Veterinária e Zootecnia

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FMVZ
2016

E X P E D I E N T E

A Revista **Veterinária e Zootecnia,** da Faculdade de Medicina Veterinária e Zootecnia-UNESP, Campus de Botucatu, publica artigos científicos originais, artigos de revisão bibliográfica, relatos de casos e comunicações curtas, referentes às áreas de Medicina Veterinária e de Zootecnia, com periodicidade trimestral, em português, espanhol, ou inglês, sendo os conceitos e opiniões emitidas, de responsabilidade exclusiva dos autores. Poderá editar e disponibilizar em sua página na internet, suplementos de eventos científicos.

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PROCEEDINGS WORKSHOP IN INTERNATIONALISATION IN VETERINARY SCIENCES: PERSPECTIVES FOR RESEARCH BETWEEN UNESP AND THE UNIVERSITY OF GLASGOW, SCOTLAND, UK

October 10th to 12th 2016

Fazenda do Lageado, FMVZ – UNESP, Botucatu, SP, Brazil

Dear colleagues,

Should research serve present-day life and represent new challenges?

It is a great pleasure for us to welcome you to the Workshop in Internationalisation in Veterinary Sciences: perspectives for research between UNESP and the University of Glasgow, Scotland, UK here in Botucatu, Brazil.

This is the first workshop between UNESP and the University of Glasgow to identify common areas of research in animal health, to establish and build links for future collaboration between Brazil and the United Kingdom, and to strengthen the internationalization programmes of both UNESP and the University of Glasgow.

The joint organizing committee is thankful for the support of UNESP - AREX (International Relations Office), PROPe, PROPG, Plano de Desenvolvimento Institucional (PDI), School of Veterinary Medicine and Animal Science (FMVZ – campus of Botucatu), and the University of Glasgow.

Finally, we believe that the workshop will be a great opportunity for all the participants to discuss perspectives for research between UNESP and the University of Glasgow and to facilitate partnerships between research groups and professional networks in Veterinary Sciences.

Yours sincerely,
The Organizing Committee

Prof. Ass. Dra. Elizabeth Moreira dos Santos Schmidt

ORGANIZING COMMITTEE

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Prof Elizabeth Moreira dos Santos Schmidt – Department of Veterinary Clinical Sciences – School of Veterinary Medicine and Animal Science, UNESP, campus of Botucatu, Brazil
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University of Glasgow, Scotland, UK

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Prof. Dra. Elizabeth Moreira dos Santos Schmidt
Prof. Dr. João Carlos Pinheiro Ferreira
Prof. Dr. José Paes de Almeida Nogueira Pinto
Prof. Dr. Peter David Eckersall

PROGRAME

Monday, Oct 10th

8:30 Opening Session

Session 1
9:15 Proteomics and acute phase proteins in infectious diseases of animals – Prof. Peter David Eckersall – University of Glasgow
10:15 Clinical pathology and acute phase proteins in nematode parasitic infections – Prof Elizabeth Moreira dos Santos Schmidt – FMVZ, UNESP, campus of Botucatu
11:00 Nutritional and hormonal strategies to improve reproductive efficiency in beef and dairy cattle – Prof José Luiz Moraes Vasconcelos - FMVZ, UNESP, campus of Botucatu

Session 2
14:00 Oral diseases of animals: peering into the unknown – Prof Marcello Riggio – University of Glasgow
15:00 Epidemiology and pathogenesis of periodontitis in ruminants – Prof. Iveraldo dos Santos Dutra – FMVA, UNESP, campus of Araçatuba

Session 3
16:10 Arbovirus-host interactions studies and the Centre for Virus research activities – Dr Alain Kohl – University of Glasgow
17:10 Epidemiological aspects of Zika virus in Brazil: challenges posted by this new public health threat – Prof Adriano Mondini – FCFAR, UNESP, campus of Araraquara

Tuesday, Oct 11th

Session 4
9:00 Improving the quality and safety of eggs – Prof Maureen Bain – University of Glasgow
10:00 Optimization modeling with spreadsheets: finding simplicity in complexity – Prof Manoel Garcia Neto – FMVA, UNESP, campus of Araçatuba

Session 5
11:10 Ovarian antral follicule differentiation – insights from the ruminant model – Prof Monika Mihm-Carmichael – University of Glasgow
12:10 Livestock genomics: the perspectives of Zebu cattle – Prof José Fernando Garcia – FMVA, UNESP, campus of Araçatuba

Session 6
14:45 The genetics and genomics of protozoan parasites – Prof William Weir – University of Glasgow
15:45 Sustainable control of parasitic gastroenteritis in ruminants – Prof Alessandro Francisco Talamini do Amarante – IBB, UNESP, campus of Botucatu

Session 7
Research experiences at the University of Glasgow
17:00 Follicular fluid proteome profile of dairy cows – MSc Rodrigo Ferrazza, FMVZ, UNESP, campus of Botucatu
17:25 Identification of the bacteria and the evaluation of tissue levels of Toll-like receptor and cytokine mRNAs associated with bovine periodontitis and oral health – MSc Ana Carolina Borsanelli, FMVA, UNESP, campus of Araçatuba
17:50 Proteomic investigation of differentially expressed proteins in buffalo (*Bubalus bubalis*) milk during mastitis – Dr André Marcos Santana, FCAV, UNESP, campus of Jaboticabal

Wednesday, Oct 12th

9:00 Posters setup, 9:45 Posters presentations
11:00 Networking Research-Coffee
13:00 Closing

CONTENTS

ABSTRACTS OF POSTER PRESENTATIONS

PROTEOMICS AND ACUTE PHASE PROTEINS IN INFECTIOUS DISEASES OF ANIMALS

Professor P. David Eckersall
Institute of Biodiversity, Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences
University of Glasgow, UK

Acute phase proteins such as bovine haptoglobin (Hp) and serum amyloid A (SAA) have become established as biomarkers of infection and inflammation in veterinary medicine. In mastitis, caused by infection of the mammary gland, the most serious animal health issue for dairy farmers, these biomarkers have been found in milk as well as in serum. However there are many addition changes in the protein content of milk during mastitis and this has now been explored using the advanced technologies of proteomics. In an experimental model of *Streptococcus uberis* mastitis, milk from 0 to 312 h post challenge (pc) has been examined by quantitative proteomic approaches to provide a wide analysis of the changes to the milk proteome that occurs following bacterial challenge. Analysis of the milk proteins of <25 kDa using capillary electrophoresis and mass spectrometry identified 75 peptides reaching a peak intensity at 81 h pc which provided a biomarker profile for mastitis detection. These peptides included casein degradation products and endogenous antibacterial agents such as serum amyloid A. Quantitative proteomic analysis of the milk samples by liquid chromatography and mass spectrometry identified 570 proteins in the milk and demonstrated that the cathelicidins, immune cell derived antimicrobial peptides, had the one of the greatest relative increases in expression in initial stages of mastitis with haptoglobin and the associated acute phase proteins having overall the most enhanced pathway with the peak response at 57 and 81 h pc. While the role of Hp and SAA as biomarkers of mastitis have been confirmed by the investigation, novel biomarkers of this disease as well as the use of biomarker profiling have been identified. The investigation was enabled by close interaction with Glasgow Polyomics of Glasgow University, a centre for proteomic as well as genomic and metabolomics investigations, which is available for collaboration in future studies.

Biography

Professor David Eckersall graduated from the University of Liverpool with a BSc in Biochemistry (1973) and a PhD in Biochemistry from University of Edinburgh (1977). Prof Eckersall is the Professor of Veterinary Biochemistry at the School of Veterinary Medicine, University of Glasgow. His research has been focused on the diagnostic applications of protein analysis in veterinary medicine and has published over 200 peer reviewed papers, holds 4 patents and co-edited the first book on animal proteomics (Methods in Animal Proteomics, Wiley). He was the Chair of the COST Action for Farm Animal Proteomics (2011-14). He was awarded the Heiner Sommer Prize of the International Society for Animal Clinical Pathology for Lifetime Contribution to Animal Clinical Biochemistry in 2008, the Siemens Prize of the Division of Animal Clinical Chemistry of the American Association of Clinical Chemistry for Contributions to Animal Clinical Chemistry in 2010 and the Lifetime Achievement Award of the Comparative Clinical Pathology Association in May 2016. His university spinout

company ReactivLab Ltd was acquired by Avacta Group Ltd in 2010. He is a Fellow of the Royal College of Pathologists, a Fellow of the Royal Society of Biology and a Member of the Academia Europaea.

Publications

MANSOR R., MULLEN W., ALBALAT A., ZEREFOS P., MISCHAK H., BARRETT D.C., BIGGS A. & ECKERSALL P.D. (2013) A peptidomic approach to biomarker discovery for bovine mastitis. Journal of Proteomics 85, 89-98.

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THOMAS FC, MUDALIAR M, TASSI R, MCNEILLY TN, BURCHMORE R, BURGESS K, HERZYK P, ZADOKS RN, ECKERSALL PD. (2116) Mastitomics, the integrated omics of bovine milk in an experimental model of Streptococcus uberis mastitis: 3. Untargeted metabolomics. Molecular Biosystems 12:2762-9. doi: 10.1039/c6mb00289g.

ORAL DISEASES OF ANIMALS – PEERING INTO THE UNKNOWN

Dr Marcello Riggio
Oral Sciences Research Group
Dental School
College of Medical, Veterinary and Life Sciences
University of Glasgow
Scotland, UK

The causes of oral diseases of animals have received relatively little attention in recent decades compared to analogous diseases in humans, despite their frequent occurrence and the pain and discomfort caused. This presentation will summarise recent research conducted on the microbial and immune basis of oral diseases of small companion and large farm animals.

Feline chronic gingivostomatitis is a chronic oral inflammatory disease that causes severe pain and distress and is challenging to treat, often necessitating total dental extraction. Recent work on the identification of putative pathogens and their interaction with the innate immune system will be presented and discussed.[1,2]

Equine periodontitis is a common and painful condition, whose prevalence increases with age, which can lead to tooth loss. Its aetiopathogenesis remains poorly understood despite recent increased awareness of this disorder amongst the veterinary profession. Bacteria and their interaction with the innate immune system is known to play an important role in human periodontitis, but such a role in equine periodontitis has not yet been examined. Data on the microbiomes associated with equine periodontitis and oral health, highlighting key differences, will be presented.[3] Changes in Toll-like receptor and cytokine expression profiles between health and disease will also be discussed.[4]

There is little known about dental disease in cattle and no routine dental treatments currently exist. Although cattle are of worldwide economic importance in the dairy and beef industries, their dentition has not been investigated as thoroughly as that in other herbivores and small animals. We have recently shown that periodontitis is frequently found in slaughtered cattle.[5] Data will be presented on the microbiomes associated with bovine periodontitis and oral health.

Knowledge of pathogens involved in the aetiopathogenesis of animal oral diseases and their interaction with the host immune system will form the basis for future development of novel therapies for their prevention, treatment and management.

