TEXAS A&M UNIVERSITY

IFRB 2013

Interdisciplinary Faculty of Reproductive Biology

Confronting and solving challenges to reproduction and health by conducting basic, clinical and translational research, and shaping science and health policy

2013, ISSUE 1 SPRING, 2013

POINTS OF INTEREST:

- The IFRB was organized in 1990 and is one of the largest Reproductive Biology Programs in the US
- Membership includes 44 faculty from 5 departments, 3 colleges , 4 TAMUS components and 2 state agencies
- IFRB sponsored activities: IFRB Repro Forum Seminar Series (22 years) Annual R.O. Berry Lecture (18 years), Texas Forum on Reproductive Sciences (19 years), Annual IFRB Retreat

INSIDE THIS ISSUE:

New IFRB Faculty Spotlight

Ashlee Watts DVM, PhD, DACVS, is the newest member of the Interdisciplinary Faculty of Reproductive Biology. Dr. Watts was recently hired to the Department of Large Animal Clinical Sciences in March of 2012 as an Assistant Professor in Equine Surgery. She founded, and is the director of the Comparative Orthopedics and Regenerative Medicine Laboratory at Texas A&M University. The laboratory is located in the new wing of the Veterinary Medical Research Building, immediately adjacent to the Large Animal Hospital at Texas A&M. The specific mission of the lab is to foster and expand multidisciplinary research and equine clinical care for orthopedic disorders and regenerative therapies. The laboratory provides for the isolation and expansion of autologous (self) mesenchymal stem cells from bone marrow for Texas A&M equine patients.

Dr. Watts received her D.V.M. degree in 2003 from Colorado State University, completed a one year internship at Pioneer Equine Hospital in Oakdale, CA and a residency in large animal surgery at Cornell University. She obtained her Ph.D. degree in 2012 from Cornell University and is a Diplomate, American College of Veterinary Surgeons, Large Animal.

Dr. Watts is an equine orthopedic surgeon at the Veterinary Medical Teaching Hospital at Texas A&M University. She works with board certified faculty members on the Equine Orthopedics, Equine Lameness and Diagnostic Imaging services at Texas A&M for the care of equine clinical patients and clinically related research. Dr. Watts collaborates with scientists at TAMU's Institute for Regenerative Medicine at

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Global Issues: Endocrine Disrupting Chemicals & Reproductive Health

Heavy Metal Endocrine Disruptors: CrVI-induced female reproductive toxicity

Heavy metal production and use started long before the industrial revolution and has increased exponentially since then. Hexavalent chromium (CrVI), one of the most toxic heavy metals, is widely used in more than 50 industries such as chrome plating, welding, wood processing and tanneries. The U.S. is one of the world's leading producers of chromium compounds and we now face great challenges in protecting human health against multiple adverse effects of CrVI. In California, 30% of drinking water sources are contaminated with significant levels of CrVI. According to the Environmental Working Group (EWG), 31 out of 36 major cities in the U.S have the drinking water contaminated with carcinogenic CrVI; and 70 million Americans consume CrVI-contaminated drinking water that can cause oral and intestinal cancers according to recent experimental research by National Toxicological Program (NTP).

The box-office hit movie **Erin Brockovich**, released in 2000 featuring Julia Roberts, is based on the biographical story of Brockovich who worked to bring litigation against the California power company Pacific Gas & Electric (PG&E). PG&E contaminated well waters and aquifers as a result of dumping 26 tons of coolant containing CrVI in Hinkley, CA. Residents reported falling sick, and some allegedly died from exposure to the known carcinogen. Breast cancer, Hodgkin's disease, lung, brain and gastrointestinal cancers, miscarriages, and kidney and ovarian tumors were blamed on the contaminated water. The case was settled in 1996 for $333 million. Interestingly, in June of 2009, Brockovich began investigating a case

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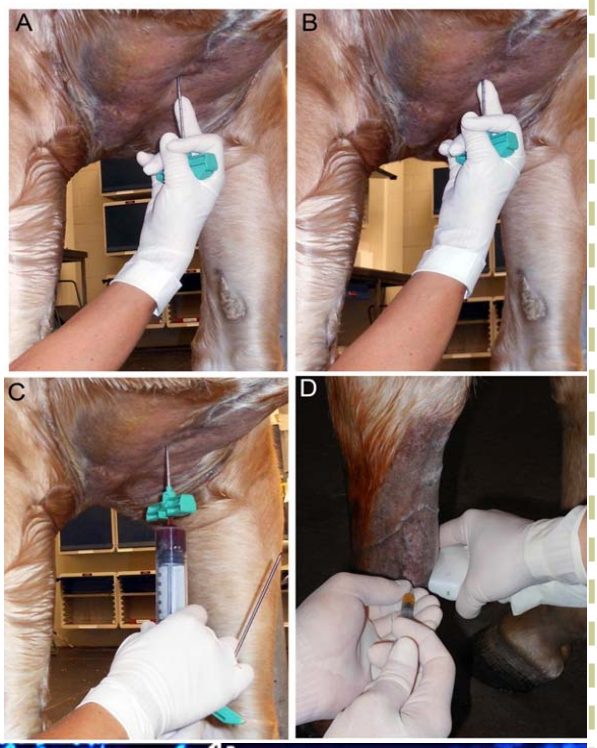
New IFRB Faculty (cont'd from page 1)

the Texas A&M Health Science Center and TAMU's Institute for Preclinical Studies for studies on the basic mechanisms of enhanced repair of orthopedic injury by adult derived stem cells. She also collaborates with veterinary practitioners in Texas and veterinary scientists at Virginia Tech, Cornell University and Colorado State University for the study of regenerative medicine. Dr. Watts' laboratory personnel include one full time technician, one post-doctoral researcher, two Undergraduate Honors students and several hourly student workers and a PhD student will start in her lab this fall.

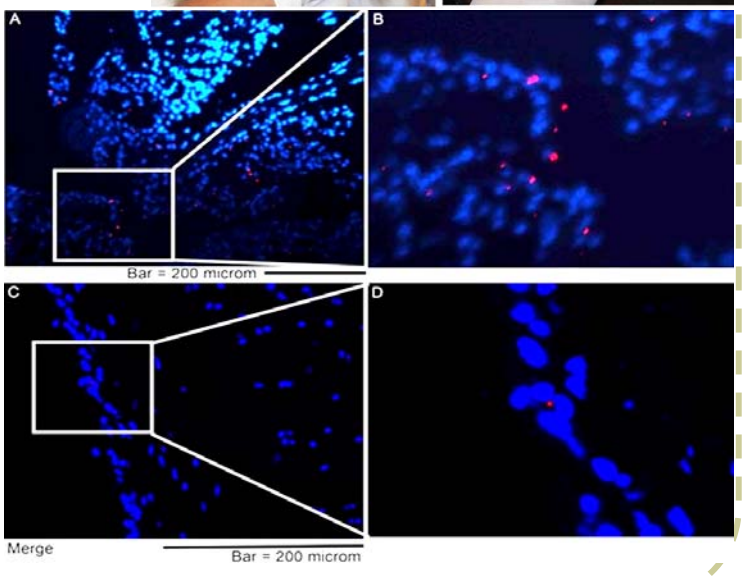
Regenerative medicine is the process of enhancing the body's own ability to heal. Regenerative medicine and stem cell therapy have been used in treating equine orthopedic injury since the late 1990s and have become a standard treatment for equine tendon injury (***see Figures at right and left***). More recently, there is increasing interest in utilizing regenerative techniques and stem cell therapy for other orthopedics injuries such as articular cartilage repair, meniscal repair, prevention of osteoarthritis progression, laminitis therapy and enhanced fracture healing. Although there are several uncontrolled reports of the efficacy of regenerative techniques and the general consensus is of improved healing, much research remains to be done. The basic mechanism(s) of enhanced repair and even the location and longevity of stem cell engraftment are still debated (***see Figure below***). Dr. Watts hopes to expand regenerative medicine research through identification of basic mechanisms of enhanced repair and ideal treatment paradigms, as well as through prospective and blinded clinical trials to better define the efficacy of treatment.

