrous cycle will be investigated by MS and ARS-IN. In these studies, ACTH will be administered at various stages of pregnancy (early, mid and late), and during the different phases of the estrous cycle (proestrus, estrus, metestrus and diestrus). Serum concentrations of progesterone and cortisol will be monitored pre- and post-challenge, and responses compared across the different stages of reproductive activity.

MO will evaluate changes in live animal characteristics associated with carcass composition and meat quality (marbling content) via ultrasonic evaluation by a trained, certified technician.. Carcass measurements will include yield and quality grade, objective color evaluation of lean tissue utilizing a HunterLab MiniScan XE Spectrophotometer to record L*-value (lightness darkness), a* (redness), b* (yellowness), chroma (color saturation), and hue-angle (measure of "true red"), intramuscular pH at various times postmortem, water holding capacity, Warner/Bratzler shear force determination (meat toughness), and chemical analysis of crude protein, crude fat, moisture and ash. MO also has the capacity for determination of total glycogen content within muscle for evaluation of the glycolytic potential. Glycolytic potential is determined by the equation; 2([glycogen] + [glucose] + [glucose-6-phosphate]) + [lactate]. Determination of glycolytic potential will provide an indication of postmortem metabolites involved in the occurrence of dark cutting beef.

KY and NE-ARS will continue with the NCPIG physiological model development and heat production responses to ambient temperature, relative humidity and wind speed will be modeled and assessed under various conditions. Data from the model will be compared to observed data collected at NE-ARS for differing lean growth potential genetics, and from other locations.
TN, TX, KY and CA will work together on projects related to transportation in horses. A project in equine response to transport will be conducted to assess characteristics of horse response to trailer environment in terms of deep body temperature and possibly other measures for various trailer designs. A model of heat and mass transfer will be developed to aid in improved trailer design, and will be compared to observed data for at least three trailer designs and/or ambient conditions during transport. Additional work will involve characterizing ammonia levels and potentially other measures of air quality in equine facilities.

Research will continue to assess dynamic responses of cattle in hot weather. KY, NE-ARS, and NY will conduct research on responses of grazing cattle to heat stress, and use of shade. This research follows from previous work by KY and others characterizing the deep body temperature response of grazing beef cattle in the presence of shade or no shade. NE-ARS, NE and MO will measure body temperature and respiratory rates of feeder cattle during heat challenges in laboratory and feedlot situations, including comparisons of shade and no shade.

TX and NE will collaborate in a effort to apply models of body temperature change developed by NE to acute changes in body temperature in elephants and tigers during transport. NE developed the models originally for use with cattle in collaboration with other members of W-173.

AZ and MO will collaborate to create a database of genomic information that integrates the Monsanto EST database of approximately 27,000 EST's, public bovine ESTs extracted from GenBank database and additional EST's generated from this project. Glass chip microarrays utilizing approximately 5000 EST's will be utilized for broad coverage of potential functional genes of interest. Nylon-arrays involving approximately 500 genes will be utilized to focus on genes in specific metabolic pathways such as the stress response, protein synthesis in mammary tissue or fatty acid metabolism in mammary tissue.

MO-ARS, IN-ARS and TN will continue collaborative work on the effects of weaning stress on behavior, neuroendocrine regulators of appetite, growth, immunological responses, and stress responses. We will examine the possible involvement of neuroendocrine appetite-regulating factors in stress responses. As a first step, levels of gene expression will be determined during early postnatal development (MO-ARS and ARS-IN). One of these factors, agouti-related protein (AGRP), has been detected in rat adrenal glands. We will determine the presence of mRNAs for recently-discovered appetite-regulating factors in tissues of the HPA axis. Depending on the results of tissue screening analyses, the effects of various stressors on gene expression will then be evaluated. Depressed appetite is a well-recognized consequence of disease stress. Preliminary evidence suggests that LPS exposure can decrease the production of orexin mRNA in the hypothalamus of the neonatal pig (Matteri et al, 2000b). MO-ARS will examine the effects of orexin injection on blood hormone and metabolite levels in baby pigs. The results will provide a direct indication of potential beneficial effects of orexin on glucose metabolism.

KS and AL will utilize disease models in pigs and sheep, respectively, to evaluate pathophysiologic sequelae of this noncognitive stressor. A prolonged surge in cortisol occurs from 24 to 72 h following infection of pigs with *Salmonella typhimurium* (Balaji et al., 2000). KS will focus on the role of cortisol in directing the immune response toward a Th1 or Th2 response. AL will continue ongoing studies of the effects of endotoxemia on the regulation of GH secretion.