Biography

Dr Riggio graduated from the University of Leicester with a BSc (Hons) degree in Chemistry-with-Biochemistry in July 1983 and with a PhD from the University of Glasgow in 1991. He is currently a Senior Lecturer in Molecular Microbiology at the the Dental School, University of Glasgow. His main research theme is to identify the bacteria involved in inflammatory oral diseases in animals and humans. Following his previous role as a Councillor, he was elected as Honorary Secretary of the British Society for Oral & Dental Research (BSODR) in 2014 and has served as its Webmaster since 2012. He has been Secretary/Treasurer of the Oral Microbiology & Immunology Group (OMIG) of BSODR since 2008. In 2011 he became an Editor for the Journal of Medical Microbiology and was appointed as a Senior (Section) Editor in 2015. He has

served as a Councillor of the Association of Basic Science Teachers in Dentistry since 2012 and was appointed as its Membership Secretary in 2015.

Publications

Dolieslager SMJ, Riggio MP, Lennon A, Lappin DF, Johnston N, Taylor D, Bennett D. Identification of bacteria associated with feline chronic gingivostomatitis using culture-dependent and culture-independent methods. *Veterinary Microbiology* 2011; 148: 93-98.

Dolieslager SM, Lappin DF, Bennett D, Graham L, Johnston N, Riggio MP. The influence of oral bacteria on tissue levels of Toll-like receptor and cytokine mRNAs in feline chronic gingivostomatitis and oral health. *Veterinary Immunology and Immunopathology* 2013; 151: 263-274.

Kennedy R, Lappin DF, Dixon PM, Buijs MJ, Zaura E, Crielaard W, O'Donnell L, Bennett D, Brandt BW, Riggio MP. The microbiome associated with equine periodontitis and oral health. *Veterinary Research* 2016; 47: 49.

Kennedy R, Lappin DF, Dixon PM, Bennett D, Riggio MP. Gingival Toll-like receptor and cytokine mRNA levels in equine oral health and periodontitis. *Equine Veterinary Journal* 2016; doi: 10.1111/evj.12597

Borsanelli AC, Viora L, Lappin DF, Bennett D, King G, Dutra IS, Riggio MP. Periodontal lesions in slaughtered cattle in the West of Scotland. *Veterinary Record* 2016; in press.

ARBOVIRUS-HOST INTERACTIONS STUDIES AND THE CENTRE FOR VIRUS RESEARCH ACTIVITIES

Dr Alain Kohl

MRC Programme Leader
MRC-University of Glasgow Centre for Virus Research
University of Glasgow, UK

My research focuses on arboviruses, and more specifically on virus replication and interaction with host responses. On the vector side we try to understand how these responses modulate the relationship between the arthropod vector and the virus, specifically with regards to RNA interference pathways. We also have a programme on the genetic modification of mosquitoes. With regards to vertebrate cells, which is a minor topic in the group, we also study innate immune responses and virus-cell interactions. All major aborvirus families are currently being worked on: *Togaviridae* (chikungunya, Semliki Forest viruses), *Flaviviridae* (dengue, Zika, tick-borne encephalitis viruses) and *Bunyaviridae* (various orthobunyaviruses and phleboviruses including Rift Valley fever viruses). Many more viruses are available. I am generally interested in topics relating to arboviruses as well other emerging viruses. In recent years I have also become more involved in virus and mosquito ecology, which involves studies with partners overseas. I currently have collaborations with partners in Brazil on Zika virus (MRC Newton Fund, UK-Brazil Neglected Infectious Diseases Partnership with FIOCRUZ Recife), and Uganda.
We have numerous tools to study arboviruses (including reverse genetics) and cellular systems at our disposal, and continuously increase these. The CVR is very well equipped for modern virology, this includes high throughput facilities for sequencing and screening, as well containment level 2 and 3 laboratories that include animal house and insectary facilities.

Biography

Following a diploma in Biology as the University of Muenster (Germany), I obtained a PhD the University Paris 7/Institut Pasteur (France) in Microbiology at the end of 1999, working on Rift Valley fever virus. After that I joined Richard Elliott's group in Scotland to continue my work on bunyaviruses at the University of Glasgow and later on in St. Andrews. In 2006 I obtained a Wellcome Trust Research Career Development Fellowship to set up my group at the University at Edinburgh, where I also later joined the Roslin Institute as Career Track fellow. In 2011 I joined the newly formed MRC-University of Glasgow Centre for Virus Research to take up a position as MRC Programme Leader. My group currently consists of 5 post doctoral fellows (including one junior PI and one MSCA Horizon 2020 Fellow), 3 PhD students and 2 technicians.

Publications

Host Inflammatory Response to Mosquito Bites Enhances the Severity of Arbovirus Infection. Pingen M, Bryden SR, Pondeville E, Schnettler E, **Kohl A**, Merits A, Fazakerley JK, Graham GJ, McKimmie CS.Immunity. 2016 Jun 21;44(6):1455-69. doi: 10.1016/j.immuni.2016.06.002.

Wolbachia Blocks Viral Genome Replication Early in Infection without a Transcriptional Response by the Endosymbiont or Host Small RNA Pathways. Rainey SM, Martinez J, McFarlane M, Juneja P, Sarkies P, Lulla A, Schnettler E, Varjak M, Merits A, Miska EA, Jiggins FM, **Kohl A**. PLoS Pathog. 2016 Apr 18;12(4):e1005536. doi: 10.1371/journal.ppat.1005536.

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Zika **virus**: a previously slow pandemic spreads rapidly through the Americas.Gatherer D, **Kohl A**.J Gen Virol. 2016 Feb;97(2):269-73. doi: 10.1099/jgv.0.000381.

IMPROVING THE QUALITY AND SAFETY OF EGGS

Professor Maureen Bain
Institute of Biodiversity, Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences
University of Glasgow, UK

The poultry industry relies on artificial incubation of eggs to limit the transfer of microorganisms from one generation to the next. Despite this the vertical transmission from broiler and layer breeders to production flocks has been identified by EFSA as the most likely route of transfer of antibiotic resistant e-coli and salmonella. There is also the opportunity for horizontal transmission to occur during the collection and transport of eggs. Irrespective of the route or site of transfer, the entry of pathogenic or zoonotic organisms to the egg contents is undesirable for food safety.

A deterioration in egg numbers combined with a decline in shell quality are the main reasons for replacing a commercial laying flocks at or around 72 weeks. Poor shell quality at any time not only results in financial loss but also results in major contamination problems for the highly mechanised egg packing and handling equipment. The long-term maintenance of the tissues and organs involved in producing eggs is a therefore a prerequisite for extending the laying cycle of commercial flocks. Breeding companies are looking to develop the 'long life' laying hen, which is capable of producing some 500 eggs in a production cycle lasting 100 weeks. This goal can only be achieved using selection programs that focus on stabilising egg quality and safety. For decades Poultry Breeding companies have been using a range of laboratory based measurements in their selection programs to improve egg quality and safety. However, although these measurements have generally responded to selection, for those relating to physical shell quality it has been notoriously difficult to prove that the measures actually relate to the susceptibility of the egg to damage or bacterial penetration in the field. The dynamic stiffness and cuticle deposition measurements developed at the University of Glasgow offer breeding companies a new opportunity to improve egg quality and safety. The dynamic stiffness measurement has a moderate heritability and provides a rapid, non-destructive measurement of eggshell quality that accurately predicts an egg's susceptibility to cracking under field conditions. This measurement is already being incorporated into breeding programs with reported success both in terms of improved hatchability and number of saleable eggs. The cuticle is the first line of defence of the egg to the penetration of bacteria. As a direct result of our research we now have a much better idea of the physiochemical and functional properties of this proteinaceous layer in terms of its role in preventing bacterial ingress. A method to quantify cuticle deposition has also been established. This measurement has a moderate to high heritability in all breeding stock (i.e. both layers and broiler) so it should respond well to selection. Genetic selection to improve cuticle deposition in both meat type and egg laying flocks is on the horizon and will improve the safety of eggs by reducing the risk of pathogenic or zoonotic organisms entering the egg contents.

Biography

Professor Maureen Bain graduated from the University of Glasgow with a BSc (Hons) in Zoology (1986) and a PhD (1990) and became a full professor in Comparative Veterinary Anatomy and Histology at the University of Glasgow in 2013. Maureen's area of expertise is in Avian Reproduction and the development of novel methods of assessing egg quality. Maureen has collaborated and published widely in the field of Egg and Eggshell Quality and works closely with key stakeholders in the Poultry Industry (Eggs and Meat). In 2014 she received the Howie Sturgenor Cup presented to an individual who has provided outstanding contribution to the UK Poultry industry. Maureen has been an invited speaker at both national and international conferences and is the chairperson of the European Poultry Federation Working Group4 (Eggs and Egg products). She is also a serving council member of the UK branch of WPSA and a director of British Poultry Science Ltd. Within the college of MVLS Maureen she has two senior management roles: Vice Dean of the School of Veterinary medicine and the Deputy Director of the Institute of Biodiversity Animal Health and Comparative Medicine. She also played a leading role in the development of the new BVMS program which was launched in 2013 and currently leads the Foundation Phase (years 1-2). She teaches veterinary undergraduate students and supervises post graduate research students.

Publications

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OVARIAN ANTRAL FOLLICLE DIFFERENTIATION – INSIGHTS FROM THE RUMINANT MODEL

Dr Monika Mihm Carmichael
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Division of Farm Animal Clinical Sciences, School of Veterinary Medicine
University of Glasgow, UK

Compromised ovarian function limits female fertility, by producing ovulatory follicles unable to contribute healthy female gametes to fertilization, or providing inadequate hormonal support for early embryonic development. The cow is, similar to the woman, a monovulatory species with a single selected dominant follicle undergoing substantial growth and differentiation essential for timely oestrus and ovulation. We have used the bovine dominant follicle model and applied different transcriptomic techniques (serial analysis of gene expression and cDNA array) to identify known and new follicular (granulosa and theca) cell genes involved in steroidogenesis, cell differentiation, proliferation and apoptosis, signalling, cell metabolism, and RNA and protein synthesis, which are differentially regulated during development of the healthy dominant follicle related to high fertility. Subsequent realtime PCR analyses and in vitro studies utilising siRNA knock-down technology further validated important candidate genes for normal differentiation, which regulate granulosa and theca cell gonadotrophin receptor response, estradiol and progesterone synthesis, and the binding of members of the inhibin and FGF growth factor families. Similar approaches combining the dominant follicle model with transcriptomic (new generation sequencing), PCR and hormonal studies identified the cholesterol synthesis and transport pathways as being affected by a negative metabolic state in lactating dairy cows or nutritionally restricted heifers, with detrimental consequences for gonadotrophin response, IGF signaling and estradiol biosynthesis in differentiating dominant follicles. It has also clearly been shown in the ruminant that adult ovarian function can be abnormally programmed by intra-uterine life experiences such as restricted nutrition or exposure to excess androgen in the dams, leading to reduced reproductive potential or sub- and infertility in early adulthood in the female offspring. Using the prenatally androgenised ewe model developed by Dr Jane Robinson, we confirmed the abnormal growth seen in large antral follicles in postpubertal androgenised ewes, and identified candidate genes for abnormal intra-uterine programming which regulate granulosa cell proliferation, apoptosis, gonadotrophin response and steroid synthesis. Therefore, our detailed understanding of follicular differentiation not only provides an insight into the pathophysiology of abnormal follicle function in monovulatory species, which is of major economic importance in the high-yielding postpartum dairy cow, but will also identify cellular and circulating targets for the development of novel diagnostic and therapeutic approaches.