The main focus of Dr. Watts' Comparative Orthopedics and Regenerative Medicine Laboratory is to serve the equine industry and veterinary and medical communities by: engaging in orthopedic and regenerative research with immediate impact to both equine and human athletes; providing guidelines and strategies for management of equine clinical cases through incorporation of innovative regenerative and stem cell technologies; evaluating and treating orthopedic disease with stem cells and regenerative therapies in equine athletes; and educating veterinary students, veterinary residents, graduate students, and visiting scientists in equine regenerative techniques for orthopedic disease.

A colony of bone marrow derived mesenchymal stem cells at 40x magnification 9 days after bone marrow collection.

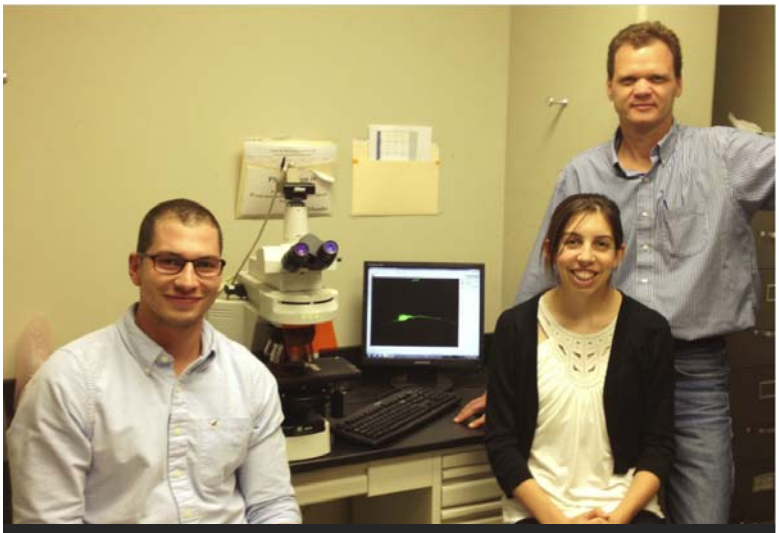


Upper Right: Bone marrow collection from the sternum of a horse (A-C) for the treatment of a deep digital flexor tendon injury by ultrasound guided intralesional injection (D).



Lower Right: Distribution of 625 nm quantum dot (Qdot) labeled mesenchymal stem cells (red) to an osteoarthritic fetlock joint synovial membrane (A, B) and articular cartilage (C, D) with a nuclear counter-stain (Hoescht). Sections are from a fetlock joint with severe osteoarthritis, one week after labeled stem cell injection. Qdot labeled cells are abundantly present in the intimal layers of the synovial membrane villi, and a single labeled cell is evident in the fibrotic surface of the OA cartilage. Images were taken at (A) 200x and (C) 400x magnification. (B, D) Expanded images from A and C denoted by the white box. Bar = 200 µm in original images

IFRB Faculty Spotlight: Dr. Marcel Amstalden



Dr. Amstalden (standing) is shown here with trainees Rodolfo Cardoso and Michelle Bedenbaugh.

Dr. Marcel Amstalden was recently elected as Chair of the Interdisciplinary Faculty of Reproductive Biology by the IFRB Executive Committee. Dr. Amstalden is an Associate Professor in the Department of Animal Science. His research program in Reproductive Neuroendocrinology contributes to solving issues of societal and economic impact such as improving reproductive efficiency in food-producing species and minimizing the impact of reproductive disorders to animals and humans. By investigating functional elements of the neuroendocrine system that are responsive to nutritional and metabolic cues during the infantile and juvenile period, Dr. Amstalden, his colleagues and trainees have advanced the understanding of mechanisms by which factors of metabolic and endocrine origin affect pubertal development and estrous cyclicity in sheep and cattle.

A major area of focus has been the characterization of mechanisms and pathways by which metabolic-sensing neurons control gonadotropin-releasing hormone (GnRH) secretion. Using an experimental model in which age at puberty is considerably reduced in early-weaned heifers fed high-concentrate diets, Dr. Amstalden, in collaboration with **Dr. Gary Williams** (Texas A&M AgriLife Research, Beeville and member of IFRB) and colleagues, have conducted vital studies in the beef heifer. Microarray technology was used to investigate the transcriptome of the arcuate nucleus, a major metabolic-sensing region of the hypothalamus, in juvenile heifers fed to gain weight at high and low rates (***see Figure***). Changes in the expression of neuropeptide Y (NPY) and proopiomelanorcortin drew particular attention as those genes are known to be involved in controlling feed intake, energy expenditure and reproductive neuroendocrine function. Subsequent studies have indicated that plasticity in neuronal projections containing NPY appears to play a role in the control of GnRH neurons in the hypothalamus in heifers that gain body weight at high rates. These observations indicated that nutrition during the juvenile period may program structural and functional hypothalamic circuitries that are important for timing reproductive maturation in females.

The role of the neuropeptide kisspeptin as stimulator of GnRH secretion has also been a significant research focus in Amstalden's laboratory. One of the physiological turning points of pubertal maturation in females is the increase in

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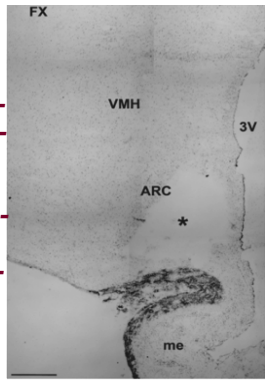


Cresyl violet-stained section through the medi-obasal hypothalamus depicting an area (asterisk) of the arcuate nucleus dissected for microarray analysis (Allen et al., 2012, J Anim Sci 90: 2222).

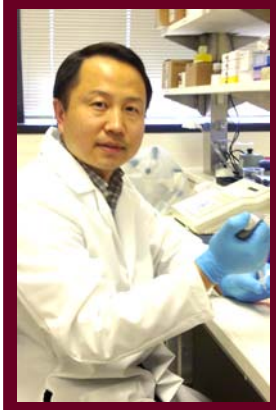
19th Annual Raymond O. Berry Memorial Lecturer Selected

Dr. Thaddeus (Ted) Golos, from the Wisconsin National Primate Research Center and School of Medicine and Public Health, and Departments of Obstetrics and Gynecology, and Comparative Biosciences, University of Wisconsin-Madison was selected by a vote of IFRB faculty in December 2012, to present the 19th Annual Dr. Raymond O. Berry Memorial Lecture which will be held in conjunction with the 7th Annual IFRB Retreat on Friday, October 11, 2013 at The Veranda in Bryan, TX.

The Golos laboratory examines questions of placental biology relevant to human health and disease, using nonhuman primate models, human clinical materials, and human and nonhuman primate embryonic stem cells in their studies. Study of the maternal-fetal immune dialogue examines nonpolymorphic MHC class I molecules expressed on placental cells and their interactions with the maternal immune system, particularly endometrial natural killer cells and macrophages in promoting pregnancy success, including placental and decidual vascularization. In addition, embryonic stem cells and primate embryos are used to model implantation and placental morphogenesis, with 2- and 3-dimensional modeling and co-culture systems for understanding how placental cells carry on a dialog with the maternal endometrium. At the interface of these projects is the direct effect of maternal immune cells on rhesus blastocyst function and growth in 3-dimensional co-culture. Maternal immune recognition of pregnancy and regulation of a healthy implantation site and placenta may play a significant role in pregnancy issues ranging from miscarriage and recurrent pregnancy loss to preeclampsia and intrauterine growth restriction. The Golos lab uses the primate model to allow direct experimentation at the maternal-fetal interface to develop experimental and therapeutic models of utero-placental function and dysfunction.

