**Project/Activity Number:** Multistate Research Project NC1184

**Project/Activity Title:** Molecular Mechanisms Regulating Skeletal Muscle Growth and Differentiation

**Period Covered:** 10/2017 to 9/2018

**Date of This Report:** 12/17/2018

**Annual Meeting Dates:** 10/26-27/2018

**Station Participants:**

|  |  |  |  |
| --- | --- | --- | --- |
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**Summary of Minutes – Annual Meeting:**

The meeting convened at 8am on Friday October 26, 2018 and concluded on Saturday October 27, 2018 in the conference room of the Stan L. Albrecht Agricultural Sciences Building at Utah State University, Logan, UT. Dr. Dirk Vanderwall, Head of the Department of Animal, Dairy, and Veterinary Sciences at Utah State University, provided the welcome and introductory remarks. These included a thorough overview of the research and teaching facilities and capabilities at Utah State University. Following this introduction, individual station reports were presented by each of the stations represented. These reports lasted approximately 30 minutes each with time for questions and discussion. In the evening, the group met for dinner, which was hosted by Dr. Kara Thornton at the Utah State University Beef Teaching Unit. NC1184 members participated in a scheduled conference call with Dr. Mark Mirando (USDA-NIFA), who outlined the current funding programs, budgets, and statistics on the number of proposals submitted annually and funding rates. An update on the plan to relocate NIFA outside of DC was also discussed. In addition, a committee led by Dr. Dave Gerrard and including Drs. Sally Johnson, Sandy Velleman, Derris Burnett, Paul Mozdiak, and others was formed to re-write the multistate project. An update on travel plans for the 2019 meeting in Starkville, Mississippi was discussed and it was determined that the 2020 meeting will be held at the University of Connecticut and hosted by Dr. Sarah Reed.

**Accomplishments:**

**Objective 1: *Characterize the signal transduction pathway that regulates skeletal muscle growth and metabolism including the influence of endogenous growth factors and various production practices.***

**Alabama Station:**

Impact of in ovo thermal manipulation on broiler chicken muscle development, growth, and satellite cell activity.

1. Completed the live animal growth performance and carcass yield data collection and sample collection and cryohistology and immunofluorescence analysis portions of the project.
2. Immunofluorescence data analysis is ongoing.
3. Presented an abstract of growth performance and carcass yields at the 2018 Poultry Science Association annual meeting.

Effects of dietary amino acid density on growth performance, satellite cell activity, collagen gene expression, and the incidence of wooden breast.

1. Completed the live animal growth performance and carcass yield data collection, sample collection, and data analysis portions of the project.
2. Presented an abstract with collagen expression results at the 2018 Poultry Science Association annual meeting.
3. Published the results in a peer-reviewed journal, *Poultry Science*.

**Connecticut Station:**

Effects of poor maternal nutrition during gestation on fetal muscle development

1. Completed metabolome analysis of longissimus dorsi muscle from offspring of over-, restricted-, and control-fed ewes at day 45, 90, 135 of gestation and within 24 h of birth.
2. Identified changes in markers of oxidative stress as a result of maternal diet in serum and muscle in offspring within 24 h of birth.
3. Initiated proteomic analysis of longissimus dorsi muscle from offspring of over-, restricted-, and control-fed ewes at day 45, 90, 135 of gestation and within 24 h of birth.

Effects of restricted maternal nutrition and realimentation during gestation on fetal muscle development

1. Histological analysis of fetal muscle tissues to determine changes in fiber cross-sectional area, Pax7(+) cells, and muscle fiber typing approximately 75% complete

**Hawaii Station:**

Neonatal suppression of MSTN on skeletal muscle growth

1. Either neonatal oral administration or intraperitoneal injection of MBP-fMSTNpro45-100mFc did not significantly affect body weight growth and gastrocnemius muscle and organ weights of mice. The results imply that the early neonatal administration of MBP-fMSTNpro45-100mFc does not enhance muscle hyperplasia in mice. However, this study did not examine either the transfer of recombinant MSTNpro into circulation or dose-response relationship. Further studies, thus, are needed to validate the potential of neonatal suppression of MSTN as a strategy to improve skeletal muscle growth of animals.

**Idaho Station**

1. Completing an aquaculture feeding trial in sablefish (Anoplopoma fimbria) examining the influence of rearing temperature and dietary composition on growth traits and temporal expression of myogenic and metabolic genes in both white and red skeletal muscle. This comprises the final research trial for an MS student with projected completion of December 2017.

**Illinois Station:**

1. Mice were genetically edited using tail-effector-like endonuclease technology (TALEN) to mimic the naturally occurring IGF2 G3072A mutation prevalent in pigs. Mice containing a mutation that disrupted the binding of a transcriptional repressor of IGF2 were successfully generated and characterized. Pigs with similar mutations display increased muscle growth but reduced fat deposition. In contrast, mice experienced increased muscle growth, increased fat deposition and increased organ weights.
2. In our ongoing work to determine the role of myostatin in muscle growth in pigs, we investigated whether epigenetic mismodifications accounted for the lack of viability in myostatin null pigs. While epigenetic changes were present when comparing the unmodified fibroblast cell line with that of cloned pigs, there was no obvious epigenetic change that accounts for the lack of viability in myostatin null pigs.
3. The abundance of beta-adrenergic receptor subtypes in beef tissues was determined by Western blotting. All three subtypes of beta-adrenergic receptors were detected in beef muscles (longissimus lumborum and psoas major), organs (lung, heart, kidney, and liver), and adipose tissues (visceral, subcutaneous, and intramuscular). Beta-1 receptor abundance was greater in muscle compared with fat tissue but beta-2 abundance was more similar between muscle and fat. Between the tissues investigated, abundance of beta-3 receptors did not vary.

**Indiana Station:**

1. Pten is a phosphatase that antagonize growth factor (IGF1) signaling. We reported deletion of Pten in embryonic myoblasts leads to postnatal muscle hypertrophy but disrupts satellite cell homeostasis (Yue et al, 2016, *Cell Reports*).