Biography

Monika graduated in veterinary medicine from Hannover, Germany, in December 1989, and after 2 years in mixed practice in Herefordshire, UK, joined the research group of

Professors Jim Roche, Maurice Boland (University College Dublin), and Jim Ireland (Michigan State University), where she pursued physiological studies into systemic and local regulators of ovarian antral follicle development in cattle, as well as the consequences of abnormal follicle function on the oocyte and fertility. She joined the Glasgow University Veterinary School in September 1997, and is now part of the new curriculum design team, clinical phase course leader, foundation and clinical phase module leader, and joint Head of the Farm Animal Division. She teaches veterinary undergraduate students in their foundation, clinical and professional phase, as well as postgraduate students and practitioners, with an emphasis on veterinary reproduction and clinical skills. Monika is keen for students, practitioners and researchers to understand the link between physiology and clinical practice, particularly in bovine reproduction, has presented plenary lectures at numerous international conferences (ESDAR, ICAR, ICFAE), and published over 20 review articles on aspects of bovine follicle development, postpartum resumption of ovulation, manipulation and fertility. She currently supervises postgraduate research projects determining the influence of mild or severe disease on antral follicle health and granulosa cell function. (You can contact Monika by emailing Monika.MihmCarmichael@glasgow.ac.uk).

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THE GENETICS AND GENOMICS OF PROTOZOAN PARASITES

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Over the last decade, the genomes of a large number of protozoan pathogens of veterinary and human importance have been sequenced. This includes tick-borne parasites, such as *Theileria* and *Babesia*, insect-borne parasites, such as *Trypanosoma* and non-vector borne pathogenic protozoa such as *Cryptosporidium*. The availability of complete genomes has facilitated a wealth of both basic and translational veterinary research. In terms of advancing animal health, this includes the identification of novel diagnostic targets for both molecular detection and serological assays. Genomic analysis has also been used to identify immunogenic proteins and to assess their suitability as subunit vaccine candidates. Arguably, one of the most important outcomes of possessing a pathogen genome sequence is the ability to develop genome-wide genetic markers for population-level analysis. Such markers have been used for investigating transmission pathways and determining sources of infection and this is increasingly being applied to viral and bacterial disease outbreaks. For protozoa, the focus of genetic studies has largely been investigating the basic diversity and population structures of parasitic species. A variety of population structures exist, from randomly mating organisms such as *Theileria* to predominantly asexual species such as *Toxoplasma*. The advent of next generation sequencing technology has facilitated a move from sparse marker-based population genetics to population genomics and this has allowed asexual organisms to be studied at a much higher level of resolution. Evolutionary theory predicts that the lack of recombination and chromosomal re-assortment in strictly asexual organisms results in homologous chromosomes irreversibly accumulating mutations and thus evolving independently of each other, a phenomenon termed the Meselson effect. We have applied a population genomics approach to examine this effect in a *Trypanosoma* species. We determined that this particular pathogen is evolving strictly asexually and is derived from a single progenitor, which emerged within the last ten millennia. We also demonstrated the Meselson effect for the first time at the genome-wide level in any organism. These findings shed new light on the genomic and evolutionary consequences of strict asexuality, which pathogens may use to exploit new biological niches.

Biography

Dr Weir graduated from the University of Glasgow Veterinary School in 1995. After spending five years in mixed practice in the North of England, Dr Weir returned to Glasgow on a Wellcome Trust Fellowship to study the molecular epidemiology of Cryptosporidium. Following a secondment to the State Veterinary Service to assist with the Foot and Mouth crisis in 2001, Dr Weir accepted a Ronald Miller Scholarship from Glasgow Veterinary School allowing him to pursue a PhD in Molecular Parasitology,

which he completed in 2006. Two years later, while undertaking post-doctoral research, Dr Weir was awarded a Master's degree in Bioinformatics. His research interests have developed in a number of areas including pathogen genetics, genomics and transcriptomics and he has published a total of 50 peer-reviewed articles. He is Principal Investigator on several projects investigating tick-borne pathogens and is currently funded to pursue research in the UK (Scottish Government, HBLB), Turkey (Turkish Research Council) and India (BBSRC). More recently he has applied his bioinformatic expertise to the identification and development of transcriptomic biomarkers for equine health and leads projects funded by the Wellcome Trust and The Donkey Sanctuary. In 2016, Dr Weir was appointed as Academic Head of the Veterinary Diagnostic Service Infectious Disease Unit at the University of Glasgow.

Publications

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CLINICAL PATHOLOGY AND ACUTE PHASE PROTEINS IN NEMATODE PARASITIC INFECTIONS

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Parasitic infections caused by nematodes are a daily routine issue in small and large animals' clinics resulting in losses in animal health, production and in some cases could represent potential zoonotic risks. In Botucatu, São Paulo, Brazil, our studies have been developed aiming to evaluate pathophysiological changes in naturally infected animals but without clinical signs of disease to understand subclinical infections, which could be considered in differential diagnosis and an indication of parasite infection. For that, a variety of biochemical markers including acute phase proteins have been screened to provide an overview of the biochemical effects produced by patent nematode parasitic infections in dogs and calves. Naturally infected dogs with *Ancylostoma* spp. (hookworm) demonstrated significant differences of selected acute phase proteins levels in serum such as C-reactive protein (CRP), haptoglobin, and also of insulin-like growth factor (IGF-1), albumin, unsaturated iron binding-capacity (UIBC), and iron concentrations. A particular acute phase protein profile response was found in dogs naturally infected with the giant kidney worm (*Dioctophyme renale*) with increased haptoglobin and cortisol without significant changes in CRP and serum amyloid A concentrations at the diagnosis time point. Although there was a significant destruction of the renal parenchyma, other routine biochemical analytes assessed were not affected by the parasites. Significant changes in biochemical analytes and acute phase proteins have been found in calves subclinically infected by gastrointestinal (*Cooperia* spp., *Haemonchus placei*, *Oesophagostomum* spp., and *Trichostrongylus* spp.) and pulmonary parasites (*Dictyocaulus viviparous*). Both GI and pulmonary parasites increased haptoglobin concentrations, only GI parasites caused decreases in the lipid profile and lungworms increased acetylcholinesterase activity. These investigations were enabled by interactions with the Institute of Biodiversity, Animal Health and Comparative Medicine of the University of Glasgow and the INTERLAB of the University of Murcia, Spain.

Biography

Professor Elizabeth Schmidt graduated with BSc in Veterinary Medicine from the University of Parana State (UFPR), Brazil in 1997; with a Master Degree in Veterinary Sciences in 2000, and from the Sao Paulo State University, Brazil (UNESP) in Veterinary Clinical Pathology with a PhD in Veterinary Medicine in 2008. She undertook a post-doctoral research study (FAPESP Grant) for two years (2008-2009) in Veterinary Pathology at Sao Paulo State University (UNESP), campus of Jaboticabal, Brazil. She spent six months as a training fellow (2013) in Veterinary Biochemistry

(Marie Curie Institute Grant - FP07 Nematode Health System) at the University of Glasgow working with Prof. David Eckersall. In 2015-2016 she spent twelve months working in a post-doctoral research project with biomarkers at the INTERLAB – University of Murcia, Spain (Science Without Borders – CNPq/Brazil, PD Grant). Since 2009 she is an Assistant-Professor of Veterinary Clinical Pathology and Parasitic Diseases at Sao Paulo State University (UNESP), Department of Veterinary Clinical Sciences – School of Veterinary Medicine and Animal Science, campus of Botucatu, Brazil.

Publications

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ZIKA: ANOTHER PUBLIC HEALTH THREAT TRANSMITTED BY *AEDES AEGYPTI*

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Zika (ZIKV), dengue (DENV) and chikungunya (CHIKV) are arboviruses transmitted to humans by the bite of infected *Aedes* females. Their main vector is the *Aedes aegypti* mosquito in Brazil, which is a domestic species linked to human activities. In recent years, the circulation of these viruses has drastically changed. A scenario, in which the circulation of DENV serotypes was predominant, is shifting to dengue co-circulating with the ZIKV and CHIKV. There were reports of febrile cases that were not related to DENV infection in various Brazilian regions at the end of 2014. In April 2015, ZIKV was identified as the etiologic agent of a number of febrile cases. In May of that year, the Brazilian Ministry of Health officially reported ZIKV circulation in the country. The spread of the virus is an additional challenge for the public health system, especially because of the risk of simultaneous infection by DENV and CHIKV, which present the same clinical symptoms. It is necessary to properly diagnose the etiologic agent of infection, so that control measures can be triggered to decrease ZIKV dispersion.

Biography

Adriano Mondini is a PhD in Health Sciences. A professor at the School of Pharmaceutical Sciences (UNESP), he teaches Public Health for graduate and undergraduate students. He has been working with arbovirus transmission since 2002. The main aspects of his research are related to molecular epidemiology of dengue and zika, virus dispersion, phylogenetic analysis, spatial distribution, and vector competence. His team is currently working on a study to perform arbovirus surveillance in febrile patients and mosquitos.

Publications

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NUTRITIONAL AND HORMONAL STRATEGIES TO IMPROVE REPRODUCTIVE EFFICIENCY IN BEEF AND DAIRY CATTLE

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A sequence of experiments were designed to evaluate the effects of excessive energy intake and supplementation with chromium propionate on insulin resistance parameters in lactating dairy cows, the effects of concentrate type and chromium propionate on insulin sensitivity, productive, and reproductive parameters of lactating dairy cows consuming excessive energy, the IGF-1, GH and leptin concentrations in 12 to 16 months old Nelore heifers and mature cows and its impact on reproduction, the effect of ground corn supplementation on Nelore cows reproduction, the effect of virginiamycin supplementation on reproductive performance of multiparous Nellore cows, the effect of interval from induction of puberty to initiation of a timed AI protocol on pregnancy rate in Nellore heifers, the effect of breed, progesterone serum concentration and eCG treatment in Bos indicus and Bos taurus x Bos indicus heifers submitted to a synchronization of ovulation protocol, the effect of adding a GnRH treatment at the beginning and a second prostaglandin F2α treatment at the end of an estradiol-based protocol for timed AI in lactating dairy cows during cool or hot seasons of the year and the effect of expression of estrus on fertility and pregnancy losses in lactating dairy cows that receive AI or embryo transfer.

J.L.M. Vasconcelos, A.D.P Rodrigues, M.H.C. Pereira, R.F.G. Peres, T. Leiva

Biography

Associate Professor of Infectious Diseases of Animals at the Department of Animal Production, School of Veterinary Medicine and Animal Science, São Paulo State University (UNESP), Botucatu – Brazil, 1989/up to now. BSc in Veterinary Medicine from the University of Minas Gerais State (UFMG) in 1980. MSc in Animal Science from the University of Minas Gerais State in 1985. PhD in Animal Science from the São Paulo State University (UNESP) in 1998. Post-doctoral research projects at the Institute of Food and Agricultural Sciences, USA in 2011, and at the Animal Sciences Department – Ohio State University in 2013.