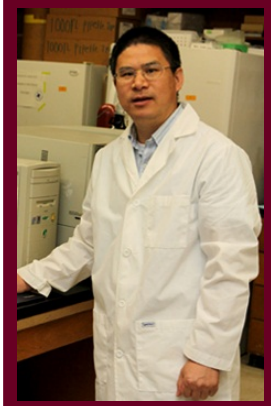
The Annual Raymond O. Barry Memorial Lecture series in honor of Dr. Raymond O. Barry, a member of the faculty from 1931 to 1960 who contributed to the establishment of the discipline of Reproductive Immunology through his pioneering studies involving embryo transfer to evaluate genetic factors affecting reproduction. The Lecture was established in 1994 by Dr. Fuller Bazer.

IFRB Faculty Awards, etc.



Dr. Qinglei Li was one o 30 young investigators selected nationwide for for the 2013 Ralph E. Powe Junior Faculty Enhancement Award.

Dr. Carey Satterfield, received the 2013 Outstanding Young Animal Scientist in Research Award from the South-



Dr. Guoyao Wu was recently named the 2013 Sigma Xi - Texas A&M Chapter Distinguished Scientist.

NEW GRANTS:

*** Guoyao Wu,** "Safety of arginine supplementation to healthy adults," International Council of Amino Acid Science, $250,000, 4/2012 to 3/2014.

*** Dr. Beiyan Zhou**, Assistant Professor, Department of Veterinary Physiology & Pharmacology, received a Junior Faculty Award from American Diabetes Association. The funding period is from January 1, 2013 to December 31, 2014.

*** Renyi Zhang, Alexi Khalizov, Fuller W. Bazer, Greg Johnson, Carey Satterfield, Guoyao Wu, Robert Burghardt**, Enhancing Teaching and Research in Health Impacts of Air Pollution," Tier One Program Grant, TAMU Dean of Faculties, $300,000, 4/1/13 to 3/31/16.

INVITED LECTURES:

*** Marcel Amstalden**, "Neuroendocrine Programming of Accelerated Puberty," Symposium on Recent Advances in the Central Control of Reproduction, Centre INRA Val de Loire, Nouzilly, France, April 11, 2013.

*** Fuller W. Bazer**, "Contributions of an Animal Scientist to Understanding Biology of the Uterus and Pregnancy," Keynote Lecture, International Embryo Transfer Society, "Hannover, Germany, January 19-23, 2013,

Plenary talk, "Roles of selected nutrients on development of the conceptus during pregnancy," 9th International Conference on Pig Reproduction Olsztyn, Poland June 9-12, 2013.

*** Robert C. Burghardt**, Microscopy and Photonics in Cancer," College of Pharmacy and Pharmaceutical Sciences, Florida A&M University, January 10, 2013.

*** Katrin Hinrichs,** "Embryo transfer, oocyte transfer and fertilization in vitro in the horse," Congress of the Sociedad Argentina de Tecnologias Embrionarias (Argentina Society of Embryonic Technologies), Corrientes, Argentina, April 12, 2013

"Assisted reproductive techniques in the horse" Annual Conference, International Embryo Transfer Society, Hannover Germany, January 20, 2013.

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IFRB International Scholars

Drs. Lagares and Stahlberg are pictured here with their children.

*** Dr. Monique de Albuquerque Lagares** is a visiting scientist from the Federal University of Minas Gerais (UFMG) in Belo Horizonte, Brazil, working with **Dr. Katrin Hinrichs** in the Equine Embryo Laboratory. Dr. Lagares research interests are in equine reproduction, mainly stallion semen and mammalian embryo cryopreservation and she is currently carrying out studies on maternal recognition of pregnancy and markers involved in its mechanism, transvaginal aspiration of ovarian follicles and in vitro fertilization of equine oocyte by the mare. She is currently funded on a one year grant from the National Council for Scientific and Technological Development (CNPq), Brazil. Dr. Lagares received her veterinary training at the Federal Fluminense University, Rio de Janeiro, Brazil and earned the PhD at the University of Veterinary Medicine, Hannover, Germany. She is Professor at the UFMG Veterinary School in Belo Horizonte and has been at the UFMG since 1999

*** Dr. Rubens Stahlberg** is a visiting scientist from the Pontifical Catholic University of Minas Gerais (PUC-Minas) in Belo Horizonte, Brazil, working with **Dr. Fuller Bazer** in the Department of Animal Science. Dr. Stahlberg is a reproductive biologist with a background in swine reproduction and management and he is currently investigating the arginine decarboxylase pathway for synthesis of agmatine that induces secretion of neuropeptides by uterine endometrium; specifically neuropeptides that are critical for development of the equine conceptus. He is funded by a one year postdoctoral grant from the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), Brazil. Dr. Stahlberg earned his veterinary degree from the Federal University do Rio Grande do Sul (UFRGS), Porto Alegre, Rio Grande do Sul, Brazil, and earned the PhD at the University of Veterinary Medicine, Hannover, Germany. He is Professor at the Veterinary School of PUC-Minas since 2001.

*** Heewon Seo, Ph.D.** recently joined the laboratories of **Drs. Greg Johnson and Kayla J. Bayless** as a postdoctoral fellow. Dr. Seo will be working on an NIH supported project to investigate how bone marrow-derived endothelial precursor cells communicate with endothelial cells and incorporate into growing blood vessels using *in vitro* model systems as well as the newly forming vasculature of pig placentae *in vivo*. He completed his doctoral studies in Korea under the direction of former IFRB trainee, **Dr. Hakhyun Ka**, who is a faculty member in the Department of Biological Science and Technology, Yonsei University, Wonju, Republic of Korea.

IFRB Seminar Series, Spring 2013

The IFRB Seminar Series, Reproductive Biology Forum, has been held weekly during the Fall and Spring Semesters since 1990. The 2013 IFRB Seminar Series, coordinated by **Dr. Marcel Amstalden**, continues to provide an excellent combination of seminars from internationally recognized reproductive biologists from outside and inside the university along with advanced IFRB trainees. Each of the seminars is followed directly by a luncheon involving graduate student and postdoctoral trainee discussions with the seminar speaker:

January 25, **Dr. Marcel Amstalden**, Chair IFRB, IFRB General Business Meeting

February1, **Dr. Milo Wiltbank**, Department of Dairy Science, University of Wisconsin-Madison, "Regression of the corpus luteum: molecular mechanisms and practical implications." Hosted by **Dr. Fuller Bazer**

February 8, **Dr. Ashlee Watts**, Department of Large Animal Clinical Sciences, TAMU College Station, "Stem Cells in Veterinary Medicine - Where We Are and Where We Are Going." Hosted by **Dr. Katrin Hinrichs**

February 15, **Dr. Jules Puschett**, Texas A&M University College Vet Med Pathobiology, "Involvement of the cardiotonic steroids in hypertensive and neurologic disorders: a tale of two syndromes." Hosted by **Dr. Nancy Ing**