**Iowa Station:**

* + - 1. We completed tissue collection for our next experiment related to modification of the PGC-1α pathway via nutraceuticals. We have expanded our interventions to include quercetin, nicotinamide riboside, Lisinopril, and Prednisone and combinations thereof. Preliminary analyses are underway and a histological and biochemical examination will begin soon.

1. We confirmed dysfunctional autophagy in dystrophic skeletal muscle. Importantly, we are the first group to document release of autophagosomes, termed autophagosome escape, from dystrophic muscle. Importantly, we also collected compelling evidence of this same phenomenon occurring in healthy muscle. As they are found in the extracellular space, they may participate in paracrine signaling and considering the mass of muscle and that they escape the muscle environment may participate in endocrine signaling.
2. We have begun experiments to better understand why autophagy is dysfunctional in dystrophic muscle, which are now focused on regulation of transcription factor EB, the primary transcription factor driving lysosome biogenesis.

**Minnesota Station:**

1. When equine satellite cell myotube cultures were treated with 408-µM leucine in the presence or absence of the mTOR inhibitor rapamycin, suppressed phosphorylation of mTOR (*P* < 0.01) and rS6 (P < 0.01) with an increase in phosphorylation of rS6 in leucine-treated cultures observed when compared to control cultures (P < 0.05). Similarly, there was a 27% increase (*P* < 0.005) in the hyperphosphorylated γ-form of 4E-BP1 compared to total 4E-BP1 in leucine treated cultures compared to control cultures with leucine-induced phosphorylation of 4E-BP1 completely blocked by rapamycin.
2. Treatment of equine satellite cell myotube cultures with β-hydroxy-β-methlybutyrate (HMB) increased the protein synthesis signal transduction pathway (AKT – mTOR – S6 and 4EBP-1) in a dose dependent manner between 3 and 12 uM HMB ((*P* < 0.05).
3. Protein synthesis was evaluated in equine satellite cell myotube cultures treated with a leucine titration ranging from 0- to 408-µ*M*. Our results show a 1.8-fold increase (*P* < 0.02) in protein synthesis at levels slightly greater than those found in the general circulation, 204- and 408-µ*M* when compared to a no leucine control (0-µ*M*). Puromycin incorporation, a nonradioactive surface sensing of translation (SUnSET) methodology, demonstrated a 180% increase (*P* = 0.0056) in puromycin incorporation in leucine compared to control cultures.

**Mississippi Station:**

Effects of melatonin supplementation during gestation on fetal and neonatal muscle development in bovine offspring

1. It was determined that Melatonin supplementation during gestation increases the primary to secondary muscle fiber ratio in beef cattle offspring at 240days of gestation.
2. Melatonin supplementation does not increase myogenic gene expression but does increase energy metabolism related expression. These findings indicate the potential for melatonin to act as a therapeutic agent during fetal development by changing the histological and metabolic disposition of skeletal muscle

**New Jersey Station:**

1. Over the past funding cycle, this station characterized the Unfolded Protein Response (UPR) in the skeletal muscle of untrained/unfit Standardbred horses (male and female) in response to an acute bout of maximal effort treadmill exercise. We also measured and assessed UPR biomarkers following an acute bout of maximal effort treadmill exercise in 12-week trained/fit Standardbred horses. These efforts revealed the UPR to be activated in untrained skeletal muscle after exercise but not in trained skeletal muscle. This station also characterized the skeletal muscle metabolome in response to acute exercise in untrained/unfit versus trained/fit Standardbred horses. These findings are together under manuscript preparation for publication in peer-reviewed journals.
2. Over the past funding cycle, this station explored the mechanism by which dietary sulfur amino acid restriction (SAAR) improves body composition in male and female mice. Specifically, we explored whether the transcription factor ATF4 is needed to increase food and fluid intake and energy expenditure, and improve body composition by regulating the hepatokine, fibroblast growth factor 21 (FGF21). It was determined that ATF4 is required for protection from adiposity but not for increased food and water intake, energy expenditure, or hepatic Fgf21 expression during SAAR and that FGF21 does not work exclusively through an ATF4-FGF21 axis. Whole body loss of *Atf4* did not promote skeletal muscle loss to dietary SAAR.

**Utah Station:**

1. Determination of mechanism through which decreased plane of nutrition in second trimester alters end-product quality of offspring in beef cattle
   1. Samples were collected from offspring of mother cows that either maintained BCS during the second trimester (MAIN) or from cows that dropped one BCS during the second trimester of pregnancy. Samples were collected from the *longissimus dorsi* at weaning, prior to beginning the feedlot phase and immediately following harvest.
   2. Completed miRNA analysis of samples from weaning, the beginning of the feedlot phase and immediately following harvest. Ten different miRNA were analyzed using qRT-PCR methods.
2. Gained insight into how different organic pastures impact dairy heifer development.
   1. A total of 6 animals per treatment were used to study 8 different pastures.
   2. Animal growth, serum IGF-1 concentration, blood urea nitrogen concentration, and parasite load were measured.

**Washington Station:**

1. During the past year, we are continuing to define mechanisms regulating myogenesis and early skeletal muscle development. In mice, we found that exercise during pregnancy can promote fetal muscle development, especially when mothers are obese and consuming a high fat diet. In addition, we found that vitamin A supplementation during the pregnancy and lactation enhances fetal and neonatal muscle development and overall animal growth, which was confirmed in beef cattle through neonatal vitamin A administration. Furthermore, we also observed enhanced marbling fat in steers derived from neonatal calves supplemented with vitamin A. In the following year, we will further define mechanisms regulating maternal impacts on fetal and neonatal muscle development.

**Wyoming Station:**

1. Completed how insulin and T3 affects titin isoform switch in a RBM20-dependent manner in Rat skeletal muscle.
2. Investigated how insulin and T3 affects different regions of titin splicing in skeletal muscle and potential molecular signaling pathway behind.
3. **Completed titin splicing pattern in different titin band in different type of muscles (Collaboration with NC State)**

**Objective 2: *Characterize the cellular and molecular basis of myogenesis***

**Alabama Station:**

Impact of *in ovo* thermal manipulation on broiler chicken muscle development, growth, and satellite cell activity.