Publications

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OPTIMIZATION MODELING WITH SPREADSHEETS: FINDING SIMPLICITY IN COMPLEXITY

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Models in research can find simplicity in complexity, because can explain and representation many challengers to real word. Then, the modelling process can be found issue investigated, with simplification, defining the essence of complex systems. There is good and simple models adequate to something complex, giving high utility with transparent, but bad models with returns of information complicated, although of simply phenomena. Then, the model to be used needs answers a specific question, contribution to performance, to save money or time, and applied to give a fuller understanding of the processes. The Practical program for Modeling -PPM (https://goo.gl/5je0GV) and Practical Program for Optimization -PPO (https://goo.gl/80rJHo) programs for estimating curves and optimization are freely available software downloads.

These programs are general purpose curve fitting and optimization for students and researchers of animal modelling, to capture the essence of complex problem, nonlinear and, many times, multi-dimensional. This is the way and the reason to use models in research.

Biography

Manoel Garcia Neto graduated from the São Paulo State University, campus of Jaboticabal with a BSc in Animal Science in 1985. Master degree in Animal Science from São Paulo State University (1989), PhD in Veterinary Medicine from the University of Minas Gerais State (UFMG) in 1993. Post-doctorate research projects at the University of Georgia, UGA, USA. Manoel Garcia Neto works with Animal Science, focusing on Nutritional Requirements of Animals (mgarcia@fmva.unesp.br).

Publications

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SUSTAINABLE CONTROL OF PARASITIC GASTROENTERITIS IN RUMINANTS

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Parasitic gastroenteritis caused by nematode infections is a major cause of economic losses in the livestock industry in Brazil because it impairs weight gain and increases mortality of cattle and small ruminants. In our region, the major gastrointestinal nematode (GIN) parasites are *Haemonchus placei, Cooperia punctata* and *Oesophagostomum radiatum* in cattle and *Haemonchus contortus, Trichostrongylus colubriformis* and *Oesophagostomum columbianum* in sheep. The environmental conditions in our area are extremely favourable for the year-round transmission of these nematodes, which leads farmers to an indiscriminate use of anthelmintic treatments. Consequently, in cattle the resistance to avermectins is widespread and in sheep is very common the presence of nematodes that exhibit multiple anthelmintic resistance. For this reason, the development of strategies that are less dependent on anthelmintic treatments is imperative for the prophylaxis of GIN infections in ruminants. The proper identification of the various nematode species is essential in epidemiological studies. Currently, we have being developing easily applied molecular and parasitological methods of identifying *H. contortus* and *H. placei* as well as their hybrids. This is essential especially when mix grazing of cattle and sheep is being employed in order to produce "clean pastures". So far, we observed that cross infections between sheep and cattle parasites are not significant, which allows the design of grazing strategies using different ruminant species to produce clean pastures. The integrated crop-livestock systems are another option to reduce the risk of heavy infections in ruminants. Supplementary feeding and breeding strategies to improve resistance to nematodes are also feasible options in the effort to reduce dependence on anthelmintic drugs to control worm infections. Nelore beef cattle present a high degree of resistance against ticks and hemoparasites, but they may present high worm burdens. With regards to the immune response against GIN infections, our studies indicate that it is possible to select Nelore cattle for resistance without jeopardize its productivity. The Santa Inês hair sheep, a naturalised breed, exhibit genetically related resistance against nematode infections compared with commercial breeds of European origin. The major problem with both Santa Ines sheep and Nelore cattle is that its carcass is considered of inferior quality. To overcome this problem, a good option has been the crossbreeding of ewes or cows of these breeds with sires of breeds with high potential for growth and good quality meat production. This strategy results in crossbred animals with high productivity and a satisfactory degree of resistance against parasites. Improvement in nutrition has also a beneficial effect on the development of resistance in young animals naturally or artificially infected with GIN. Finally, we have been testing a vaccine against haemonchosis containing native *H. contortus* intestinal glycoproteins, which have afforded significant protection to lambs and calves in both pen and field trials.

Workshop in Internationalisation in Veterinary Sciences: perspectives for research between UNESP and the University of Glasgow, Scotland, UK, October 10th to 12th 2016, Fazenda do Lageado, FMVZ – UNESP, Botucatu, SP, Brazil.

Biography

Alessandro F. T. Amarante is currently Professor of Veterinary Parasitology at Universidade Estadual Paulista in Botucatu, Sao Paulo State, Brazil. His research focuses on the Gastrointestinal Nematodes of Ruminants and he has published over 100 peer-reviewed scientific papers. His projects in development include studies about genetics of *Haemonchus* spp. and vaccine against haemonchosis. These researches are being developed in co-operation with scientists from Calgary University – Canada and Moredum Research Institute – UK, respectively. He is also involved in studies about immunology, epidemiology and anthelmintic resistance with a group of 2 pos-doc, 5 PhD and 1 MSc graduated students under his supervision.

Publications

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LIVESTOCK GENOMICS: THE PERSPECTIVES OF ZEBU CATTLE

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After the huge success of genomics in the taurine dairy world, which has impacted in the entire genetics production chain in breeds such as Holstein and Jersey, several attempts to implement this "golden model" into the tropical cattle (either dairy or beef) has shown limited success up to date. The bottlenecks are less scientific or technical and more on the scale and particularities of the current breeding programs in countries such as Brazil. Alternative methods such as genome wide association studies (GWAS), runs of homozigosity, signatures of selection and haplotype oriented matings will be discussed in the presentation, with emphasis on the improvement of methods and processes for breeding relying on the concept of "functional genomics", where the use of information related to the genes themselves (or the use of their genomic coordinates to indirectly access their effects) is proposed. The functional view of the genomics and its exploration can be of extreme value in situations such as crossbreeding (taurine x indicine), highly prevalent in the tropical world and used to increase productivity without losing rusticity. Besides this application, the use of genomics can provide additional value by combining several applications in one single test (parentage, genetic defects detection, genomic and functional selection, breed/product traceability). We will discuss the potential of using genomic information to increase the speed of breeding and selection in an economically sustainable way, which is the major requirement in indicine populations nowadays. Finally, we will touch base on the use of gene editing methods to move the genetic progress even faster.

Biography

Associate Professor on Animal Biotechnology, School of Veterinary Medicine (FMVA), São Paulo State University (UNESP), Araçatuba – Brazil, 1997/up to now. BSc in Veterinary Medicine – University of São Paulo (USP), Brazil – 1989. MSc in Livestock Parasitic Diseases – Federal University Rio Grande do Sul (UFRGS), Brazil – 1992. PhD in Animal Reproduction – University of São Paulo (USP), Brazil – 1995. Master of Business Administration (MBA) – Fundação Getúlio Vargas (FGV), Brazil – 2008. Four sabbatical periods in livestock genetics and genomics (around two months each). Università Cattolica, Department of Animal Sciences, Piacenza – Italy (2010). USDA, Bovine Functional Genomics Laboratory, Beltsville – USA (2011, 2012 and 2014). Scientist and Technical Officer at the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, Vienna – Austria, 2003-2005. Vice Executive Coordinator of the UNESP Biotechnology Institute, Botucatu – Brazil, 2012/2014.

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EPIDEMIOLOGY AND PATHOGENESIS OF PERIODONTITIS IN RUMINANTS

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Periodontitis in ruminants is a dramatic and scientifically challenging disease. Its high prevalence, associated with the management of pastures in extensive regions of the Atlantic Forest biome, savanna, wetland and Amazon is responsible for significant economic losses to animal health and welfare in Brazil. It has been verified a variable prevalence of chronic or aggressive forms of the disease in cattle, sheep, goats and deer in captivity and fed with the best feed and diets. But often periodontitis is not noticed by producers and is rarely diagnosed by veterinarians. An epidemiological feature of periodontitis in herds is the reduction of its incidence over the years; however, in endemic areas, there is an intensification of disease outbreaks after the adoption of modern agricultural practices. Putative periodontal pathogens recognized in humans and other animals are present in the lesions. However, the possible risk factors that determine the disease are still unknown. Currently, the challenges are to evaluate the prevalence of periodontitis and its economic impact on animal production, especially in the Amazon region, whose low productivity of cattle contributes to the pressure by opening up new areas of forests. Similarly, we intend to develop in the elucidation of possible risk factors, focusing on its effects on oral bacterial dysbiosis and the mechanisms that trigger the inflammatory process that promotes the destruction of the periodontal tissues.

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Biography

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FOLLICULAR FLUID PROTEOME PROFILE OF DAIRY COWS

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Bovine follicular fluid (FF), secreted by granulosa and theca cells, has been shown to contain biologically active molecules and proteins that may affect follicular growth, oocyte maturation and ovulation. Follicular fluid proteomic profile has a significant impact in the identification of biomarker for oocyte quality estimation and, maybe, for in vitro fertilization success improvement, however is still poorly characterized. We performed a study involving healthy non-lactating Holstein cows to determine the proteome profile of FF in key-stages of the follicular development. Follicles were aspirated in vivo at predeviation, deviation, post deviation and preovulatory stages of the estrous cycle, which were confirmed by measurement of estradiol and progesterone concentrations. Pooled FF samples from each stage were depleted, reduced, alkylated and digested with trypsin. The resulting peptides were labelled with Tandem Mass Tag (TMT-6 plex; Thermo Scientific) according to the protocol supplied by manufacturer. Proteins were identified using LC-MS/MS (Orbitrap Elite ETD) and data analysis was carried on Proteome Discoverer^{TM} 2.1 software. A total of 524 proteins were identified and assigned to a variety of functional processes, including protein binding, enzyme regulator activity, metal ion binding, and catalytic activity. Twenty-two differentially ($P<0.05$) expressed proteins were found between stages indicating intrafollicular changes, with deviation a critical time point for modulation of follicular growth. For instance, inhibin, follistatin, serglycin, protein HP 20, anti-testosterone antibody, complement C4, fibrinogen, amongst other, were shown to correlate with the first stages of the follicular development. In contrast, alpha-2-macroglobulin, beta-2-glycoprotein, antithrombin-III and immunoglobulins were altered during later stages of the estrous cycle. In conclusion, these differentially expressed proteins provide insights into the size-dependent protein changes in the ovarian follicle microenvironment that may influence follicular function.

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IDENTIFICATION OF THE BACTERIA AND EVALUATION OF TISSUE LEVELS OF TOLL-LIKE RECEPTOR AND CYTOKINE MRNAS ASSOCIATED WITH BOVINE PERIODONTITIS AND ORAL HEALTH

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Bovine periodontitis is an oral disease that occurs under specific epidemiological conditions and is predominantly associated with presence of anaerobic bacterial microflora in the subgingival biofilm. It is now increasingly apparent that bovine periodontitis is likely to impact significantly on the welfare of affected animals, since it can lead to difficulty in feeding which consequently leads to loss of body condition, weight loss and decreased productivity. This evidence suggest that periodontitis may be a hidden financial loss to farmers and a reason for culling cows at an earlier age than expected. Toll-like receptors play an important role against invading pathogens and one of the most important strategies utilized by the host immune system to defend against microbial challenges is to produce and secrete cytokines. The aim of this study was to investigate the expression of TLR4, TNF-α, IFN-ɣ, IL-1β, and IL-4 mRNAs in 20 orally healthy bovine and 20 with periodontitis, by quantitative PCR. In addition, we used high-throughput 16S rRNA gene sequencing to determine the composition of the complex microbiota associated with bovine periodontitis (40) and oral health (38). The abundance of mRNA encoding TLR4 ($p<0.01$), TNFα ($p<0.01$), IL-1β ($p<0.001$), IL-4 ($p<0.01$) and IFN-ɣ ($p=0.01$) was increased in the periodontitis samples compared to the healthy samples and this increase was statistically significant. The analysis of partial results of high-throughput sequencing showed the prevalence of phyla Proteobacteria and Actinobacteria in the periodontally healthy animals and the phyla Fusobacterium, Bacteroidetes, Firmicutes and Synergistetes in the group with periodontitis. In conclusion, bovine periodontitis appears to be associated with an increase in expression of TLR4, TNF-α, IFN-ɣ, IL-1β, and IL-4 mRNAs and the two cohorts of cattle examined harboured distinct profiles.