February 22, **Ted Wing**, DVM, Department of Veterinary Integrative Biosciences, TAMU, "Osteopontin binds to the αv integrin promoting porcine endothelial progenitor cell (EPC) incorporation into developing vasculature." Hosted by **Dr. Greg Johnson**

March 1, **Dr. John McCracken**, Department of Animal Science, University of Connecticut, "Milestones in reproductive endocrinology including the birth control pill." Hosted by **Dr. Joe Arosh**

March 8, **Dr. Carole Mendelson**, Department of Biochemistry and Obstetrics & Gynecology, University of Texas Southwestern Medical Center, Dallas, "miRiad roles of microRNAs in pregnancy and labor." Hosted by **Dr. Qinglei Li**

March 15, Spring break – No Forum

March 22, **Dr. Joan Jorgensen**, Department of Comparative Biosciences, University of Wisconsin, "Reproductive development pathways are linked to adult diseases." Hosted by **Dr. Sakhila Banu**

March 29, Reading day – No Forum

April 5, **Dr. Jan-Dirk Haeger**, Department of Anatomy, University of Veterinary Medicine Hannover, Hannover, Germany, "Complex spheroid-based culture models for bovine placental cells - implications for the study of reproduction and beyond." Hosted by **Dr. Greg Johnson**

April 12, **Dr. Shaye K. Lewis**, Scott Department of Urology, Center for Reproductive Medicine, Baylor College of Medicine, "Genomic disorders associated with genital anomalies and midline Fusion Defects." Hosted by **Dr. Robert Burghardt**

April 18-19, **Texas Forum of Reproductive Sciences,** 19th Annual Meeting, Texas Children's Pavilion for Women, Houston, TX

April 26, **Dr. Caurnel Morgan**, Department of Nutrition and Food Sciences, TAMU
Title: "Photoperiodic control of fertility markers in Syrian hamster trials." Hosted by **Dr. Marcel Amstalden**

Dr. Tom Spencer Gives Local Lecture

Dr. Tom Spencer, Professor Department of Animal Sciences, Washington State University, presented a keynote address at the Texas A&M Health Science Center-College of Medicine - Graduate Student Organization 2013 for the Graduate Student Research Symposium on April 26th, 2013. The title of his talk was "Pregnancy success and failure: Tails from different species."

Dr. Spencer his doctoral degree in Physiology of Reproduction at Texas A&M University with **Dr. Fuller W. Bazer** in 1995 and carried out postdoctoral studies with Dr. Bert O'Malley in the Department of Cell Biology, Baylor College of Medicine. He joined the Department of Animal Science at Texas A&M University in 2001 as an Assistant Professor of Reproductive Biology and Physiological Genomics and rose to the rank of Professor and AgriLIFE Research Faculty Fellow. He was recruited to Washington State University in 2011 as the new Baxter Professor of Beef Research. He is continuing to pursue studies to reach his long-term goal to discover and understand key hormonal, cellular and molecular mechanisms regulating development and function of the uterus and placenta using a variety of animal models including sheep, cattle and mice to advance understanding of basic uterine biology and pregnancy, as well as provide the basis for rational design of therapies aimed at the prevention and treatment of infertility and pregnancy loss in domestic animals and humans.

Dr. Spencer has continued to earn recognition for his work. He will receive the Research Award from the Society for the Study of Reproduction at the society's 46th annual meeting to be held in Montreal, in July. The SSR Research Award is an award of the highest honor that recognizes an active, regular member of SSR for outstanding research published during the previous six years. He was recently selected to become one of the next Editors-in-Chief of *Biology of Reproduction* with Dr. Franco DeMayo from the Baylor College of Medicine beginning July, 2013. He also recently awarded a $3 million grant from the U.S. Department of Agriculture, National Institute for Food and Agriculture to lead a team to help address cattle infertility, which is one of the biggest barriers to global competitiveness for American dairy farmers. The five-year grant, includes scientists from Washington State, University of Idaho and University of Florida and components in research, outreach and teaching.

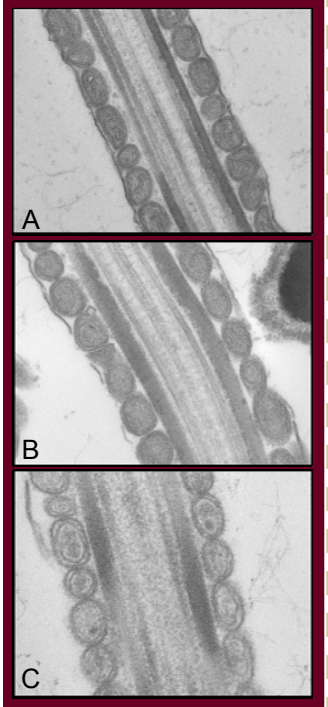
Trainee Spotlight

Shavahn Loux, is a doctoral candidate in the Department of Veterinary Physiology and Pharmacology, under the mentorship of **Dr. Katrin Hinrichs.** Her research has primarily been focused on the regulation of hyperactivated motility in stallion spermatozoa.

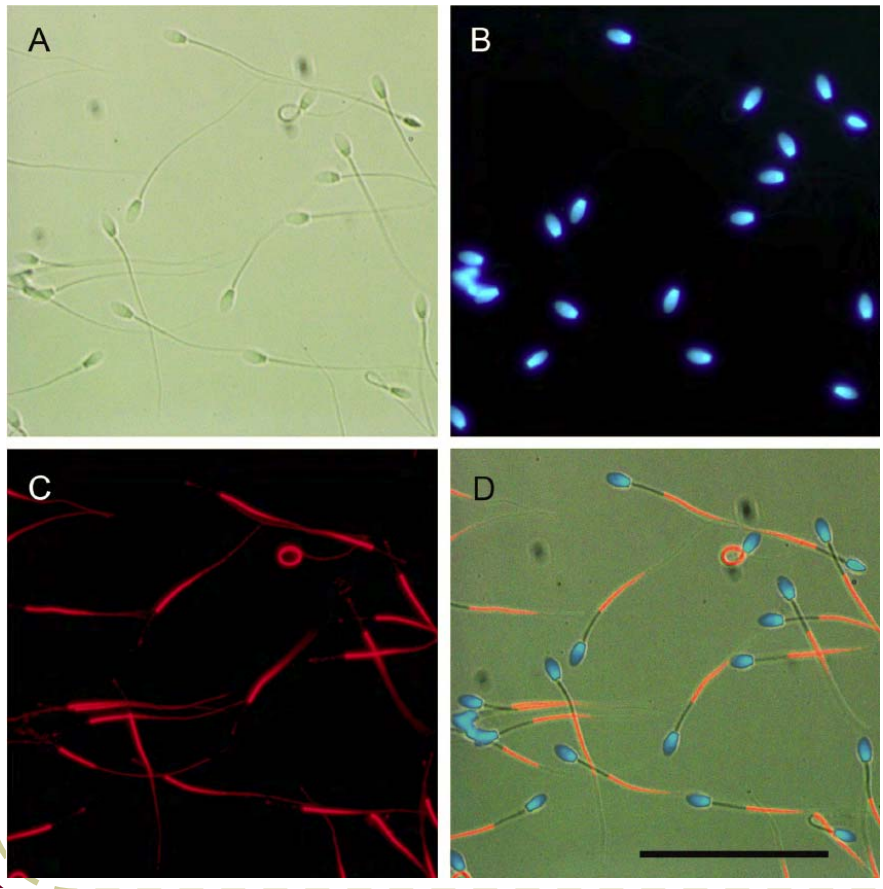
Shavahn's studies began by confirming the presence of the CatSper channel, a pH-gated calcium channel localized to the principal piece (***see Figure below***), in equine sperm. From there, she evaluated the effects of pharmacological and physiological effectors of hyperactivated motility, including high pH medium, procaine, ammonium chloride and 4–aminopyridine, on the kinetics of calcium, intracellular pH and motility. The CatSper channel inhibitors, mibefradil and NNC 55-0396 were used in combination with the above physiological and pharmacologic agents of hyperactivation. The results are complex, but indicate that equine sperm are particularly sensitive to intracellular calcium, and that motility declines rapidly as intracellular calcium increases. Further experiments revealed that the equine CatSper channel is not responsive to progesterone or prostaglandin E1, both effectors in human sperm.