1. Completed the live animal growth performance and carcass yield data collection and sample collection and cryohistology and immunofluorescence analysis portions of the project.
2. Immunofluorescence data analysis is ongoing.
3. Presented an abstract of growth performance and carcass yields at the 2018 Poultry Science Association annual meeting.

Effects of dietary amino acid density on growth performance, satellite cell activity, collagen gene expression, and the incidence of wooden breast.

1. Completed the live animal growth performance and carcass yield data collection, sample collection, and data analysis portions of the project.
2. Presented an abstract with collagen expression results at the 2018 Poultry Science Association annual meeting.
3. Published the results in a peer-reviewed journal, *Poultry Science*.

**Indiana Station:**

1. We demonstrated that Shisa2 regulates the fusion of muscle progenitors.
2. Demonstrated transdifferentiation of muscle satellite cells to adipocytes.

**Michigan Station:**

1. Examination of transcript abundance and DNA methylation patterns in longissimus dorsi muscle at two stages of pig fetal development revealed that differential methylation is enriched in gene regulatory regions and impacts genes associated with skeletal muscle development.
2. Turkey hatchlings from a slow-growing random-bred control line (RBC2) and fast-growing line selected from RBC2 for 16-week body weight (F line) were brooded for 3 days at one of 3 temperatures: control (35°C), cold (31°C), or hot (39°C). Samples of the pectoralis major were harvested and subjected to RNA deep sequencing to determine whether genes were differentially expressed as a function of temperature.

**Nebraska Station:**

1. Myoblasts from IUGR fetal sheep induced by maternal hyperthermia exhibit intrinsic deficits in proliferation and differentiation *ex vivo*. Using the traditional model of hyperthermia-induced placental insufficiency (PI) to produce IUGR fetuses in sheep, we found that PI-IUGR fetal myoblasts exhibited lower proliferation rates after three days in complete growth media than control fetal myoblasts whether incubated in the presences or absence of inflammatory cytokines. Likewise, fewer PI-IUGR fetal myoblasts were positive for the early differentiation marker Myogenin or the late differentiation marker desmin than control fetal myoblasts.
2. Myoblasts from IUGR fetal sheep induced by maternofetal inflammation exhibit greater proliferation but intrinsically reduced differentiation *ex vivo*. In a new model of sustained LPS-induced maternofetal inflammation (MI) to produce IUGR, we found that MI-IUGR fetal myoblasts proliferated at greater rates after 3 days in complete growth media. Conversely, fewer MI-IUGR fetal myoblasts were positive for the differentiation markers Myogenin or desmin than control fetal myoblasts.
3. Inflammatory adaptations that enhance TNFα and TLR4 signaling pathways in IUGR fetal muscle play a role in limiting myoblast function, particularly differentiation.
4. Inflammatory adaptations alone likely do not explain all myoblast dysfunction in the IUGR fetus, as two experimental models for IUGR in sheep had similar effects on myoblast differentiation, insulin sensitivity, and muscle growth, but differing effects on myoblast proliferation.

**North Carolina Station:**

1. Increased myostatin expression, resulting in muscle loss, has been associated with hyperammonemia in mammalian models of cirrhosis.

**Ohio Station:**

**Effect of Thermal Stress on In Vivo Breast Muscle (with Michigan Station)**

1. Poultry selected for growth have an inefficient thermoregulatory system and are more sensitive to temperature extremes.
2. Satellite cells are precursors to skeletal muscle and mediate all post hatch muscle growth. Their physiological functions are affected by temperature.
3. The objective of the current study was to elucidate the effects of continuous heat exposure the first 2 wk. of age on breast muscle development in broilers
4. Results showed a high level of sensitivity in the satellite cells during the early post hatch period to chronic heat, leading to impaired myogenicity and increased fat.
5. Growth selected turkeys respond to thermal stress through changes in genes predicted to downstream effects on muscle growth. Slower growing turkeys respond to thermal stress through the modulation of lipid related genes suggesting a reduction in lipid storage, transport, and synthesis. These changes are consistent with energy metabolisms required to maintain body temperature.

**Reduction in the Wooden Breast Myopathy (with Arkansas Station)**

1. Male Cobb500 broilers were fed Quatum Blue. Quatum Blue improved feed conversion and breast weight. Only high doses of Quantum Blue reduced the severity of wooden breast.
2. Reduced oxygen status altered the fate of the adult myoblasts and was accompanied by a loss of muscle repair.

**Wyoming Station:**

The role of RBM20 in regulation of skeletal muscle regeneration after injury.

1. Completed muscle injury model in WT and RBM20 KO rats and investigated whether deficiency of RBM20 affects skeletal muscle regeneration. This work has been performed in rat model.
2. Completed how RBM20 deficiency affect skeletal muscle regeneration after injury in Rat model. The manuscript is in preparation.

**Objective 3: *Characterize mechanism of protein assembly and degradation in skeletal muscle***

**Utah Station:**

1. Determination of mechanism through which decreased plane of nutrition in second trimester alters end-product quality of offspring in beef cattle
   1. Analysis of mRNA expression to different muscle fiber types of the offspring at the beginning of the feedlot phase and at harvest was completed.
   2. Previously, we observed changes in miRNA expression in these samples. We are currently analyzing mRNA expression of the downstream targets of these miRNA.
2. Gained insight into the molecular mechanism responsible for development of beef tenderness during aging.
   1. Samples were collected from the *longissimus dorsi* of steaks that had been aged for 14 days. Samples were then analyzed for tenderness with WBSF methods.
   2. Protein expression of HSPβ1, PARK7 and HSP70 was analyzed in 100 samples that vary in tenderness.
   3. An animal trial was conducted to determine the impact of pre-mortem stress on development of meat quality. We are currently analyzing these samples.