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PROTEOMIC INVESTIGATION OF DIFFERENTIALLY EXPRESSED PROTEINS IN BUFFALO (*BUBALUS BUBALIS*) MILK WHEY DURING MASTITIS

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The aim of this study was to investigate, by performing conventional 2D-electrophoresis (2-DE) and 2D-DiGE, modifications of the milk whey proteome profile in buffaloes during mastitis. Milk samples were collected from healthy (G1, negative bacteriology, SCC<100,000 cells/mL) and mastitic (G2, positive bacteriology, SCC>100,000 cells/mL) buffaloes. To perform 2-DE, samples with low (0.10-0.50 μg/mL, n=6) and high (1.41-12.08 μg/mL, n=12) haptoglobin levels were selected from G1 and G2, respectively. To perform 2D-DiGE, pools with low (0.10-0.44 μg/mL) and high haptoglobin levels (5.98-12.08 μg/mL) were prepared using milk whey samples from G1 and G2, respectively. 2-DE was accomplished by loading 200μg of total protein of each sample into 11cm, pH3-10 IPG strips, followed by SDS-PAGE. 2D-DiGE was accomplished by loading 50μg of total protein of two different pools (healthy vs mastitis), labelled with fluorescent dyes (Cy3, Cy5), into 24cm pH4-7 IPG strips, followed by SDS-PAGE. Gel images were analyzed using SameSpot (2-DE) and DeCyder software (2D-DiGE). Spots of interest were excised and subjected to tryptic in-gel digestion and analysed by LC-ESI-MS/MS. Protein identifications were assigned using NCBI databases. 2-DE highlighted ten spots differently expressed during mastitis. From these, seven spots were increased, where host-defence proteins (lactoferrin, complement C3, imunoglobulin light chain) and clusterin were identified. Three spots were decreased, where high abundance proteins (β-Lactoglobulin, α-Lactalbumin) were identified. 2D-DiGE highlighted 29 spots differently expressed during mastitis. From these, 15 spots were increased, where host-defense proteins (lactoferrin, complement C9, imunoglobulin light chain, endopin 2B and apolipoprotein A1) and clusterin were identified. 14 spots, identified as β-lactoglobulin and α-lactalbumin were decreased. In conclusion, 2-DE and 2D-DiGE allowed a comparison of protein spots between healthy and mastitic buffalo milk whey samples and the two techniques combined could give important information regarding potential biomarkers of mastitis. This project received financial support from FAPESP (2013/26498-5) and University of Glasgow.

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CORTICOSTERONE LEVELS IN BLOOD SERUM MEASURED BY ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) AS PARAMETER OF WELFARE IN BROILERS

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Transport is a stressful process that can result in mortality, injuries and increased physiological stress with associated effects on meat quality. In the UK, over 700 million broilers (meat chickens) are produced annually, and all of these birds are transported to abattoirs for slaughter. Data were collected from a large abattoir in Midlands Region of England. Corticosterone levels in blood serum (measured by enzyme-linked immunosorbent assay ELISA test) were examined over the winter in three transport distances from farm to abattoir (S=short, 0-50km, M=medium, 51-150km and L=long, 151-300km), in December 2015 and January 2016 (312 blood samples: 104 for each treatment). Regarding corticosterone levels, English data showed that levels decrease when transport is longer. Among the distances the mean (SD) was 1,137.07pg/ml (378.16), 991.46pg/ml (292.66) and 704.95pg/ml (176.50) in short, medium and large distances respectively. In the UK, corticosterone levels were significantly lower for long transport compared to short and medium which did not differ from each other. Brazilian corticosterone data will be collected in December 2016 (summer in Brazil), season more stressful for the broilers, to compare with the English data of winter (season more stressful for the broilers in England). Concluding, long transport in the UK was less stressful (as measured by corticosterone levels) than short or medium distance transport, probably because the animals get more stressed right after hanging and loading. In Brazil, the results can be different, with higher levels in large distances, because the hot weather can be more stressful than the hanging and loading.

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MORTALITY AND INJURIES IN BROILER TRANSPORT: COMPARISON BETWEEN A BRAZILIAN AND AN ENGLISH SLAUGHTERHOUSE

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In the UK, over 700 million broilers are produced annually while in Brazil the equivalent number is 6 billion; all of these birds are transported to abattoirs for slaughter, being transport stressful process that can result in mortality and injuries. Data were collected from two large abattoirs, one in Brazil and one in UK. Numbers of broiler birds dead on arrival (DOA) and numbers of injuries on post-mortem inspection were examined over two seasons (summer and winter) and three transport distances from farm to abattoir (S=short, 0-50km, M=medium, 51-150km and L=long, 151-300km). DOA and injuries in Brazil was measured in 2013 (n=5.632.767 broilers) and in UK in 2015 (n=7.183.193 broilers). DOA was higher in Brazil than in England (0.72% vs. 0.13% overall). In Brazil, in winter there was no relationship (rs=0.092; P=0.33) between DOA and distance while in summer there was positive correlation (rs=0.203; P=0.035). In both seasons there was a positive correlation between injures and distance [rs=0.273; P=0.004 (winter) and rs=0.286; P=0.003 (summer)]. In UK, long distance winter transport resulted in higher mortality (P<0.01) but there were no season/distance interactions in summer. Percentages of birds with injuries were higher in Brazil than the UK (0.97% vs. 0.06%). Concluding, DOA were higher at a large Brazilian abattoir than an equivalent abattoir in the UK, observing that mortality in transport is related to heat stress in Brazil and cold stress in the UK.

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TEST SPOT ON THE LAWN FOR DETECTION QUORUN SENSING

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Research such as the use of probiotics with pro detection therapies and anti-quorum work by modulating bacterial chemical communication circuits have become a reference and an alternative to the use of antibiotics, for most of the bacteria have a self-inducer to communicate quorum sensing, which is a process of cell-cell communication that controls the Bacteria-year collective behavior (HOANG 2015). In the present work we tested the existence of quorum sensing between *Lactobacillus* spp. and the pathogen Salmonella Enteritidis through the test spot on the lawn. For the experiment the control sample (AC) used was a *Lactobacillus* spp. resistant to antibiotics rifampin (RIF) and nalidixic acid (NAL). In the tests where the AC had contact with the pathogen sensitive NAL / RIF was grown together with the AC in DeMan-Rugosa-Sharpe broth (MRS) and Brain Heart Infusion (BHI) broth and incubated. A-post this culture was transferred to MRS broth containing RIF / NAL and incubated so that in this way the pathogen was inactivated and remained only the Lactobacillus spp. and further cultured in MRS only. The tubes were centrifuged and filtered AC to obtain a cell-free supernatant. In this filtrate was added 10-shows the Lactobacillus spp. and incubated separately. After the samples of *Lactobacillus* spp. They were subjected to the test spot on the Law (SANTOS, 1993) against Salmonella Enteritidis. This same methodology was used to test the samples of Lactobacillus spp. in which the AC had no contact with Salmonella Enteritidis. After incubation the halos were measured. It was concluded that there was quorum sensing between Lactobacillus spp., since the average of the halos of the cultured samples of broth filtrate AC without contact with the pathogen were smaller than those grown in broth AC had contact with pathogen. References: Hoang, D. Lu, et al. Modulating Vibrio cholerae Quorum-Sensing-Controlled Communication Using Auto-inducer Loaded Nano particles. Nano Letteres, v.15, p.2235-2241, 2015. Santos, W.L.M. Aislamiento y partial caracterizacion una bacteriocina producida by Pediococcussp. 347, Carnico of origin. Thesis (Ph.D.) - Complutense University of Madrid 1993.

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SELECTION AND CHARACTERIZATION IN VITRO PROBIOTIC LACTOBACILLUS SPP

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For improvements in poultry production, which Brazil is the position of this that worldwide (ABPA, 2016), and the growing concern with the bacteria resistance to antimicrobials that are not fully effective in fighting pathogens, reduce the natural microflora of the gastrointestinal tract of birds, and increase the colonization of it by microorganisms (BELCHIERI and NETO, 2009; OIE, 2015), the search industry to introduce changes that improve the processes of creation. The lacto-bacilli are bacteria that are part of the natural microflora and are used as probiotic replacing antimicrobial, protecting the gastrointestinal tract against the action of micro-organisms such as Salmonella Heidelberg, harmful pathogens to animal and human health. This work was proposed to characterize probiotic in vitro de 170 samples of bacteria from feces of turkeys. The samples were tested for resistance to gastric pH between 2.5 and 7.0, bile salts with Oxgall enzyme potential hydrophobicity with Hexadecane enzyme potential multiplication, production of hydrogen peroxide with use of TMB-plus modified antagonism against Salmonella Heidelberg with tests *spot on the lawn, radial streak, agar well difussion, liquid coculture assay and cross-streak* modified, antibiotic susceptibility test with 12 antimicrobials and antimicrobial resistance genes integrons C. As the best-selected samples in all analyzes were submitted to sanger sequencing. The results were analyzed using Bioedit Mega7.0 and programs, and readings made at the National Center for Biotechnology Information. It is concluded that the samples analyzed, 11 were selected, one of *Lactobacillus frumenti* 9 *Lactobacillus reuteri* and *Lactobacillus johnsonii*, all with probiotic potential, proven in vitro, and election for use invivo.

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PPO BROILER CHICKEN: A PROGRAM FOR DETERMINATION OF OPTIMAL MARKET AGE OF BROILERS

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Broiler production has been increasing over the years and has a high economic return due to its short production cycle. The production indexes, like body weight gain, feed intake, age, mortality, are very known and extremely utilized. However, the interpretation of these indexes is done separately, making difficult the profitability analysis. The costs in broiler production is complex, incurred mainly to feed and operational costs, and profits, coming from the sale of the product, as the live bird or whole carcass, are dynamic, ranging through each production and according the market conditions and consumers preferences. The mathematical modelling can associate different indexes, providing a better understanding of the results and investments returns. The Practical Program for Optimization (PPO/ http://goo.gl/80rJHo) is a Microsoft Excel-based workbook that was developed aiming to provide a new referential in the business management, ensuring a better vision of the production. The PPO allows that the user make comparisons between different scenarios. The profit-maximizing analysis is composed of three stages. First, data about the cumulative mortality, body weight and cumulative feed intake have to be inserting. Then, the bottom "Fitting the NarushinTakma Model" have to be pressed to occur the curve fitting, that is made through the Solver feature of Excel, firstly been activated the Evolutionary Solving method and, after, the GRG Non Linear Solving method. The mathematical model was proposed by Narushin and Takma (2003), been composed for three equations and is chosen that have the better goodness of fit (R^2). Second, inputs about the broiler price ($/kg), broilers housed, price of feed, according the age, and operating cost have to be included. Finally, the bottom "Solver" has to be activated, to give the estimation of the optimal market age.