Recently, Shavahn developed a technique for functional demembranization of equine sperm which maximizes membrane permeability and motility while minimizing damage to the intracellular structures of the sperm (***see Figure at left***). This model is ideal for studying motility as the sperm remain motile for significantly longer than traditional demembranization models. Shavahn is currently using this model to study the direct effect of calcium and pH on the equine axoneme, and the data model suggests that equine sperm require only picomolar amounts of calcium to maintain motility. Shavahn has found species-specific differences in the response of sperm motility to pH, with equine sperm preferring a much more acidic axonemal environment. She is currently working to determine if nickel and cadmium are toxic to demembranated sperm, as they are in sperm of other species, as they competitively bind with calcium.

While at Texas A&M, Shavahn has co-authored four publications, with another currently under review. She has two additional first-author manuscripts in progress as well. Before attending Texas A&M University, Shavahn received her Bachelor's degree at Cal Poly, San Luis Obispo in Animal Science. She is scheduled to graduate in December, 2013. She is thankful to have had the support of her family through her graduate education, including her husband Dean, seven-year-old daughter, Tyler and one-year-old son, Daxton.



Transmission electron microscopy of stallion sperm exposed to different concentrations of Triton-X for 30 sec to induce demembranization: A) 0%, showing intact plasma membrane; B) 0.02%, illustrating disruption of the plasma membrane with maintenance of mitochondrial integrity; C) 0.2% Triton X, showing extensive disruption of the plasma membrane and loss of mitochondrial integrity.



Photomicrographs of equine sperm after labeling. A) Brightfield; B) DAPI; C) Rabbit anti-human CATSPER1 immunostaining D) Overlay of all three images. Bar = 50 µm.

A Snapshot of IFRB Research

The IFRB is recognized as one of the most productive interdisciplinary research and education programs in reproductive biology in the U.S. The following "snapshot" of research productivity illustrates the multiple investigator research activities of the IFRB involving extensive participation of trainees during the 4 month period January 1 through May, 2013:

Albarzanchi AM, Sayes CM, Ridha Albarzanchi MT, Fajt VR, Dees WL, Kraemer DC (2013) Cilostazol blocks pregnancy in naturally cycling mice. Contraception 87(4):443-448

Antoniazzi AQ, Webb BT, Romero JJ, Ashley RL, Smirnova NP, Henkes LE, Bott RC, Oliveira JF, Niswender GD, Bazer FW, Hansen TR (2013) Endocrine delivery of interferon tau protects the corpus luteum from prostaglandin F2-alpha-induced luteolysis in ewes. Biol Reprod 2013 Apr 24. [Epub ahead of print]

Arnold CE, Love CC (2013) Laparoscopic evaluation of oviductal patency in the standing mare. Theriogenology 79(6):905-910.

Bazer FW (2013) Contributions of an animal scientist to understanding the biology of the uterus and pregnancy. Reprod Fertil Devel 25:1–19.

Bousquet M, Zhuang G, Meng C, Ying W, Cheruku PS, Shie AT, Wang S, Ge G, Wong P, Wang G, Safe S, Zhou B (2013) MiR-150 blocks MLL-AF9 associated leukemia by repressing multiple oncogenes. Mol. Cancer Res. Epub April 19.

Browning ZS, Wilkes AA, Mackenzie DS, Patterson RM, Lenox MW (2013) Using PET/CT imaging to characterize 18 F-fluorodeoxyglucose utilization in fish. J Fish Dis. 2013 Mar 14. doi: 10.1111/jfd.12081. [Epub ahead of print]

Bruno RG, Farias AM, Hernández-Rivera JA, Navarrette AE, Hawkins DE, Bilby TR (2013) Effect of gonadotropin-releasing hormone or prostaglandin F(2a)-based estrus synchronization programs for first or subsequent artificial insemination in lactating dairy cows. J Dairy Sci 96(3):1556-1567.

Carnahan MN, Veazey KJ, Muller D, Tingling JD, Miranda RC, Golding MC (2013) Identification of cell-specific patterns of reference gene stability in quantitative reverse-transcriptase polymerase chain reaction studies of embryonic, placental and neural stem models of prenatal ethanol exposure. Alcohol ;47(2):109-20. doi: 10.1016/j.alcohol.2012.12.003. Epub 2013 Jan 11.

Chadalapaka G, Jutooru I, Sreevalsan S, Pathi S, Kim K, Chen C, Crose L, Linardic C, Safe S (2013) Inhibition of rhabdomyosarcoma cell and tumor growth by targeting specificity protein (Sp) transcription factors. Int J Cancer 132 (4):795-806.

Choi YH, Norris JD, Velez IC, Jacobson CC, Hartman DL, Hinrichs K (2013) A viable foal obtained by equine somatic cell nuclear transfer using oocytes recovered from immature follicles of live mares. Theriogenology 79: 791-796.

Li C, McDonald TJ, Wu G, Nijland MJ, Nathanielsz PW. (2013) Intrauterine growth restriction alters term fetal baboon hypothalamic appetitive peptide balance. J Endocrinol 217:275-282.

Dai Z, Wu Z, Yang Y, Wang J, Satterfield MC, Meininger CJ, Bazer FW, Wu G (2013) Nitric oxide and energy metabolism in mammals. Biofactors. 2013 Mar 29. doi: 10.1002/biof.1099.

Das PJ, McCarthy F, Vishnoi M, Paria N, Gresham C, Li G, Kachroo P, Sudderth AK, Teague S, Love CC, Varner DD, Chowdhary BP, Raudsepp T (2013) Stallion sperm transcriptome comprises functionally coherent coding and regulatory RNAs as revealed by microarray analysis and RNA-seq. PLoS One. 2013;8(2):e56535.

Del Follo-Martinez A, Banerjee N, Li X, Safe S, Mertens-Talcott S (2013) Resveratrol and quercetin in combination have anticancer activity in colon cancer cells and repress oncogenic microRNA-27a. Nutr Cancer 65 (3):494-504.

Dorniak P, Welsh TH Jr, Bazer FW, Spencer TE (2013) Cortisol and interferon tau regulation of endometrial function and conceptus development in female sheep. Endocrinology 154(2):931-941.