7.2 The following are collaborative research efforts to address the second objective (item 6.2 above).

Cooling systems for dairy cattle under various housing conditions will be evaluated. A study will be conducted in CA because of predictable periods of heat stress. Ten stalls in a free-stall barn will be outfitted with the new cooling system developed in NY. Cow responses under the new system will be compared to cows using fans over the free-stalls and sprinklers at the feed alley. Investigators from NY, CA, and HI stations will record core body temperatures, respiration rates, behavioral responses, milk production, and meteorological data. The new cooling system utilizes a master controller to regulate the timing between cooling events. It will monitor and integrate relative humidity, ambient air temperature and time of day. These data will be used to determine when the next cooling event will be initiated. The interaction and value of humidity, temperature and time of day for an appropriate cooling event will be determined experimental and using interpretations from the literature. NY will continue its collaborate experiments with CA, HI and MS stations at commercial or experiment station dairy farms to test the efficacy of the new cooling system. Using similar devises, TX and NY stations will apply this technology to cooling elephants. A mathematical model (NE) will be used to analyze effects of cooling strategies in conjunction with AZ, NY and HI.

In dairy cattle research, KY will examine technologies such as evaporative cooling, tunnel ventilation, and direct wetting of cows to reduce heat stress and assist lactating cows in coping with heat stress conditions. Gates' previous work in time integrated ventilation as a management system will be continued and system designs tested for improving cow environments under heat stress. KY will perform field tests to assess benefits and considerations in adaptation of the available technology. Cooperation with other stations includes AZ, NY and KY who are all working on heat stress in dairy cattle. A tunnel ventilation system will be established at MS that will house 20 cows. Physiological measurements including immune status (ARS-IN) will be taken on cows inside and outside of the facility. Group feed intakes, ambient environmental data, milk production data and SCC will be analyzed. Serum hormonal profiles (progesterone and cortisol) that reflect heat stress and cooling will be determined (MS). Cow behavior in the tunnel will be recorded by video (ARS-IN) to provide indications of cow comfort and to shed light on differences between cows in the tunnel ventilation facility and traditional free-stall arrangements with respect to important production variables.

An experiment will investigate the efficacy of sunshades during hot summer weather for beef cattle in feedlots. NY will provide its portable calorimeter to identify the component of the cattle's energy budget under shade and under sunlight at the ARS-NE facility. Respiration rates, body temperatures, feed and water intake, and meteorological data will be recorded to characterize the cattle's thermoregulatory responses. In beef systems, shade will be evaluated on grazing systems and its use by cattle for no, intermediate and high shade levels in a variety of direct and diffuse solar radiation environments.THI work will be conducted by KY, and will complement work by ARS-NE and NY in the area of shade effects on cattle.

In application of the GPS technology, GPS and GIS will be used to develop improved management strategies for cattle grazing systems. Work will be conducted to examine cattle behavior in relation to shade and water systems, and implications for riparian zone management. KY will lead this effort. ARS-NE will work with KY on cattle movement using GPS. Work performed under this objective at the KY will include studies of heat stress mitigation strategies for dairy, beef and swine, and on assessing management strategies for cattle grazing using GPS technology.

Data (animal performance and thermal conditions) is being collected within the ARS-NE swine production facilities as a means of evaluating the NCPIG model under production conditions, using two distinct genotypes differing in carcass lean content. A series of experiments is planned for the environmental chambers and calorimeters to compare groups of pigs to individual animals under heat stress conditions, using hot cyclic and constant temperatures and two genotypes. Each of these factors will be used to evaluate an aspect of NCPIG as well as to collect basic data on number of pigs per pen, temperature pattern and genetic effects as influenced by heat stress.

Prenatal and neonatal stress has a lasting impact on an animal's ability to cope with stressors. ARS-IN will examine how stress of the pregnant dam can affect her developing fetus and how manipulation of pen systems for sow housing could minimize aggression and the expression of abnormal behavior in swine. ARS-MO and ARS-IN will collaborate on these studies. Behavioral and immune measures will be provided by ARS-IN and neuroendocrine measures by ARS-MO. TX will collaborate with NE to determine the efficacy of imprint training of foals in reducing the stress response of foals during handling and training. TX will conduct the trials with over 200 foals and UNL will develop the analysis of the data.