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IN VITRO EVALUATION OF THE INHIBITORY ACTIVITY OF LACTOBACILLUS SPP. WITH PRIOR CONTACT AGAINST SALMONELLA SPP

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BA Nagayoshi
EL Mibradt
TM Silva

Commercial poultry has as target high productivity, producing protein with quality and low cost, as matched against infections caused by *Salmonella* spp. can reduce productivity and cause toxic infection in humans, causing restriction on the use of some antibiotics in animal production, consequently stimulating the interest in the use of probiotics, as already reported that some species of probiotics have activity on *Salmonella* spp. The bacteria notice changes in the environment and modify their behavior to survive and thrive by means of a collective response, called quorum sensing mechanism, in which the bacteria produce and release auto inducers that regulate physiological activities as production of bacteriocins with antibiotic activity. And with that the objective of this study was evaluate the inhibition capacity of seven *Lactobacillus* sp. front of *Salmonella* spp., where they were grown in filtered lactobacillus broth that had come into contact with Salmonella spp. the inductor. To assess the potential inhibition was carried out at spot-on the-lawn technique with modifications. The results of seven samples of *Lactobacillus* sp. analyzed showed a biggest inhibition halo twice *Salmonella* spp. when previously cultured with the inductor. And it can be concluded that there is interaction between *Lactobacillus* sp. and the inductor.

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Workshop in Internationalisation in Veterinary Sciences: perspectives for research between UNESP and the University of Glasgow, Scotland, UK, October 10th to 12th 2016, Fazenda do Lageado, FMVZ – UNESP, Botucatu, SP, Brazil.

INFLUENCE OF ENVIRONMENTAL EFFECTS IN THE LOSS OF EGGS OF PATRIDGES FOR ARTIFICIAL INCUBATION

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During the breeding season of partridge (*Rhynchotus rufescens*), males are responsible for natural incubation of the eggs and care about young patridges. Artificial incubation of eggs is recommended to avoid the loss by pecking due to male carelessness. The aim of this study was to evaluate the environmental factors of male and female proportion (M:F), average age of parents (AA) and number of animals by pen (NPEN) for egg loss percentage (ELP) of population of partridges *Rhynchotus rufescens.* The experiment was conducted from October to December of 2015 in the Setor de Animais Silvestres of Faculdade de Ciências Agrárias e Veterinárias (UNESP). 87 males and 67 females of patridges *R. rufescens* were randomly distributed in 22 pens with six different M:F (2:4, 3:4, 1:1, 3:2, 5:3 and 5:2). A total of 247 eggs were collected. ELP by pen was analyzed by least-squares with the GLM procedure of the SAS program. The statistical model was defined by fixed effects of M:F and NPEN and covariate effect of AA. Means comparisons were performed using the Tukey-Kramer test ($P<0.05$). The effects of M:F and NPEN were statistical significant ($P<0.05$) for ELP. Means of ELP for the proportions were 32.1%, 27.2%, 32.3%, 5.6%, 20% and 24% for 2:4, 3:4, 1:1, 3:2, 5:3 and 5:2, respectively. Only the means of 1:1 and 3:2 proportions showed statistical difference ($P<0.05$). Results of these study showed M:F and NPEN influenced the occurrence of broken eggs. Strategies considering M:F and NPEN must be implemented to minimize the losses of hatching eggs, targeting higher profitability for commercial creation of partridges.

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CHEMICAL COMPOSITION AND ENERGY VALUES OF DIFFERENT CORN CULTIVARS FOR MEAT-TYPE QUAILS

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AC Furlan[2]
MR Fachinello[2]
SL Ferreira[2]
LMD Huepa[2]
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Corn is a widely used ingredient in diets for quails and contributes with a high content of metabolizable energy (ME), however, the ME values differs among corn batches due to its variation in the chemical composition. The objective of this study was to determine the chemical composition, the apparent ME (AME) and the AME corrected for nitrogen balance (AMEn) of different corn cultivars for meat-type quails. A metabolism assay was realized being used 160 meat-type quails, males, averaging 22 days of age, distributed in a completely randomized design with eight treatments, four replicates and five animals per experimental unit. Treatments consisted of a basal diet (BD) and seven corn varieties, which replaced 20% of the BD. The feed intake and excretas were quantified during the trial period and gross energy (GE) of corn cultivars, diets and excreta were determined using an isoperibolic calorimeter. The values of crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), acid detergent fiber (ADF), mineral matter (MM) and phosphorus (P) of the corn cultivars were determined. The AME:GE and AMEn:GE, expressed in percentage, were submitted to the SNK test at 5% probability. The CP, EE, NDF, ADF, MM and P ranged, respectively, from 7.55 to 9.01%; 2.94 to 4.22%; 3.36 to 4.29%; 10.89 to 11.82%; 1.03 to 1.22% and 0.229 to 0.311%. This variation in chemical composition may be related to the type of seed, climate, fertilizer and grain storage, but these changes in the chemical composition did not affect (P>0.05) AME:GE and AMEn:GE that ranged from 86.34 to 90.88%; and from 83.87 to 88.07%, respectively. It is concluded that there were no differences in the metabolization of GE between the studied corn cultivars for meat-type quails, with values of AME and AMEn ranging from 3868.88 to 4068.25; and 3758.27 to 3942.21 kcal/kg, respectively.

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IN VITRO PRODUCTION OF BOVINE EMBRYOS EVALUATING DIFFERENTS BUFFER SYSTEM IN TWO GASEOUS ATMOSPHERES

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VM Codognoto
PH Yamada
FR de Ruediger A Dantas
PF Lainetti
E Oba

The development of a complete *in vitro* culture medium that meets all the requirements of the embryo has been the objective of several studies. The presence of sodium bicarbonate and its interaction with the temperature and gaseous atmosphere still require an incubator with O2 and CO2 atmospheric control, since none of these buffering agents provides sufficient stability for the maintenance of embryonic development under atmospheric conditions. The aim of the present study was to investigate the effect of different buffer systems in the pH stability of culture medium and in the developmental capacity of IVP embryos. After fertilization, the zygotes were cultivated according with treatments (T): T1- control group with NaHCO3 in medium HTF Irvine Scientific®; T2- group with 25mM NaHCO3 in medium SOFaa; T3- group with 9mM NaHCO3 and 4mM HEPES in medium SOFaa and T4- group with 9mM NaHCO3 and 12mM sodium phosphate in medium SOFaa. The embryos were exposed in two ambient: atmosphere with 5% CO2 + 5% O2 + 90% N2 at 39°C and atmosphere with 5% CO2 in air at 39°C and 100% humidity. The results obtained in this work showed that the addition of low concentration of HEPES to the culture media did not negative influence for blastocyst formation. Media buffering depends on the gaseous atmosphere utilized during culture and embryo development in sensitive to pH variation during culture, especially in bench manipulations. The pH stability obtained in these conditions didn't damage embryonic developmental and may be beneficial for embryo quality during cleavage rates evaluation and feeding when embryos were exposed to ambient atmosphere.

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IS THE MORPHOLOGY OF IN VITRO MATURED BOVINE OOCYTES ASSOCIATED WITH EMBRYO QUALITY?

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The aim of this study was to evaluate the nuclear maturation rate and in vitro embryonic development of oocytes from slaughtered bovine females in unknown cyclical phase considering the morphological classification criteria of *Cumuli Complexes*-oocytes (COCs). Oocytes were obtained from abattoir ovaries, transported in thermal container, stored between 30 to 35° C, containing saline solution of 0.9% NaCl plus 100 mg of Streptomycin and 50,000 IU of penicillin G-Potassium. 145 COCs were handled, sorted and matured for 24 hours in medium TCM-199 plus 10% BFS, 50 μg/mL gentamicin, 2 Mm sodium pyruvate, 20 Mm HEPES, 0.5 μg/mL FSH, 0,03 IU/mL of LH at 38° C and 5% CO2 in air. In the first step of the experiment, the oocytes were distributed in 3 treatments (T) proposed by Leibfried & First (1979): T1-COCs quality 1; T2-COCs quality 2 and T3-COCs quality 3 and 4. The evaluation of chromosomal configuration of *in vitro* matured oocytes did not indicate statistically significant differences for the nuclear maturation between the COCs degrees. In the second stage of the experiment, the oocytes were matured and fertilized as the 3 previous treatments and the T4-pool of COCs with all quality degrees. Were used to profile analysis a significance level of 5%. Cleavage averages were: T1 (18); T2 (16); T3 (11) and T4 (17) and morula: T1 (14), T2 (11); T3 (4) and T4 (9). The results indicated that regardless of the morphological classification and the females' estrus cycle stage, after 24 hours the co-cultivated oocytes had ability to develop in vitro to the MII stage in all treatments, before fertilization. After fertilization, just T3 presented embryonic development lower than the others, indicating that COCs quality 3 and 4 did not provide satisfactory development when they were grown separately.

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HISTOPATHOLOGY OF DIFFERENT CULTURE PROTOCOLS OF BOVINE ENDOMETRIAL EXPLANTS

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L Maia
CN Moraes
FC Landim-Alvarenga
JCP Ferreira*

The aim of this study was to test qualitatively different techniques to culture bovine endometrial explants and determine the best one to preserve tissue characteristics. For this, three bovine uteri presenting corpus luteum phase 1 (n = 2) or 2 (n = 1) were obtained from slaughterhouse. Then, the uteri were opened and endometrium was rinsed in alcohol for 30 seconds and then in saline solution. The endometrium and myometrium was separated, and endometrial fragments were manually obtained by successive cuts of 1mm3. The fragments were weighed and samples of 50mg assigned to one of the following conditions: A) 6-well plate treated with polylysine for tissue adhesion; B) 6-well plate using fetal bovine serum (FBS) for tissue adhesion; C) 3D system using a 15ml tube; D) 6-well plate with fragments on a 1.5% agarose support, measuring 10x10x5 mm, immersed in the culture medium; E) 6-well plate with no treatment. For all conditions, the maintenance medium was DMEM high glucose, 10% FBS, 100 IU/ml penicillin, 100 μg/ml streptomycin, amphotericin 2,5 μg, 10 μl/ml ITS (insulin, transferrin and selenium). After 24 hours, the medium was replaced and the explants were culture for more 24 hours. Fragments were collected before and after the 48-hours culture, fixed in formalin to perform histopathology. The integrity of endometrial glands, presence of indicative cellular response to stress such as atrophy, hypertrophy, hyperplasia and metaplasia, as well cell injury and death were analyzed. Treatments A and B did not allow the maintenance of adhesion to the plate, as was intended. The treatment E allowed the highest cell survival rate. On the other hand, treatment C resulted in the highest autolysis. We conclude that, in experimental conditions, the culture technique that better allowed the maintenance of histological integrity of bovine endometrium, for further application in in vitro models, was E.