Forde N, Mehta JP, McGettigan PA, Mamo S, Bazer FW, Spencer TE, Lonergan P (2013) Alterations in expression of endometrial genes coding for proteins secreted into the uterine lumen during conceptus elongation in cattle. BMC Genomics. 10;14(1):321. [González-Fernández L, Macías-García B, Loux SC, Varner DD, Hinrichs K (2013) Focal adhesion kinases and calcium/calmodulin-dependent protein kinases regulate protein tyrosine phosphorylation in stallion sperm. Biol. Reprod. doi: 10.1095/biolreprod.112.107078

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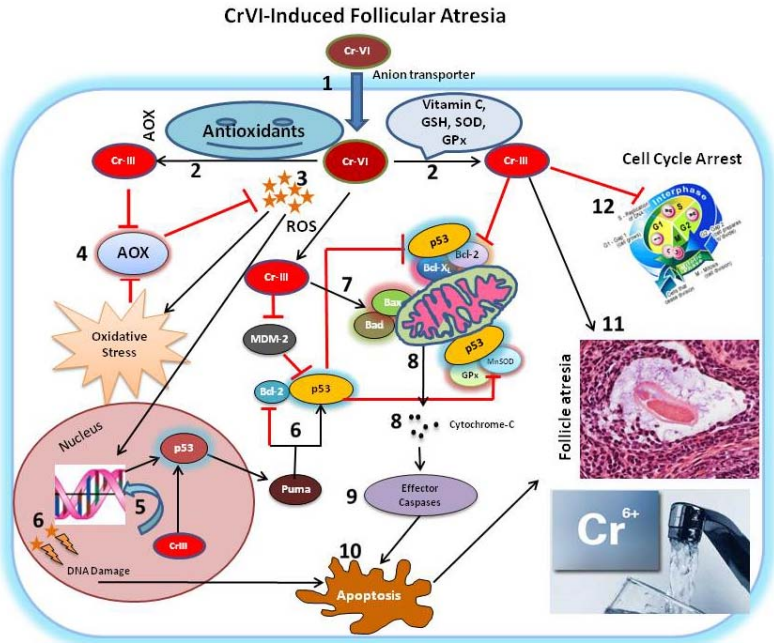
Stanley JA, Sivakumar KK, Nithy TK, Arosh JA, Hoyer PB, Burghardt RC, Banu SK (2013) Postnatal exposure to chromium through mother's milk accelerates follicular atresia in F1 offspring through increased oxidative stress and depletion of antioxidant enzymes. Free Radic Biol Med. Mar 4.[Epub ahead of print]

Global Issues (cont'd from page 1)

of contaminated water in Midland, Texas. "Significant amounts" of CrVI were found in the water of more than 40 homes in the area, some of which have now been fitted with state-monitored filters on their water supply. Brockovich said, "The only difference between here and Hinkley, is that I saw higher levels here than I saw in Hinkley" ("Brockovich: Midland, Texas Water Sullied." CBS News. June 10, 2009).

More than two million tons of CrVI processing wastes contaminate 150 sites in Northern New Jersey alone, as it is known as the chromate capital of the world. According to the National Tap Water quality data base and Environmental Working Group report (March, 2008), 1.7 million people from 42 communities living in the New Jersey area drink chromium contaminated water and are predisposed to various health problems including oral and lung cancers. Overall, in the U.S., at least 74 million people in nearly 7000 communities drink tap water polluted with chromium. Despite conservation and recycling efforts in U.S., over 20,000 metric tons of CrVI are released into the environment every year with over 5,000 metric tons released as atmospheric emissions. Non-occupational sources of CrVI include contaminated soil, air and water. The US Environmental Protection Agency (EPA) sets safe drinking water levels of Cr as 0.05 mg/L. Chromium contamination is a global issue affecting not only the U.S., but also India, Bangladesh, China and other countries. Reports from the Blacksmith Institute, and local news media indicated the "World's Top 10 worst polluted places" include the above nations. News articles about "India's threatened water supplies," and "drinking water pollution with CrVI in Tokyo, Japan" are few of the examples.

The extensive toxicokinetic and genotoxic literature associated with oral exposures to CrVI in animals and humans indicates that CrVI causes various health hazards including cancers, dermatitis, damage to the liver and kidneys, infertility in both males and females, defects in embryo/fetal development, and developmental problems in young children. A Cr supplement, Cr picolinate (a trivalent form of Cr) from health food stores has been proven to cause liver and kidney damage. Cr usage is increasing exponentially worldwide; and Cr pollution is a continuous, ongoing problem. According to The Blacksmith Institute, various drinking water sources such as lakes, rivers, and wells in developing countries are highly polluted with CrVI (31 mg/L water), where its concentration far exceeds the USEPA approved safer limits in drinking water of 0.05 mg/L for total Cr.



Schematic diagram of chromium-mediated apoptosis of granulosa cells, atresia of ovarian follicles and cell cycle arrest. *(1) CrVI is taken up by cells via the anionic transport system; (2) CrVI is reduced into CrIII by antioxidants, predominantly vitamin C and GSH; (3) Large amounts of free radicals (ROS) are generated during reduction of CrVI into CrIII; (4) AOX are consumed by cells to quench excess ROS that results in decreased AOX levels; (5) CrIII binds with DNA and forms DNA-adducts and induces mutation; (6) Free radicals can also directly induce DNA damage. DNA damage/mutation results in the up regulation of p53; (7) CrIII upregulates pro-apoptotic Bax and Bad proteins. All these changes culminate in change in mitochondrial membrane potential and release of cytochrome c (8) and (9) activation of effector caspases, that results in (10) apoptosis of granulosa cells and (11) atresia of ovarian follicles. (12) CrIII arrests cell cycle, and delays follicle development. Bars (red) indicate inhibition; Arrows (black) indicate stimulation.*

While the dangers of Cr in causing cancer are known, it's role in reproductive failures and infertility has not caught notice from the public, despite Cr being recognized as a teratogen. **Dr. Sakhila K. Banu** is an Assistant Professor in the Department of Veterinary Integrative Biosciences whose research is focused on identifying the molecular mechanism of CrVI-induced female reproductive toxicity and on developing molecular and nutritional preventive and therapeutic intervention strategies to mitigate or inhibit the adverse effects of Cr on female reproduction and embryo/fetal development. This research has been funded by National Institute of Environmental Health Sciences (NIEHS), NIH, and an Indo-US collaborative proposal award by the Indian Council of Medical Research, Federal Government of India.

Work in Dr. Banu's lab has shown that Cr passes through milk and the placenta and can gain access to the developing embryo/fetus through placenta; and developing offspring through mother's milk. Thus Cr has the potential to adversely affect early in utero and postnatal development of female reproductive organs, mainly the ovary and the uterus. Dr. Banu and lab members have discovered that postnatal exposure to Cr through mother's milk delays spuberty, decreases steroid hormone synthesis by the ovary, increases death of follicles in the ovary leading to premature ovarian failure and early reproductive senescence in laboratory rodents. Cr also reduces the number of pregnancies and number of pups born to the exposed females. Because Cr leaches up to 160 ft deep into the ground through contamination of soil and ground water, bioremediation of Cr from the soil or preventing the global production or use of Cr may not be very practical or successful.

Dr. Banu believes that the most important and feasible way to solve Cr problems on human health is to develop organ-based intervention strategies to prevent Cr-induced health problems. Interestingly, her research has identified and validated the efficacy of vitamin C against Cr-induced reproductive toxicity. Vitamin C is water soluble, easily available and affordable making transnationally relevant to protect human health against the adverse effects of CrVI. In addition, her research also identified the potential mechanism of CrVI-induced ovarian toxicity through increase in free radicals generation in blood, ovary and breast milk, depletion of various intracellular antioxidants, and p53 (a tumor-suppressor gene)-mediated mechanisms (**see Figure**). Her recent findings are published in several peer-reviewed journals such as *Toxicology and Applied*

(continued on page 12)

IFRB Trainee News

*** James (Will) Frank**, graduate student in the laboratory of **Dr. Greg Johnson**, earned second place honors in the platform competition at the 19th Annual Texas Forum for Reproductive Sciences held on April 18-19, 2013 at The Texas Children's Hospital Pavilion for Women, Houston, TX.

Will also received a TAMU College of Veterinary Medicine High Impact Research Achievement Award for Large Grant (a USDA National Institute of Food and Agriculture (NIFA) Graduate Fellowship).