**Impact Statements:**

1. Committee members gained insight into how various environmental factors, such as diet and temperature, impact skeletal muscle growth in poultry, livestock, and companion animal species. This includes an improved understanding of how supplemental amino acids, nutraceuticals, and vitamins alters metabolism within both the skeletal muscle and adipose of these species. The committee also increased our current knowledge regarding the effects that different in utero environments have on the fetal and postnatal skeletal muscle growth of the resultant offspring. These results provide novel insights that can be used to improve growth of skeletal muscle in meat animals and performance animals. These data also inform the mechanisms that underlie dysfunctional skeletal muscle conditions such muscular dystrophy so that mitigating therapeutics can be developed. Lastly, these findings will improve the precision of management practices in poultry, dairy, and beef cattle in order to increase overall productivity and efficiency.
2. Committee members investigated different signaling pathways that are responsible for altering the growth of skeletal muscle at a molecular level in several different species in health and disease. An improved understanding of the molecular determinants of skeletal muscle growth will provide new opportunities to maximize skeletal muscle growth and efficiency in livestock and performance animals and will help to reduce the deleterious impact of dysfunctional muscle growth and development by targeting specific signaling molecules.
3. Committee members have redoubled their efforts to conduct collaborative research to better leverage the resources and expertise of the individual members on the complex and dynamic objectives outlined by this group. These efforts have yielded grants, publications, and datasets that have contributed to these objectives and are outlined below.

**Collaborative Research between Stations**

1. The Ohio and Michigan Stations are collaborating on the effect of thermal stress on in vivo breast muscle growth and development.
2. The Ohio Station is also collaborating on the reduction of wooden breast myopathy with the Arkansas Station.
3. The Mississippi Station is collaborating with the Alabama station on the effect of protein source on muscle and adipose development in weanling pigs.
4. The North Carolina Station is collaborating with the Wyoming station on characterization and RBM20 regulation of the Z-band and M-band titin splicing in striated muscles across species, muscle types and during development and RBM20 regulation of TTN novex specific exons and splicing variation across species and their functional role in cardiomyopathies.

**Collaborative Publications between Stations:**

1. Chen, Z., R. Maimaiti, C. Zhu, H. Cai, R. A. Stern, P. Mozdziak, Y. Ge, S. P. Ford, P.W. Nathanielsz, and W. Guo, 2018. Comprehensive characterization and RBM20 regulation of the Z-band and M-band titin splicing in striated muscles across species, muscle types and during development. Journal of Cellular Biochemistry (In Press).
2. Chen, Z., J. Song, L. Chen, C. Zhu, H. Cai, M. Sun, R. A. Stern, P.E. Mozdziak, and W. Guo, 2018. RBM20 regulation of TTN novex specific exons and splicing variation across species and their functional role in cardiomyopathies. Genes (In Press).
3. Maaenia, Y., M, S. B. Leeb, I.H. Ismaila, P. Mozdziak, and Y. S., Kim, 2018. Cloning of Japanese quail (Coturnix Japonica) follistatin and production of bioactive quail follistatin288 in E. coli
4. Barnes, N.E., Mendoza, K.E., Strasburg, G.M., Velleman, S.G., and Reed, K.M. 2018. Thermal challenge alters the transcriptional prolife of the breast muscle in turkey poults. Poult. Sci. (in press).
5. Flees, J., Greene, E., Anthony, N., Rochell, S., Kidd, M.,Walk, C., Velleman, S., Dridi, S. 2018. Quantum blue supplementation redues the severity of woody breast myopathy in broiler chicks. Poult. Sci. 97: (E-suppl. 1).
6. Zhao L, McMillan R, Xie G, Won S, Baumgard L, El-Kadi S, Selsby JT, Ross JW, Gabler NK, Hulver M, and Rhoads R. Heat stress decreases metabolic flexibility in skeletal muscle of growing pigs. American Journal of Physiology – Regulatory, Integrative, and Comparative Physiology. In Press.

1. Abuajemieh M, Kvidera SK, Mayorga EJ, Kaiser AR, Lei SM, Seibert JT, Horst EA, Sanz Fernandez VM, Ross JW, Selsby JT, Keating AF, Rhoads RP, and Baumgard LH. The effect of recovery time from heat stress on circulating bioenergetics variables and biomarkers of leaky gut. Journal of Animal Science. In Press.
2. Ganesan S, Brownstein A, Pearce S, Hudson M, Gabler NK, Baumgard L, Rhoads R, and Selsby JT. Prolonged environment-induced hyperthermia alters autophagy in oxidative skeletal muscle from Sus scrofa. Journal of Thermal Biology. In Press.
3. Seelenbinder KM, Zhao LD, Hanigan MD, Hulver MW, McMillan RP, Baumgard LH, Selsby JT, Ross JW, Gabler NK, and Rhoads RP. Effects of heat stress during porcine reproductive and respiratory syndrome virus infaction on metabolic responses in growing pigs. Journal of Animal Science. 96:1375-1387, 2018.
4. Ganesan S, Pearce S, Gabler NK, Baumgard LH, Rhoads RP, and Selsby JT. Short-term heat stress results in increased apoptotic signaling and autophagy in oxidative skeletal muscle. Journal of Thermal Biology. 72:73-80, 2018.
5. Ganesan S, Summers CM, Pearce SC, Gabler NK, Baumgard LH, Rhoads RP, Valentine RJ, and Selsby JT. Short-term heat stress altered metabolism and insulin signaling in skeletal muscle. Journal of Animal Science. 96:154-167, 2018.

**Collaborative Teaching Efforts between Stations**

Members of the NC1184 group including Dr. Sandy Velleman (Ohio), Marcia Hathaway (Minnesota), Dave Gerrard (Virginia), Sally Johnson (Virginia), and Derris Burnett (Mississippi) collaborate to team-teach an advanced animal growth and development class in the spring semester of each year. This unique delivery method arose directly from the NC1184 group and continues through the efforts and interactions of the NC1184 members.

**Grants and Contracts:**

**Alabama Station:**

Grant Title: Effect of dietary protein source and litter condition on broiler chicken growth performance and muscle stem cell activity.

Funding Agency: Auburn University College of Agriculture Internal Hatch and Multistate Competitive Funding Program.

Amount: $50,000

Duration: Oct. 2018 to Sept. 2020

PD (J. Starkey)

Grant Title: Role of Muscle Stem Cells in the Wooden Breast Chicken Meat Quality Defect.

Funding Agency: USDA-NIFA AFRI Foundational Grant Program (CRIS Number: 1014981).