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GENE EXPRESSION OF IN VITRO MATURATED OOCYTES CAN BE MODULATED BY FOLLICLE EXOSOMES FROM COWS KEPT UNDER THERMO-NEUTRAL OR HEAT STRESS CONDITION

FM Dalanezi[1]
JCP Ferreira

There are several intrafollicular agents that have the ability to interfere with the development of the oocyte; among these we highlight the exosomes (EXOs). Thus the aim of this study was to evaluate the capacity of EXOs extracted from the follicular fluid (FF) of cows kept under thermo-neutral (TN) or heat stress (HS) conditions to modulate oocyte maturation in vitro. Twenty-four Holstein cows were subjected to the following treatments: HS or TN for 14 days, and had their follicles aspirated. All FF from cows was pooled forming the groups (HS and TN). The EXOs were obtained by ultracentrifugation of FF. *Bos indicus* cumulus oocytes complex (COC) collected from ovaries obtained in commercial slaughterhouse, were pooled in groups of 20 COCs and randomly subjected to one of the following treatments: *Control*–maturated in standard medium; *HS-EXO* or *TN-EXO*–matured in standard medium added with 10 µl of a solution of follicular EXOs from HS or TN cows respectively. The procedures were repeated four times. After 22 hours of maturation, COCs were recovered and the expression of genes related to cell viability (BCL2, CDCA8, CPT1B, STAT3, RPL15), oocyte maturation (BMP15 and HAS2) and heat stress protection (HSF1) were assessed. Statistical test used was ANOVA and Tukey. All genes, except CPT1B, showed lower expression in TN-EXO oocytes when compared with control and HS-EXO ($P<0.05$). CPT1B showed a higher expression in HS-EXO oocytes ($P<0.05$). The results showed that the addition of EXOs from exogenous follicles can modulate the expression of oocytes genes. The lower expression of these genes in TN-EXO, suggested that the EXOs obtained in TN condition attenuate several genes related to the oocytes viability. Surprisingly, the control oocytes showed a similar gene expression pattern of the HS-EXO. In conclusion EXOs derived from FF of cows submitted to a TN or HS condition can modulate the gene expression of oocytes maturated in vitro.

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GENE EXPRESSION IN THE CORPUS LUTEUM FOLLOWING INTRAUTERINE PULSES OF LOW DOSES OF PROSTAGLANDINS E1 AND F-2 ALPHA IN CATTLE

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JC Mota[5]
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R Median[6]
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In ruminants, natural luteolysis is triggered by the uterine release of pulses of prostaglandin F2alpha (PGF), whereas prostaglandin E1 (PGE1) is considered to be a luteoprotective mediator. This study was designed to study the effect of low doses of PGE1 (2mg / infusion) infused into the uterine lumen, on the luteal responses to intrauterine PGF (0.25 mg / infusion). Cows on day 10 of the estrous cycle received intrauterine infusions of 4 different treatments: saline (4 infusions; n=5), PGE (4 infusions; n=5), PGF (4 infusions; n=5), and PGE+PGF (4 infusions; n=5) at 6-h intervals in a 2 X 2 experimental design. Progesterone (P4) concentrations were determined and luteal volume was evaluated by ultrasonography. Concentrations of PGFM and PGEM were measured before and 10 minutes after the first two infusions. A luteal biopsy was collected from each cow at 30 minutes after the third infusion in order to determine gene expression. Concentrations of PGFM after infusions were greater in cows receiving treatments with PGF and PGE+PGF than in saline or PGE-treated cows. Concentrations of PGEM after infusions were greater in cows that were treated with PGE and PGE+PGF than in saline and PGF-treated cows. Concentrations of P4 in the PGF group decreased compared to those in the saline group by 12 h (48.9% of control) after first infusion of PGF, at 24 h (20.2% of control), and all subsequent time points ($P < 0.05$). No differences in P4 concentrations were found between Saline, PGE, and PGF+PGE. There was a decrease of luteal volume between the PGF group and the other three groups that was detectable at 24 (56.4% of control), 48 (30.6% of control), and 72 (20.4% of control) h after PGF treatment ($P<0.05$). There were no differences in luteal volume between saline, PGE, or PGE+PGF. Gene expression in the luteal biopsy indicated a typical response to the PGF treatments (FGF2, EGR1, FOS and FAS increased; PTGFR, VEGFA, NR5A1 and STAR decreased) and that simultaneous PGE1 treatment completely blocked these gene expression changes. In summary, simultaneous intrauterine infusions of PGE blocked the luteolytic actions of intrauterine PGF pulses in cattle. Funding was provided by WI Experiment Station as Hatch Project WIS01240 to MCW and by BARD IS-4788-15.

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MOLECULAR DIAGNOSIS FOR CASES OF INTERSEXUALITY IN CATTLE

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LSLS Mota[1]

The cattle-ranching is one of the highlights of Brazilian agribusiness in the world scenario. The occurrence of abnormality during the sexual differentiation and / or embryo development is described in cattle. These abnormalities cause damage to the sector, since they are associated with reproductive problems including sterility or spread of aberration by their descendants. In many cases, analysis of the chromosomes of the affected animal is not being requested molecular diagnosis. Considering the economic importance and the damage caused by infertility or subfertility, this research evaluated the causes of deformities found in a bovine animal through the analysis of its DNA and, therefore, establish a protocol for this diagnosis. A undefined breed (UB) bovine, aged two years and six months presenting slightly masculinized female phenotype was forwarded to the veterinary hospital. The animal had changes throughout the female genital tract, with anomaly of the vulva and vagina. After slaughter, it was verified the presence of testicles in the reproductive tract. DNA was extracted from a blood sample and subsequently subjected to polymerase chain reaction using the set of primers AMELX and TSPY. The amelogenin gene (AMEL) is responsible for encoding an important protein in the formation of teeth enamel and, in most mammals, has an allele located on chromosome X (AMELX) and other allele on chromosome Y (AMELY) with differences in size and nucleotide sequence. Moreover, the TSPY primer amplifies a specific gene region of the Y chromosome related to spermatogenesis. After an analysis in agarose gel of the amplified products, it was found the presence of a fragment of 241pb regarding AMELX region and a fragment of 200pb specific region Y. For this result, we discard the occurrence of freemartinismo and androgen sensitivity syndrome. We suggest that this is a case of pseudo-hermaphrodite involving the translocation of a Y chromosome segment of the X chromosome.

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CHARACTERIZATION OF ANGIOGENIC FACTORS PRESENT IN CORPORA LUTEA OF PREGNANT AND NON-PREGNANT BITCHES

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AAP Derussi

The luteal phase in dogs is very similar among pregnant and non-pregnant females. This similarity is one of the most interesting features about the reproduction of canids and although numerous studies have been conducted, the results are still very inaccurate. In this study, the angiogenic factors present in gestational and cyclic corpora lutea (CLs) will be analyzed in order to evaluate the differences between these reproductive phases. To achieve this goal, will be used ovarian samples already collected in the project entitled: "Luteal function in the cyclic and gestational canine diestrous: a cellular approach to genetic and metabolic point of view," FAPESP process: 2011 / 17768-3. From the results obtained by the technique of RNA-Seq, the angiogenic factors will be selected for validation. The monitoring of the estrous cycle and ovariohysterectomy of females in cyclical diestrous (n = 20) and pregnancy (n = 20) were performed and the CLs were collected between days 7-11, 21- 25, 40-44 and 61-64 after pre-ovulatory surge of LH in all females. For analysis of gene profile, CLs of the right ovary were subjected to RNA-Seq technique and subsequently with the results obtained among the angiogenic factors and their receptors, were selected VEGFA, FGF, ANGPT, IGF and its binding proteins (IGFBP) for validation by qRT-PCR and immunohistochemistry. Expression validation of selected factors will be assessed as normal and variances, it can be used ANOVA or Kruskal Wallis among the group of pregnant and nonpregnant females. The data will be analyzed using the GraphPad Prism program 5. It will beconsidered the significance level of $p < 0.05$.

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THE FAILURE TO DETECT PIROPLASMS OF SYLVATIC RODENTS FROM BOTUCATU, SÃO PAULO, BRAZIL

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LC Demoner
MRL Silva
NM Magro
LH O'Dwyer

Rodents could be infected by many blood parasites, including piroplasms species that are zoonotic parasites. In Brazil, human infection with *Babesia* spp. was recorded twice, with the description of *Babesia* merozoites in blood smear, without molecular confirmation. Also, there are few reports of piroplasms in rodents from Brazil, in two of them the authors detected parasites only in blood smears, without molecular characterization, and, in the other the authors made only the molecular diagnostic. The aim of this study was to investigate whether piroplasms are present in wild small rodents from forest fragments that surround rural areas in Botucatu County, São Paulo, Brazil. Sixty seven rodents belonging to five species were live-trapped in forest fragments. Blood samples were obtained for blood smear confection and molecular detection. Blood was submitted to DNA extraction and conventional and nested PCR targeting 18S rDNA fragments of piroplasms group. We tried two nested PCR with the primers BTF1/BTR1 and BTF2/BTR2 followed by the primers BT18SF1/BT18SR1 and BT18SF2/BT18SR2. Two additional PCR were tested with the primers BabF/BabR and BAB2 143-167/BAB2 694-667. Ten from the 67 animals had blood structures similar to small piroplasms and eight animals were infected with *Hepatozoon* sp. Despite the suspicion of piroplasm infection, all the PCR and nested PCR exams were negative. The reactions with the primers BabF and BabR, and also with BAB2 143-167 and BAB2 694-667 detected *Hepatozoon* sp., although they were designed to detect piroplasms. The negative molecular results may be related to the amount of DNA presented in the samples that probably was too small to detect, even with molecular methods. Another explanation is the possibility that the parasites observed were not piroplasmas, but haemoplasmas or even Rickettsiae, which could be confounded with small piroplasmas in blood smear examination.

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MORPHOLOGICAL AND MOLECULAR DETECTION OF PIROPLASMS IN *DIDELPHIS ALBIVENTRIS* FROM BOTUCATU, SÃO PAULO, BRAZIL

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F Fornazari
LC Demoner
H Langoni
CR Teixeira
LH O'Dwyer

Didelphis albiventris may act as reservoir of various pathogens that can infect humans and domestic animals. Piroplasms are blood-borne parasites transmitted mainly by tick vectors and little is known about their prevalence in *D. albiventris*. In the present study, we assessed the presence of piroplasms in 67 *D. albiventris* trapped in urban and periurban region of Botucatu, using morphological and molecular methods. Blood samples were collected for DNA isolation and blood smear preparation. Molecular detection was performed using nested PCRs based on amplification of 18S rDNA and sequencing. The microscopic examination of blood smears revealed intraerythrocytic inclusions in 14 (20.89%) *D. albiventris*. The organisms observed were single intraerythrocytic inclusions and showed spherical, oval, pyriform or irregular-shape. In five samples, intraerythrocytic inclusions were large, while four animals showed small piroplasms and in five animals, both piroplasms were presented. These observations suggest the presence of two different piroplasms infecting *D. albiventris*. Eighteen (26.86%) samples were positive in the PCR and ten partial piroplasm 18S rDNA sequences were obtained, of samples that showed large piroplasms in blood smears, with lengths ranging from about 1305–1382 bp. The sequences of the study were identical among them and BLAST search revealed 95% of similarity to *Theileria* sp. from Australian marsupial (JQ682879) and 94% of similarity to *Babesia* sp. from Brazilian seabirds (KC754965). These are the first molecular detection of piroplasm in *Didelphis albiventris*, and the similarity (95%) observed among sequence of piroplasms obtained in the present study and sequences deposited in the Genbank indicate the occurrence of at least a new species of piroplasm. Further studies will be conducted for identify the species that infect *D. albiventris* and understand the phylogeny of these parasites.