TX, ARS-TX, FL, ARS-IN, and IL will continue the study of transportation stress with swine, beef cattle, dairy cattle, and horses. Behavioral (TX, ARS-TX, ARS-IN, IL), physiological

(ARS-IN, FL), and immunological (ARS-TX, ARS-IN, and IL) endpoints will be utilized to measure the effectiveness of new management practices during the transportation process. Assistance with data analyses will be provided by NE to determine the most effective measure or group of measures in determining stressed animals.

8. MEASUREMENT OF PROGRESS AND RESULTS:

Outputs: The committee will continue to produce a steady flow of refereed journal articles, abstracts, symposium papers and articles for industry journals. In addition, new, improved and modified techniques needed to evaluate and measure animal stress will continue to be developed.

Outcomes or projected impacts: Collectively, research directed toward both objectives should advance the understanding of the biology of the stress response and important components and measures of animal well-being. In addition, these cooperative efforts will identify management practices that will improve animal environments and reduce animal stress responses to those practices. The animal response measurements provides a basis to develop response functions that can be used to predict outcomes and to optimize management of the thermal environment. Similarly, use of the dynamic response measurements has given us a basis to predict when an animal is under stress or distress and in need of attention to minimize detrimental effects. We expect this information to become more readily usable for direct application in animal production systems. Such application will reduce animal stress and increase animal productivity resulting in increases in net income for livestock enterprises.

Milestones: Relative to the first objective, the group collaborating on evaluation of the effect of the adrenal on reproduction will first have to validate procedures to model experimental induction of milk let down and pseudopregnancy before ACTH challenge studies can be initiated. Researchers evaluating the effect of thermal stress on muscle quality must establish a reliable beef cattle model that produces desired changes in muscle color and post-mortem muscle biochemistry prior to evaluating intervention approaches. Stations collaborating in the horse transportation area must establish important physiological and behavior measures before transportation management that minimizes stress can be evaluated. Researchers from MO and AZ will have to establish the appropriate experimental heat stress environment prior to utilizing microarray technology to identify important stress-induced genes in mammary tissue. Relative to the second research objective, stations collaborating on dairy cattle cooling methodology must fabricate cooling equipment to be utilized at multiple research cites prior to comparing the technology to free-stall fans. Also, collaborators evaluating shading technologies for grazing cattle and cattle in feedlots will have to conduct preliminary studies to verify appropriate shade design prior to evaluation of shade density on the thermoregulatory response in hot environments. Finally, researchers collaborating in the area of transportation stress will first have to establish immunological, behavioral, and physiological endpoints that are reliable indicators of stressful transportation. These measures will be important in asking appropriate experimental questions relevant to animal friendly transportation technologies.

9. PROJECTED PARTICIPATION:

Participant Name	E-Mail	Institution & Dept	Research						Extension		Objectives				
			CRIS Codes			Personnel					1	2	3	4	5
			RPA	SOI	FOS	SY	PY	TY	FTE	Program					
Robert J. Collier	rcollier@ag.arizona.edu	University of Arizona Animal Sciences	303 305 306	3310 3410	1040 1020	1.0	0.5	0.5			x	x			
J. L. Sartin	sartinl@vetmed.auburn.edu	Auburn University Animal Health	315	3999	1020	0.1									
Carolyn L.Stull	clstull@ucdavis.edu	UCDavis Vet. Med.	305 306 307	3310 3410 3810	1020 1010	0.2			0.3	Agriculture	x	x			
John Arthington	jarth@gnv.ifas.ufl.edu	University of Florida Range Cattle	307	3310	1010	0.1						x			
C. N. Lee	chinl@hawaii.edu	University of Hawaii HNFAS Dept	306 306	3410 3310	1020 1010	0.1 0.1	0.5	0.5	0.3	Agriculture	x	x			
James R. Carpenter	cjim@hawaii.edu	University of Hawaii HNFAS Dept	306 306	3310 3410	1010 1020	0.2 0.1			0.1	Agriculture	x	x			
Douglas L. Vincent	vincent@hawaii.edu	University of Hawaii HNFAS Dept	306 306	3310 3410	1010 1020	0.1 0.1			0.1	Agriculture	x	x			
Janeen L. Salak-Johnson	johnso17@uiuc.edu	University of Illinois Animal Science	306	3510	1020	0.2					x	x			
J.Ernest Minton	eminton@oznet.ksu.edu	Kansas State Univ. Animal Sciences	305	3599	1090	0.1					x				
Frank Blecha	blecha@vet.ksu.edu	Kansas State Univ. Anatomy & Physio	305	3599	1090	0.1					x				
Larry W. Turner	lturner@bae.uky.edu	University of KY Bio & Ag Engr	306 315.402	3910	2020	0.1	0.1	0.1			x	x			
Richard S. Gates	gates@bae.uky.edu	University of KY Bio & Ag Engr	306 315.404	3910	2020	0.1	0.1	0.1			x	x			