He was also selected to present a plenary talk at the 9th International Conference on Pig Reproduction ICPR 2013) 9 - 12 June 2013, Olsztyn, Poland. The title of his talk is "Integrin αv is necessary for the attachment of trophectoderm cells to osteopontin to mediate adhesion of the conceptus to the uterus during implantation in pigs," Co-authors are Fuller W. Bazer, Robert C. Burghardt, Kayla J, Bayless and Greg A. Johnson.

*** Yang Gao**, a graduate student in the laboratory of **Dr. Qinglei Li**, was awarded a CVM Graduate Trainee Research Grant, during the Spring semester, 2013. His research focuses on the Smad signaling pathway in female reproductive system. He is currently investigating how transforming growth factor β signaling regulates extracellular matrix protein expression during myometrial development

*** Wei (Eddie) Ying**, graduate student working with **Drs. Beiyan Zhou and Fuller Bazer**, earned second place honors at the Texas A&M Student Research Week for his oral presentation entitled "A novel paradigm of microRNA regulation of insulin resistance: miR-150 regulates insulin sensitivity through controlling B cell functions." Eddie will also present platform and poster session presentations at the 5th Chinese American Diabetes Association in Chicago on June 20 in Chicago and the America Diabetes Association 73rd Scientific Sessions, being held June 21-25 at McCormick Place, Chicago, IL. Presentations include:

Ying W, Cheruku PS, Meng C, Wu G, Safe S, Bazer FW, Zhou B. Interferon tau alleviates adipose tissue inflammation and insulin resistance by regulating macrophage polarization.

Ying W, Mashoof S, Cheruku PS, Zhuang G, Bazer FW, Wu G, Safe S, Cristiello M, Zhou B. A novel paradigm of microRNA regulation of insulin resistance: miR-150 regulates insulin sensitivity through controlling B cell functions.

***Ted Wing,** graduate student working in the labs of **Drs. Greg Johnson, Kayla J. Bayless** and **Robert Burghardt** has been selected as a Finalist in competition for the Society for the Study of Reproduction (SSR) Trainee Research Award - Platform Presentation. The Finalists for this will compete together in a special platform session at the 46th Annual SSR Meeting , July 22-26, in Montreal Quebec.

Ted was also selected to receive a 2013 Animal Reproduction in Agriculture Research Fellowship funded by the United States Department of Agriculture, National Institute of Food and Agriculture, Agriculture and Food Research Initiative.

* **Michelle Bedenbaugh** has been elected by IFRB trainees to serve as the Trainee Representative on the IFRB Executive Committee and will replace Rodolfo Cardoso beginning in the Fall Semester.

Congratulations to IFRB Spring Semester Graduates:

*** Jennifer Thorson** completed her doctoral program in Physiology of Reproduction in December 2012, when she defended her dissertation entitled "Interactive roles of gonadotropin-releasing hormone and RF-amide related peptide 3 in adenohypophyseal physiology and reproduction in the mare." She was awarded her degree at the May 11, College of Agriculture and Life Sciences graduation ceremony. During her program, Jennifer authored or co-authored 8 abstracts presented at national and international meetings, co-authored a published review article, and has 3 manuscripts currently submitted or in revision for peer-reviewed publication. In 2012, Jenny was awarded a Tom Slick Graduate Research Fellowship from the College of Agriculture and Life Sciences. She is currently a postdoctoral research associate at USDA-ARS in Clay Center, Nebraska working in the laboratory of Dr. Clay Lents. **Drs. Gary Williams and Marcel Amstalden** served as co-chairs of her graduate advisory committee.

*** JeHoon Lee** completed his doctoral program in Biomedical Sciences in February, 2013 when he defended his dissertation entitled "Cellular transport of prostaglandins in the ovine uterus." During his graduate program, he authored or co-authored 13 peer-reviewed publications and numerous abstracts. **Dr. Joe Arosh** served as chair of his graduate advisory committee.

*** Jacob Thorne** completed his M.S. program in the Physiology of Reproduction under the direction of **Dr. David Forrest** in May 2013. The title of his thesis was "Fertility of beef recipients following a fixed-time embryo transfer protocol that includes follicle stimulating hormone diluted in hyaluronan." Jacob is currently a Research Assistant with Dr. Dan Waldron in Animal Breeding & Genetics at the Texas A&M AgriLife Research and Extension Center in San Angelo, TX.

IFRB Faculty Spotlight (cont'd from page 3)

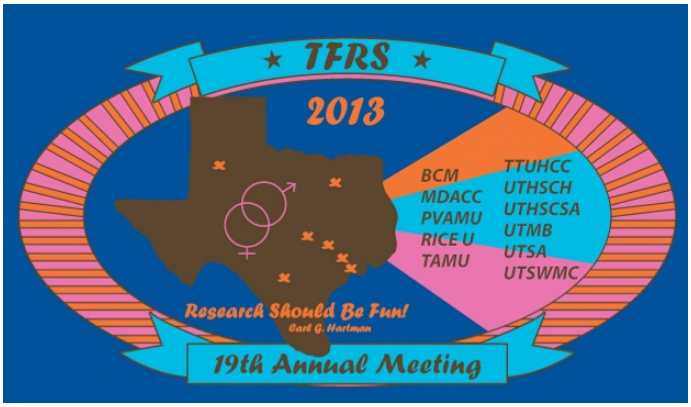
the frequency of episodic release of luteinizing hormone (LH) that precedes first ovulation. Amstalden's group has demonstrated that intermittent injections of kisspeptin stimulated episodic release of LH, increased ovarian steroidogenesis, and caused ovulation/follicle luteinization in peripubertal ewe lambs. In addition, the increase in frequency of LH release was associated with an increased expression of Kiss1, the gene encoding kisspeptin, in arcuate nucleus neurons. This observation indicates that the arcuate population of kisspeptin neurons serves as a primary excitatory pathway for the activation of GnRH neurons during pubertal transition. Using immunocytochemistry and image analysis approaches, it was observed that NPY neuronal fibers are in close proximity to kisspeptin neurons, and that this structural association appears to form functional synaptic inputs. Therefore, the NPY system may represent a potential candidate for mediating the nutritional regulation of kisspeptin neurons in sheep. More recently, Amstalden's group began investigations to determine whether the decreased sensitivity to estradiol negative feedback that is characteristic of peripubertal transition in the female involves regulation of kisspeptin neuron function.

Dr. Amstalden has been invited to present his research during the 2013 Annual Meeting of the Society for the Study of Reproduction in a talk entitled "Emerging Neuroendocrine Pathways Controlling the Onset of Puberty in Ruminants." and during the American Dairy Science Association-American Society for Animal Science Joint Annual Meeting this summer in a talk entitled "Neuroendocrine Programing of Accelerated Puberty in Heifers."

Funding for Amstalden's research has been provided by the Texas Beef Enhancement Program through Texas A&M AgriLife Research, by the National Institute of Food and Agriculture, and by the National Institute of Child Health and Human Development. Since joining the faculty at Texas A&M University, Dr. Amstalden has contributed to various IFRB programs. He coordinated the Annual Reproductive Biology Retreat from 2007 to 2010, and the Reproductive Biology Forum from 2010 to 2013. Dr. Amstalden has served in the Executive Committee of IFRB since 2007 and now serves as Chair of IFRB.