Amount: $447,675

Duration: Mar. 2018 to Feb. 2021

PD (J. Starkey)

Co-PD (C. Starkey)

Grant Title: Effect of feed enzyme (ADM Empirical NSP Mixer) inclusion on broiler chicken growth performance and carcass parts yield.

Funding Agency: ADM Animal Nutrition

Amount: $85,129

Duration: June 2018 to May 2019

PD (C. Starkey)

Co-PD (J. Starkey)

Grant Title: Investigating Rovabio® Advance and Phytase Interactions in Broiler Diets with Reduced Nutrient Density on Broiler Chicken Growth Performance, Carcass Parts Yield, and Bone Mineralization.

Funding Agency: Adisseo, Inc.

Amount: $86,867

Duration: Jan. 2018 to Dec 2018

PD (C. Starkey)

Co-PD (J. Starkey)

Grant Title: Effect of combining maternal and post-hatch 25-hydroxycholecalciferol (Hy·D®) supplementation on broiler chicken skeletal muscle developmental characteristics, satellite cell mitotic activity, growth performance, and carcass yield.

Funding Agency: DSM Nutritional Products, Inc.

Amount: $272,268

Duration: Jan. 2018 to Dec. 2020

PD (J. Starkey)

Co-PD (C. Starkey and J. Wilson)

**Connecticut Station:**

Grant Title: The effects of nutrient restriction and realimentation on offspring liver and muscle growth and metabolism

Funding Agency: USDA/AFRI

Amount: $489,826

Duration: 01/01/2017-12/31/2021

Role: co-PD (PD K. Govoni, Co-PD S. Zinn, K. Vonnahme)

**Indiana Station:**

Grant Title: Polymeric nanoparticles for inductive browning of white adipose tissue

PI: M. Deng (Subaward PI: S. Kuang)

Funding Agency: NIH/NIDDK R43DK115277

Duration: 07/06/2018 – 07/05/2019 ($198,323)

Grant Title: Notch signaling in liposarcoma

PI: S. Kuang

Funding Agency: NIH/NCI R01CA212609

Amount: $1,937,500

Duration: 06/01/2017 – 05/30/2022

Grant Title: Engineering a biomaterial niche of satellite cells for skeletal muscle regeneration

PI: M. Deng (co-PI: S. Kuang)

Funding Agency: NIH/NIAMS R03AR068108

Amount: $226,640

Duration: 06/01/2016 – 05/31/2019

**Wyoming Station:**

Neogen GeneSeek Corp. Donation Funds for Research Support,

Role: W. Guo (PI)

Amount: $56,000

Duration: 2/28/2018-3/1/2020

UW-College Collaborative Grant,

Funding Agency: NIH-Wyoming INBRE; NIH P20 NIGMS 103432,

Role: W. Guo (co-PI)

Amount: $50,000 ($25,000 to W.Guo)

**Ohio Station:**

Title: Influence of Thermal Challenge on Turkey Muscle Development and Meat Quality.

Role: G.M. Strasburg (PD), W.D. Atchison (co-PD), K.M. Reed (co-PD), S.G. Velleman (co-PD).

Funding Agency: USDA NIFA AFRI. $975,000.

Amount: $975,000

Duration: March 15, 2014 - March 14, 2018 (Extended to March 14, 2019)

Title: Integrated analysis of genetic marker, mRNA and miRNA data to unravel mechanisms controlling growth and meat quality traits in pigs.

Role: C.W. Ernst (PD), J.P. Steibel (Co-PD), R.O. Bates (Co-PD), S.H. Zhao (Co-PD).

Funding Agency: USDA NIFA AFRI.

Amount: $499,985.

Duration: January 1, 2014 – December 31, 2017 (Extended to December 31, 2018).

Title: Functional annotation of the porcine genome.

Role: C.K. Tuggle (PD), H. Zhou (Co-PD), T.P.L. Smith (Co-PD), C.W. Ernst (Co-PD), J.M. Reecy (Co-PD), J.E. Koltes (Co-PD), P.J. Ross (Co-PD), D.J Nonneman (Co-PD), C.R. Loving (Co-PD), J.P. Steibel (Co-PD), W. Huang (Co-PD).

Funding Agency: USDA NIFA AFRI.

Amount: $2,500,000.

Duration: February 1, 2018 – January 31, 2022.

**South Dakota Station:**

Title: Growth management strategies to optimize feedlot cattle production

Funding Agency: Hatch

Amount: $128,000

Role: (PD) Zachary Smith

Duration: 10/15/18 to 10/14/21

**Illinois Station:**

Title: Internal and external validation of a porcine dystrophinopathy model

Role: J. Selsby (PD)

Funding Source: 1R21NS106112-01

Amount: $765,000

Duration: 5/2018-5/2020

Title: Therapeutic approaches to heat stress: targeting mitochondria.

Role: J. Selsby (PD), L. Baymgard, and R. Rhoads

Funding Source: USDA 2017-05931

Amount: $497,000

Duration: 1/2018-12/2021

**Minnesota Station:**

Title: Homeostatic Responses to Amino Acid Insufficiency

Define the contribution of the integrated stress response to the early molecular and physiological responses that function to maintain proteostasis during dietary amino acid insufficiency.

Funding Agency: NIH/NIDDK R01DK109714-01A1

Role: Lead PI

Duration: 01/01/17 – 12/31/21

Title: The major goal of this project is to examine the impact of PERK deletion on the metabolic phenotype associated with dietary methionine restriction.

Funding Agency: NIH/NIDDK R01DK096311-05A1

Role: Subcontract PI

Duration: 04/01/2017 – 03/31/2019

**Mississippi Station:**

Grant Title: Determining the temporal and spatial regulation of marbling development in the longissimus muscle of porcine offspring from weaning through finishing.