Jose R. Bicudo	jbicudo@bae.uky.edu	University of KY	306	3910	2020	0.1		0.1	0.1	Agriculture	x	x
		Bio & Ag Engr	315.402									
Anne M. Parkhurst	aparkhurst@unl.edu	UNL	306	7310	2090	0.5	0.5	0.5	0.2			
		Biometry										
Scott T. Willard	swillard@ads.msstate.edu	Mississippi State	306	3410	1020	0.1	0.5	0.1			x	x
		Animal & Dairy										
Don Spiers	spiresD@missouri.edu	University of MO	306	3310	1020	0.2		0.2			x	x
				3410	1020	0.2		0.2				
Erie Antoniou	antoniouE@missouri.edu	University of MO	306	3840	1020	0.1					x	x
Eric Berg	bergEP@missouri.edu	University of MO	306	3510	1020	0.2					x	x
Henry G. Kattesh	hkattesh@utk.edu	University of TN	305	3910	1020	0.3	0.5	0.1			x	
		Animal Science										
Peter E. Hillman	peh1@cornell.edu	Cornell Univ.	306	3410	2020	0.4						x
		Ag & Bio Engr.										
Kifle Gebremedhin	Kgg1@cornell.edu	Cornell Univ.	306	3410	2020	0.3						x
		Ag & Bio Engr.										
Ted Friend	t-friend@tamu.edu	Texas & AM Univ	306	3410	1060	0.1	0.1	0.1			x	x
		Animal Science										
John Nienaber	nienaber@email.marc.usda.gov	MARC, USDA/ARS	306	3510	2020	0.5					x	x
Tami Brown-Brandl	brandl@email.marc.usda.gov		306	3310	2020	1.0					x	x
Roger Eigenberg	eigenberg@email.marc.usda.gov		306	3310	2020	0.3					x	x
Susan Eicher	spruiett@purdue.edu	USDA/ARS	305	3410	1090	0.5					x	x
			315	3510								
Robert L. Matteri		USDA/ARS	306	3510	1020	0.5	0.5	1.0			x	x
Jeffery A. Carroll		USDA/ARS	306	3510	1020	0.8		0.8				x
Julie Morrow-Tesh	julie.morrow@ttu.edu	USDA-ARS,SPA	305	3510	1020	0.3					x	x
			307	3310								
Terry E. Engle	engle@lamar.colostate.edu	Colorado State Univ	302	3310	1010	0.2	0.3				x	x
		Animal Science										
TOTALS						9.3	3.6	4.3	1.1			

10. OUTREACH PLAN:

The project is expected to result in peer-reviewed scientific publications, as well as abstracts of research presented at national and international meetings, non-refereed research reports, extension publications and theses/dissertations. Additionally, many of the project participants hold appointments at land-grant institutions and have colleagues within their home departments who hold extension appointments. Data generated from the current project that appears to have practical application will be evaluated by extension personnel for appropriate dissemination at producer meetings. The committee has a long history of organizing and participating in scientific symposia. It is anticipated this activity will continue.

11. ORGANIZATION AND GOVERNANCE:.

The Executive Committee of the Regional Technical Committee shall consist of the Chair, Secretary, and immediate Past-Chair. A new Secretary will be elected each year by the voting members of the Technical Committee. The previous Secretary will become the Chair for one year and then will move to the Executive Committee for and additional year. The Executive Committee will have the authority to act on behalf of the Regional Technical Committee. If any member of the Executive Committee resigns, the remaining member shall, with the advice and consent of the Administrative Advisor, appoint a member of the Regional Technical Committee to fill the vacancy. The term of the office will end at the adjournment of the regular annual meeting. The new immediate Past-Chair will prepare the annual progress report and submit it to the Administrative Advisor. The new Chair (previous Secretary) will prepare a set of minutes of the annual meeting and send it to the Administrative Advisor for distribution to the Regional Technical Committee.

12. AUTHORIZATION:

May 11, 2001

Colin Kaltenbach, Administrative Advisor | Date

Western Association of Agricultural Experiment Station Directors

August 6, 2001

APPENDIX A-References

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