19th Annual Texas Forum for Reproductive Sciences

The 19th Annual Texas Forum for Reproductive Sciences (TFRS) was held on April 18-19, 2013 at The Texas Children's Hospital Pavilion for Women, Houston, TX. Over 110 participants from all major Texas universities registered for the meeting. Keynote lectures were presented by **Drs. Douglas Stocco**, Texas Tech University Health Science Center, Lubbock, TX, "The Role of Hormone Sensitive Lipase in the Regulation of StAR and Steroidogenesis" and **Cheryl Walker**, Institute for Biostatistics and Technology TAMU Health Science Center and College of Veterinary Medicine & Biomedical Sciences, "Reprogramming the Developing Reproductive Tract: Crosstalk between the Environment and the Epigenome." The annual meeting serves as a venue for the exchange or research ideas and discoveries in the spectrum of reproductive sciences. Platform and poster sessions focus almost exclusively on trainee presentations.



*-The logo for the 19th Annual TFRS Meeting was designed by **Dr. Greg A. Johnson**.*

Texas A&M University was well represented at the meeting. Four trainees presented platform session talks. **Xiaoqui Wang**, "Arginine and secreted phosphoprotein 1 act synergistically to stimulate MTORC1/MTORC2 cell signaling and cytoskeletal organization for proliferation, migration and adhesion of ovine trophectoderm cells." **Theodore Wing**, "Osteopontin binds to the αv integrin promoting porcine endothelial progenitor cell (EPC) incorporation into developing vasculature," **James W Frank,** "Loss of integrin subunit β3 during the peri-Implantation period leads to fetal growth retardation in sheep," **Ashley Keith,** "Microarray analysis portrays an adaptive placental response to nutrient restriction in ewes."

Trainees presenting posters included **JeHoon Lee,** "Selective inhibition of prostaglandin E2 receptors EP2 and EP4 inhibits growth of experimental endometriosis in nude mice evaluated by real-time noninvasive imaging and fluorescence microscopy," **Shavahn C. Loux**, "The effect of calcium and pH on the axoneme of demembranated stallion spermatozoa," **Jing Xu,** "Effects of interferon tau on ovine trophectoderm cell proliferation and gene expression," **Irene Ruiz-Gonzalez, "**Potential roles of exosomes, TLR7 and TLR8 in the regulation of enJSRV expression in the ovine conceptus during early pregnancy," **Grace Chang, "**Expression of α2β1 and Claudin-4 protein at the uterine-placental interface suggests roles in transport of proteins and nutrients from mother to fetus in pigs," **Xiaoling Zhu,** "Comparison of phenotypic markers of porcine conceptus trophectoderm, trophectoderm cell line and macrophages," **Chelsie A Burroughs,** "Possible roles of transporters SLC2A5 and SLC2A8 in fructose movement between maternal and fetal tissues during pig pregnancy," **Hannah M. DelCurto**, **"**Anterior gradient Homolog 2 in ovine placentome and its potential roles in pregnancy," **Wei Ying,** "Arginine decarboxylase/agmatinase: An alternative pathway for synthesis of polyamines in ovine conceptuses and uterus," **Yang Gao**, "Transforming growth factor β signaling regulates extracellular matrix protein expression during myometrial development."

Several current and former trainees currently in Research Scientist or Postdoctoral positions also presented their work at the meeting including Drs. **Young-Ho Choi, Shaye K. Lewis, Xilong Li,** and **Haijun Gao.**

Save the date for the 20th Annual Texas Forum for Reproductive Sciences at the University of Texas at San Antonio Downtown Campus, April 10-11, 2014.

Global Issues (cont'd from page 9)

Pharmacology, Free Radical Biology and Medicine, and *Reproductive Toxicology*. Ongoing research in her lab is following these studies with the use of nutrioxidants such as resveratrol, curcumin, and antioxidants such as glutathione and N-acetyl cysteine to inhibit the toxic effects of CrVI. Her long-term research goal is to determine molecular and cellular mechanisms of heavy metal endocrine disruptors on female reproduction and fetal development, discover trans-generational effects of metal toxicity on female reproduction and reproductive cancers through epigenetic pathways.

- IFRB thanks Dr. Sakhila Banu for her help with the preparation of this Global Issues report.

Faculty Awards, etc. (cont'd from page 4)

INVITED LECTURES continued

*** Qinglei Li**, "TGFβ signaling in female reproduction," Department of Molecular & Cellular Medicine, Texas A&M Health Science Center, May 14, 2013.

*** Guoyao Wu**, plenary talk "Maternal and fetal amino acid metabolism in gestating sows." 9th International Conference on Pig Reproduction, Olsztyn, Poland., June 9-12, 2013.

HONORS & AWARDS:

*** Dr. Greg A. Johnson,** Associate Professor of Veterinary Integrative Biosciences was appointed to the Editorial Board of *Reproduction*.

Qinglei Li, Assistant Professor in the Department of Veterinary Integrative Biosciences was one o 30 young investigators selected nationwide for for the 2013 Ralph E. Powe Junior Faculty Enhancement Award. These awards are intended to enrich the research and professional growth of outstanding junior faculty and result in new funding opportunities.

*** Dr. Carey Satterfield,** Assistant Professor in the Department of Animal Science, received the 2013 Outstanding Young Animal Scientist in Research Award from the Southern Section of the American Society of Animal Science.

*** Dr. Guoyao Wu,** Distinguished Professor, University Faculty Fellow, and AgriLife Research Senior Faculty Fellow in the Department of Animal Science was recently named the 2013 Sigma Xi - Texas A&M Chapter Distinguished Scientist.

46th SSR Annual

SSR

Society for the Study of Reproduction

The 46th Annual Meeting of the Society for the Study of Reproduction will be held in Montréal, Québec, Canada July 22-26, 2014. The theme of the Meeting is "Reproductive Health: Nano to Global." Speakers include SSR's own Martin Matzuk, M.D., Ph.D., delivering the Keynote Address; Lonnie D. Shea, Ph.D., Jerry Taylor, Ph.D., and Jackson Kirkman-Brown, Ph.D., in the President's Symposium; Sabra L. Klein, Ph.D., in a special Plenary Session; Mitinori Saitou, M.D., Ph.D., will present the State-of-the-Art Lecture; JoAnne S. Richards, Ph.D., will deliver the Historical Perspectives Lecture; and Serdar E. Bulun, M.D., will present the ASRM Exchange Lecture.

To date, eleven trainees who have had first authored abstracts accepted for presentation at the meeting will receive travel support to attend the meeting. Trainees include: Michelle Benenbaugh, Chelsie Burroughs, Rodolfo Cardoso, Hanna De.Curto, Will Frank, Yang Gao, Ashley Keith, Xiaoqui Wang, Ted Wing, Jean Xu, and Eddie Ying. Three other trainees including Brittni Littlejohn, Meghan Roberts, and Sarah Schmidt will receive support to attend the joint American Dairy Science Association/American Society of Animal Science Annual Meeting. The IFRB gratefully acknowledges funding provided by the Texas A&M University Division of Research and Deans of the Colleges of Agriculture and Life Sciences and Veterinary Medicine & Biomedical Sciences.

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IFRB RESEARCH AND TRAINING MISSION:

Reproductive Biology is at the epicenter of the life sciences. Focal areas of research and graduate/postdoctoral training in the IFRB are interdisciplinary and cover both genders, encompass humans, domestic animals, laboratory animals and wildlife, and include: assisted reproductive techniques, biological clocks, cloning, conservation of endangered species, contraception, developmental biology, diseases of the reproductive tract, endocrinology, fertilization, fetal growth retardation, gametogenesis, gender-biased diseases and health issues, immunology, infertility, lactation, pregnancy and pregnancy-related disorders, premature labor, recovery of function, science and health policy, stem cell biology, systems biology and functional genomics, reproductive toxicology, and uterine biology. The outcomes of this research are impacting Texas, our nation and the world.