Funding Agency: National Pork Board

Role: PI

Duration 5/2018-5/2019

**Utah Station:**

Grant Title: Establishing a protocol for receiving cattle that are at-risk of having a mineral deficiency

Funding Agency: USDA-WSARE

Amount: $206,208

Duration: 06/18-05/21

Role: PD

Grant Title: Effects of pre-mortem stress on heat shock protein expression and oxidation relative to meat quality

Funding Agency: USDA-NIFA Seed

Amount: $149,879

Duration: 04/18-03/20

Role: PD

Grant Title: Elucidation of the molecular mechanism through which trenbolone acetate and estradiol improve skeletal muscle growth in beef cattle.

Funding Agency: USDA-NIFA

Amount: $490,000

Duration: 01/18-12/21

Role: PD

Grant Title: Understanding how a novel alfalfa product impacts production of beef cattle, dairy heifers, and milking cows

Funding Agency: private company

Amount: $89,185

Duration: 01/18-12/20

Role: PD

**Illinois Station:**

Grant Title: Muscle Proteomic Approach to Characterize Biochemical Mechanisms responsible for Woody Breast and Pale, Soft and Exudative Conditions in Broilers.

Funding Agency: USDA-NIFA

Amount: $454,986

Duration: 2017-2021

Role: M.W. Schilling (PI), W. Zhai (co-PI), S. Suman (co-PI), A. Dilger (co-PI). $454,986.

2017-2021.

Grant Title: Defining and mitigating the effects of maternal infection and piglet muscle and immune system development.

Funding Agency: USDA-NIFA

Amount: $500,000

Duration: 2018-2022

Role: A. Dilger (PI), R. Dilger (co-PI), D. Boler (co-PI), J. Beever (co-PI).

Grant Title: Title: Characterizing the amount, variability and cellular mechanisms of intramuscular fat deposition throughout the loin using barrows and gilts from two genotypes.

Funding Agency: National Pork Board

Amount: $99,869

Duration: 2018-2019

Role: D. Boler (PI), A. Dilger (co-PI), J. Beever (co-PI), C. Stahl (co-PI).

**Publications (peer reviewed journals):**

1. Meloche, K. J., W.A. Dozier, III, and J. D. Starkey. 2018. Skeletal muscle growth characteristics and myogenic stem cell activity in broiler chickens affected by wooden breast. Poult. Sci. Accepted 6/10/18. doi: 10.3382/ps/pey287.
2. Jones, M. K., K. E. Richardson, C. W. Starkey, N. M. Dale, and A. J. Davis. 2018. Impact of extended heat treatment on the amino acid digestibility and TMEn content of a formaldehyde-treated diet. J. Appl. Poult. Sci. <https://doi.org/10.3382/japr/pfy038>
3. Jones AK, Hoffman ML, Pillai SM, McFadden KK, Govoni KE, Zinn SA, Reed SA. 2018. Gestational restricted- and over-feeding promote maternal and offspring inflammatory responses that are distinct and dependent on diet in sheep. Biol Reprod. 98:184-196. doi: 10.1093/biolre/iox174.
4. Jones AK, Hoffman ML, Pillai SM, McFadden KK, Govoni KE, Zinn SA, Reed SA. 2018. Gestational restricted- and over-feeding promote maternal and offspring inflammatory responses that are distinct and dependent on diet in sheep. Biol Reprod. 98:184-196. doi: 10.1093/biolre/iox174.
5. Farzaneh, M., F. Attari, S. E. Khoshnam, and P. E. Mozdziak, 2018. Chicken whole embryo culture system, the strong in-vitro model. British Poultry Science (In Press).
6. Zand-vakili, M., A. Golkar-Narenji, H. Eimani, and P. E. Mozdziak, 2017. An in vitro study on cumulus oocyte complexes COCs and follicles of autotransplanted mouse ovaries with the effect of VEGF. Journal of the Turkish German Gynecological Association 18: 167-173.
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10. Farzaneh, M, S., N. Hossani, P.E. Mozdziak, and H. Baharvand. 2017. Avian embryos and related cell lines: a convenient platform for recombinant proteins and vaccines production. Biotechnology Journal 12: DOI 10.1002/biot.201600598/
11. Stern, R. A., S. Dasarathy, and P.E. Mozdziak, 2017. Ammonia elicits a different myogenic response in avian and murine myotubes. In Vitro Cell Dev. Biol. Animal. 53: 99-110.
12. Bazgir, B., R. Fathei, M. Rezazadeh Valoujerd, P. Mozdziak, and A. Asgari, 2017. Satellite cell contribution to cxercise mediated muscle hypertrophy and repair: A review article. Cell Journal 18: 473-484.
13. Chen, Z., R. Maimaiti, C. Zhu, H. Cai, R. A. Stern, P. Mozdziak, Y. Ge, S. P. Ford, P.W. Nathanielsz, and W. Guo, 2018. Comprehensive characterization and RBM20 regulation of the Z-band and M-band titin splicing in striated muscles across species, muscle types and during development. Journal of Cellular Biochemistry (In Press).
14. Chen, Z., J. Song , L. Chen, C. Zhu, H. Cai , M.Sun, R. A. Stern, P.E. Mozdziak , and W. Guo, 2018. RBM20 regulation of TTN novex specific exons and splicing variation across species and their functional role in cardiomyopathies. Genes (In Press).
15. Maaenia, Y., M, S. B. Leeb, I.H. Ismaila, P. Mozdziak, and Y. S., Kim, 2018. Cloning of Japanese quail (Coturnix Japonica) follistatin and production of bioactive quail follistatin288 in E. coli
16. Barnes, N.E., K.M. Mendoza, G.M. Strasburg, S.G. Velleman and K.M. Reed. 2018. Thermal challenge alters the transcriptional profile of the breast muscle in turkey poults. Poultry Science. doi: 10.3382/ps/pey401.
17. Daza, K.R., J.P. Steibel, D. Velez-Irizarry, N.E. Raney, R.O. Bates and C.W. Ernst. 2017. Profiling and characterization of a *longissimus dorsi* muscle microRNA dataset from an F2 Duroc x Pietrain pig resource population. Genom. Data. 13:50-53.
18. Reed, K.M., K.M. Mendoza, G.M. Strasburg and S.G. Velleman, 2017. Response of turkey muscle satellite cells to thermal challenge. II. Transcriptome effects in differentiating cells. Frontiers in Physiology. 8:948.
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22. Wang, B., C. L. Harris, W. Nie, X. Fu, J. M. Deavila, M. J. Zhu, M. Maquivar, S. M. Parish, J. R. Busboom, M. L. Nelson, and M. Du. (2018). Neonatal vitamin A injection promotes cattle muscle development and increase oxidative muscle fibers. *Journal of Animal Science and Biotechnology*, In press.
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24. Ma, Y.N., B. Wang, Z.X. Wang, N. A. Gomez, M. J. Zhu, and M. Du. (2018). Three dimensional spheroid culture of adipose stromal vascular cells for studying adipogenesis in beef cattle. *Animal*, 12: 2123-2129.
25. Zhao, L., T. Zou, N. A. Gomez, B. Wang, M. J. Zhu, and M. Du. (2018). Raspberry alleviates obesity-induced inflammation and insulin resistance in skeletal muscle through activation of AMP-activated protein kinase (AMPK) α1. *Nutrition & Diabetes*, 8: 39.
26. Zhang, S., Y. Zhang, X. Zhou, X. Fu, J. Michal, G. Ji, M. Du, and Z. Jiang. (2018). Alternative polyadenylation drives genome-to-phenome information detours in the AMPKα1 and AMPKα2 knockout mice. *Scientific Report,* 8: 6462.
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36. Wang, B., X. Fu, X. Liang, J. M. Deavila, Z. Wang, L. Zhao, Q. Tian, J. Zhao, N. A. Gomez, S. C. Trombetta, M. J. Zhu, and M. Du. (2017). Retinoic acid induces white adipose tissue browning by increasing adipose vascularity and inducing beige adipogenesis of PDGFRa+ adipose progenitors. *Cell Discovery*, 3:17036.
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38. Gao, P. F., X. H. Guo, M. Du, G. Q. Cao, Q. C. Yang, Z. D. Pu, Z. Wang, Q. Zhang, M. Li, Y. S. Jin, X. J. Wang, H. Liu, and B. G. Li. (2017). LncRNAs profiling of skeletal muscles in Large White pigs and Mashen pigs during development. *Journal of Animal Science*, 95: 4239-4250.
39. Wang, B., Z. Wang, J. M. de Avila, M. J. Zhu, F. Zhang, N. A. Gomez, L. Zhao, Q. Tian, J. Zhao, J. Maricelli, H. Zhang, B. D. Rodgers,andM. Du. (2017). Moderate alcohol intake induces thermogenic brown/beige adipocyte formation via elevating retinoic acid signaling. *FASEB Journal*, 31: 4612-4622.
40. Wang, B., X. Fu, M.J. Zhu, and M. Du. (2017). Retinoic acid inhibits white adipogenesis by disrupting GADD45A mediated Zfp423 DNA demethylation. *Journal of Molecular Cell Biology*, 9: 338-349.
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**Book Chapters:**

None

**Abstracts, Posters, and Professional Presentations:**

1. Ferreira, T. Z., L. Kindlein, K. J. Meloche, S. Vieira, V. Nascimento, and J. D. Starkey. 2018. Characterization of myogenic stem cell populations in broiler chickens affected with the Wooden Breast myopathy. Poult. Sci. Vol. 97 (E-Suppl 1).
2. Starkey, J. D., C. W. Starkey, and A. Morey. 2018. Integration of student e-portfolio use into the Auburn University poultry science curriculum. Poult. Sci. Vol 97 (E-Suppl 1).
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4. C. W. Starkey, R. B Shirley, A. L. Welsher, O. J. Tejeda, L. F. Spencer, and J. D. Starkey. 2018. Effect of dietary protein source and bacillus subtilis probiotic (Alterion®) supplementation on growth performance and meat yield of broiler chickens reared to 46 days of age. Poult. Sci. Vol. 97 (E-Suppl 1).
5. J. E. Powell, O. J. Tejeda*,* J. D. Starkey, and C. W. Starkey. 2018. Effect of particle size and proportion of fines in growth performance of broiler chickens reared to 21 days of age. Poult. Sci. Vol. 97 (E-Suppl 1).
6. L. F. Spencer, A. J. Calderon, O. J. Tejeda, K. Estes, B. Barton, T. Powell, J. D. Starkey, and C. W. Starkey. 2018. Effect of increased dietary choline chloride concentrations on growth performance and carcass characteristics of broiler chickens reared to 32 days of age. Poult. Sci. Vol. 97 (E-Suppl 1).
7. A. J. Calderon, L. F. Spencer, O. J. Tejeda, K. Estes, B. Barton, T. Powell, C. W. Starkey, and J. D. Starkey. 2018. Effect of increased dietary choline chloride concentrations on growth performance and carcass characteristics of broiler chickens reared to 66 days of age. Poult. Sci. Vol. 97 (E-Suppl 1).
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9. Martin, D.E., A.K. Jones, M.L. Hoffman, S.M. Pillai, K.E. Govoni, S.Z. Zinn, and S.A. Reed. 2018. Effects of poor maternal nutrition during gestation on the offspring muscle metabolome.
10. Govoni, K.E., S.A. Reed, and S.A. Zinn Cell Biology Symposium: Poor maternal nutrition during gestation alters whole body and cellular metabolism in offspring
11. Reed, S.A. Meat Science and Muscle Biology Symposium: Roles for inflammation in livestock muscle growth and repair.
12. Iannitti, H.R., A. K. Jones, M. L. Hoffman, S. M. Pillai, K. E. Govoni, S. A. Zinn, and S. A. Reed. Effects of poor maternal nutrition during gestation on offspring oxidative stress
13. Maimaiti Rexiati, Mingming Sun and Wei Guo, RBM20 Deficiency Postpones Skeletal Muscle Regeneration after Injury and Promotes Fibrotic Tissue Formation. EB Meeting. 2018. Abstract 5067. Poster A676.
14. Maimaiti Rexiati, Mingming Sun and Wei Guo, RBM20 Modulates Myofiber Maturation and Skeletal Muscle regeneration. Myofilament Meeting, Madison, Wisconsin, 2018. Abstract and Poster 40. Page 123
15. Krieger, J. A. Yesinovskiy, P. Mozdziak, and M. Kim, 2018. Developing scaleable production methods for cultured pork and turkey meat: beyond. Proceedings of the Recprocial Meat Conference.
16. Corbett, R.J., N.E. Raney and C.W. Ernst. 2018. Transcriptional and genome-wide methylation profiling in fetal pig skeletal muscle. Plant and Animal Genome XXVI Conference. San Diego, CA. https://pag.confex.com/pag/xxvi/meetingapp.cgi/Paper/28250. Invited Oral Presentation.
17. Daza, K., D. Velez-Irizarry, S. Casiro, J.P. Steibel, N.E. Raney, R.O. Bates and C.W. Ernst. 2018. Genomic co-localization of microRNA expression QTL target genes with phenotypic QTL in the Michigan State University Duroc x Pietrain pig resource population. Plant and Animal Genome XXVI Conference. San Diego, CA. https://pag.confex.com/pag/xxvi/meetingapp.cgi/Paper/30021.
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22. Dylan J. Klein, Emily T. Mirek, Tracy G. Anthony and Kenneth H. McKeever (2018) Exercise Training in Standardbred Horses Alters the Skeletal Muscle Metabolome and Plasma Amino Acid Profile: Implications for the “Athlete’s Paradox”. Abstract Number: 855.27. Published: April 20, 2018. https://www.fasebj.org/doi/abs/10.1096/fasebj.2018.32.1\_supplement.855.27
23. Tracy G. Anthony, "Dietary Sulfur Amino Acid Restriction (SAAR) and the Integrated Stress Response: Role of eIF2 kinases versus ATF4” presented on August 7, 2018, at the FASEB Science Research Conference entitled, Nutrient Sensing and Metabolic Signaling.
24. Tracy G. Anthony, “Exercise and the Integrated Stress Response”, presented in the session on Exercise and Energy Restriction to Improve Health: The Crossroads of Energetics and Protein Turnover at the ACSM Conference on Integrative Physiology of Exercise, San Diego, CA on Sept 7, 2018.
25. Tracy G Anthony, “Dietary Sulfur Amino Acid Restriction and the Integrated Stress Response”, presented at the Department of Nutritional Sciences Seminar Series at Rutgers University, Sept 12, 2018
26. Tracy G Anthony, “Dietary Sulfur Amino Acid Restriction and the Integrated Stress Response”, presented at Iowa State University, Department of Animal Sciences, Sept 26, 2018.
27. Yates DT - Nebraska Sheep & Goat Producers Association, 2018. Impact of Maternal Stress on muscle growth & metabolism in the fetus & offspring.
28. Cadaret CN - Western Section, American Society of Animal Science, 2018. Sustained maternal inflammation during the early third trimester yields fetal adaptations that impair subsequent skeletal muscle growth and glucose metabolism in sheep.
29. Posont RJ - Western Section, American Society of Animal Science, 2018. Changes in myoblast responsiveness to TNF⍺ and IL-6 contribute to decreased skeletal muscle mass in intrauterine growth restricted fetal sheep. Translational Animal Science.
30. Barnes TB - Western Section, American Society of Animal Science, 2018. Impaired muscle stem cell function in cows with high concentrations of androstenedione in their follicular fluid.
31. Kubik RM - Western Section, American Society of Animal Science, 2018. Investigation of the skeletal muscle transcriptome in lambs fed β adrenergic agonists and subjected to heat stress for 21 d.
32. Duffy EM - Western Section, American Society of Animal Science, 2018. Rumen bacterial composition in lambs is affected by β adrenergic agonist supplementation and heat stress at the phylum level.
33. L T Honegger, B N Harsh, J E Beever, D Boler, A C Dilger; (2018) Comparative Analysis of the Porcine IGF2-G3072A Mutation and Reduced Myostatin Function on Carcass and Meat Quality Characteristics. Journal of Animal Science, Volume 96 (Supplement 2) 105–106. Midwest Meeting of ASAS and ADSA, Omaha, NE. March 2018.

**Theses and Dissertations:**

1. “Fit As A Horse: From Skeletal Muscle Metabolism to Whole Body Physiology", a Doctoral Dissertation by Dylan Joseph Klein submitted to the Rutgers University School of Graduate Studies with a 2 year embargo on publication to allow for research articles currently under preparation to be published first.
2. "Role of ATF4 in Dietary Sulfur Amino Acid Restriction", a Master’s Thesis by Nicholas Margolies submitted to the Rutgers University School of Graduate Studies with a 2 year embargo on publication to allow for research articles currently under preparation to be published first.
3. Xiaoxing Xu, Plan A MS degree (2016 Spring – 2018 Summer), “The effect of neonatal administration of recombinant myostatin propeptide on skeletal muscle growth in mice.”
4. Taylor Barnes. 2018. MS Thesis: “Stress and other factors and their effect on skeletal muscle growth and metabolism; Strengths-based lab groups improve learning of undergraduate anatomy and physiology concepts.”
5. Rachel Kubik. 2018. MS Thesis: “Genomic investigation of beta agonist supplementation and heat stress in livestock species.”
6. Jessica Lancaster. 2018. MS Thesis: “Utilization of Depth - Enabled Identification and Tracking System to Identify and Track Individual Pigs and Analyze Individual Pig Activity.”
7. Lauren Kett. 2018. MS Thesis: “Evaluation of the Interaction of Beta-Adrenergic Agonists Supplementation and Heat Stress on Growth Performance and Carcass Composition in Feeder Lambs.”
8. Martin, D.E. 2018. The Effects of Poor Maternal Nutrition on Offspring Muscle Metabolism. M.S. Thesis. Univ. Connecticut, Storrs, CT.
9. Bailey Harsh. PhD Thesis. Effects of ractopamine hydrochloride on nutrient digestibility, environmental nitrogen excretion, regulation of skeletal muscle growth and beta-receptor subtypes in finishing beef steers. August, 2018
10. Thompson, R. C. 2018. Effect of maternal melatonin supplementation during late gestation on programming and metabolic disposition of adipose tissue and skeletal muscle in bovine offspring. M. S. Thesis, Mississippi State University, Mississippi State, MS.