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REVIEW OF TOLTRAZURIL PROTOCOLS FOR CONTROLLING COCCIDIOSIS IN PIGLETS IN BRAZIL

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Coccidiosis is one of the most important diseases affecting suckling piglets. *Cystoisospora suis* causes diarrhea, low performance and significant economic losses. Toltrazuril administration is used worldwide for coccidiosis prevention, however, there is no research regarding the correct prophylaxis of this disease in Brazil. Thus, this study aimed to evaluate different protocols of toltrazuril administration (FARMACOX® 5%, oral suspension, Farmabase Animal Health, SP, Brazil). These protocols were used for specific conditions of production in Brazil for controlling coccidiosis under field conditions. For that, 495 piglets were randomly allocated. The animals were divided into four different groups: 125 piglets (control group) which did not receive toltrazuril; 127 piglets that received toltrazuril on the third day of life (group two); 116 piglets which received toltrazuril on the fifth day (group three), and 127 piglets that received two dosages of toltrazuril with 3 and 7 days-old (group four). Excretion of oocysts was evaluated by coproparasitological analyses when the animals were 7, 12, 21 and 63 days-old. They were individually weighed at 7, 21 and 132 days-old to evaluate the growth performance between treatments. Data were analyzed using the MIXED procedure of SAS, using the third day weight as an independent covariate. Results were adjusted by Tukey ($P \leq 0.05$). Groups 2 and 4 did not excrete *C. suis*. One animal from the control group and group three (63 days-old) excreted *C. suis* oocysts. The control group demonstrated lower weight at weaning when compared to group 4. When the piglets from the control group were 132 days-old the body weight was significant lower when compared to groups 2 and 4. The control group also demonstrated significant lower daily weight gain (DWG) then group 4 at weaning and lower significant DWG at 132 days-old then groups 2 and 4. These results suggested that toltrazuril is effective in controlling coccidiosis and improving performance indexes.

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EFFICACY OF BARBERVAX® IN GRAZING LAMBS AND CALVES AGAINST HAEMONCHOSIS*

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GFJ Newlands[2]
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A vaccine containing integral membrane glycoproteins from the intestine of *Haemonchus contortus* was evaluated in young calves and lambs, naturally infected by gastrointestinal nematodes. Vaccinated animals received 5 µg or 50 µg of the antigen and 1 mg of saponin adjuvant, while the controls received adjuvant alone. Lambs received six vaccinations each three weeks apart over the course of the trial. The first vaccination was given when the animals were only four weeks old on average. The rationale was to try to stimulate the response to the vaccine before the lambs became anaemic or sick due to haemonchosis and less likely to respond well to vaccination. All lambs were weaned when approximately 10 weeks old, one week after their third vaccination. Those used for worm counts were euthanised three weeks after their last vaccination. The circulating antibody response of both vaccinated groups (5 µg or 50 µg of the antigen) was very similar and followed the same pattern, rising after each immunisation and then declining. In the case of the lower antigen dose this was associated with significantly less anaemia, 72% reduction in the overall number of *Haemonchus* spp. eggs produced and significantly fewer worms compared with control lambs. Vaccinated calves received initially three doses of vaccine, 3 weeks apart before weaning and then four more times at 6 weeks intervals. Three weeks after the last immunisation all of the calves were euthanised for worm counts. Immunisation stimulated high titre antibodies against the vaccine antigens, reduced the egg output of *Haemonchus* spp. by 85% and the numbers of *Haemonchus placei* and *Haemonchus similis* by 63% and 32%, respectively, compared with control calves. It was concluded that vaccination with intestinal membrane glycoproteins from *H. contortus* could be a useful additional tool for controlling *Haemonchus* spp. infection in ruminants raised in Brazil.

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VACCINATION OF EWES WITH BARBERVAX® DURING PREGNANCY AND LACTATION AND IN THEIR LAMBS*

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Haemonchus contortus is the most important parasite of small ruminants, mainly in the tropics and subtropics. The vaccine Barbervax® was evaluated against *H. contortus* in grazing ewes during pregnancy and lactation and in their lambs. Ewes were divided in two groups: one was supplemented in order to increase the body score condition, while the other had access to a basal nutrition in order to avoid changes in the body score. Half of the animals received three doses of the vaccine every 3 weeks (starting at mid-pregnancy) followed by three more doses at 6 weeks intervals until weaning. The remaining animals of each group were the controls. The lambs were divided in two groups: one received four doses of the vaccine every 3 weeks, from two-months of age, the second were unvaccinated. At three months of age, the lambs were weaned and moved to a feedlot. The body score condition, packed cell volume (PCV) and total plasma protein were higher in the supplemented groups. Due to high faecal egg counts (FEC) associated with low PCV values, several ewes received precautionary anthelmintic treatments. In both supplemented and basal diet ewes fewer treatments were necessary in vaccinates than controls. Within vaccinates, fewer treatments were needed in the supplemented group. Meanwhile, the FEC of vaccinated lambs were more than 80% lower than in controls. In conclusion, the vaccine conferred a significant protection in lambs against haemonchosis. Such protection, however, was less evident in pregnant and lactating ewes.

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CORRELATIONS BETWEEN ACUTE PHASE PROTEINS AND FECAL EGG COUNTS IN CALVES NATURALLY INFECTED BY GASTROINTESTINAL NEMATODES

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This study aimed to evaluate possible correlations in selected serum acute phase proteins (APPs) and fecal egg counts (FEC) in calves naturally infected by gastrointestinal nematodes. We used 51 clinically healthy crossbred calves, two to 24 months old, which belonged to two private farms in São Paulo state, Brazil. FEC were determined using the modified McMaster technique. Blood was collected from the jugular vein. Serum concentrations of total protein (TP), haptoglobin (Hp), paraoxonase-1 (PON-1), and albumin were determined using an automated analyzer (Olympus Diagnostica GmbH). The data are reported as median. The Mann Whitney test was used to compare the groups as data was not normally distributed. The correlations were assessed by Spearman's correlations. Statistical significance was set at $P < 0.05$ for all analyses. The calves were divided into two groups according to the results of eggs per gram of feces (EPG) in group A (GA) (n=28): EPG ≤ 200 and group B (GB) (n=23): EPG > 200. The EPG ranged between 50 to 200, and 700 to 6750 in group A and B, respectively. Group B had significant higher concentrations of Hp (0.4 g/L) when compared to GA (0.2 g/L). No significant differences were observed for the others analytes (GA; GB): TP (6.8; 6.9 g/dL), PON-1 (7.3; 6.2 UL/mL), and albumin (2.8; 2.9 g/dL). There was a moderate significant negative correlation between GB and PON-1 (r = - 0.4). No significant correlations were observed for TP (r = - 0.2), Hp (r = - 0.1), and albumin (r = - 0.1) in this group. No significant correlations were observed in GA for TP (r = - 0.3), Hp (r = 0.3), PON-1 (r = 0.1), and albumin (r = 0.1). These results could indicate that the parasite burden produced an inflammatory reaction as Hp and PON-1 are positive and negative APPs, respectively.

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CHANGES IN BIOCHEMICAL ANALYTES IN CALVES INFECTED BY NEMATODE PARASITES IN FIELD CONDITIONS

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The aim of this study was to evaluate possible alterations in selected serum biochemical analytes in calves sub-clinically infected with gastrointestinal (GI) and pulmonary nematodes in field conditions. We used 86 clinically healthy crossbred calves, two to 24 months old, which belonged to two private farms in São Paulo state, Brazil. Samples of feces and blood were collected. Fecal egg counts (FEC) were determined using the modified McMaster technique with a sensitivity of 50 eggs per gram of feces (EPG). First stage-larvae of *Dictyocaulus viviparus* were identified by a modified Baermann method. The biochemical profile was measured using an automated analyzer (Olympus Diagnostica GmbH) and the analytes determined were: haptoglobin, paraoxonase type 1, acetylcholinesterase, butyrylcholinesterase, triglycerides, cholesterol, HDL, LDL, iron, UIBC, total protein, albumin, amylase, lipase, phosphorus and calcium. The calves were divided into four groups according to the results of EPG and the modified Baermann method. Group 1: healthy control animals (n=16); Group 2: calves with only GI parasites (n=51): This group was sub-divided into sub-groups according to the EPG threshold: 2a - GI parasites with low EPG (n=28) (≤ 200), and 2b - GI parasites with high EPG (n=23) (≥ 200). Group 3: animals with only lungworms (n=5) and Group 4: calves with lung + GI parasites (n=14). The Kruskal-Wallis test was used to compare the groups and Dunn's post-test was used for multiple comparisons as the data was not normally distributed ($P < 0.05$). Haptoglobin concentration significantly increased in calves with GI and pulmonary parasites. A significant increase in acetylcholinesterase activity was observed in calves infected with lungworms. The lipid profile was decreased but lipase activity increased in calves with GI parasites. These findings in calves sub-clinically infected could provide an indication of GI parasites and lungworm infection, especially in an endemic area.

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DETECTION OF PERIODONTOPATHOGENS IN MICROFLORA OF BOVINE PERIODONTITIS

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Bovine periodontitis is a progressive purulent infectious process associated with the presence of strictly and facultative anaerobic subgingival biofilm and epidemiologically related to soil management in large geographic areas of Brazil. By using PCR, an independent method of cultivation, this study sought to potential periodontal bacterial pathogens in periodontal pockets of cattle (n=26) with chronic lesions deeper than 5mm on probing and gingival sulcus of animals (n=25) considered periodontally healthy. In the qualitative analysis, we employed primers of 35 bacteria known to be present in the subgingival microbiota of animals and man. Prevalence and risk analysis were performed using Student´s T test and Spearman Correlation Test. The bacterial prevalence in the bovine periodontal pocket samples were as follows: *Fusobacterium nucleatum* (96.2%), *Actinomyces naeslundi* (80.7%), *Fusobacterium necrophorum* (80.7%), *Porphyromonas endodontalis* (80.7%), *Prevotella melalinogenica* (73.1%), *Treponema amylovorum* (73.1%), *Prevotella intermedia* (61.5%), *Tannerella forsythia* (61.5%), *Treponema pectinovorum* (61.5%), *Eikenella corrodens* (53.8%), *Porphyromonas asaccharolytica* (53.8%) and *Prevotella oralis* (50%). In bovine without lesions prevailed *F. nucleatum* (84%), *Eikenella corrodens* (72%), *F. necrophorum* (68%), *P. endodontalis* (40%), *P. loeschei* (40%) and *T. forsythia* (40%). Data evaluation by T test, enabled to verify that ocorrence of *Actinomyces naeslundii* (p=0.0002), *Enterococcus faecium* (p=0.002), *Porphyromonas asaccharolytica* (p=0.000003), *P. endodontal*is (p=0.0023), *Prevotella buccae* (p=0.0017), *P. intermedia* (p=0.0020), *P. melaninogenica* (p=0.00006), P. *oralis* (p=0.0028), *Treponema denticola* (p=0.0042) and *T. pectinovorum* (p=0.0000) is correlated with bovine periodontitis. The same results were obtained by Spearman correlation test. In addition to cataloguing the qualitative composition of the subgingival microbiota of cattle with chronic periodontal lesions and in animals considered periodontally healthy, these findings will be critical to filling in the Socransky postulate, and therefore necessary for studies of etiopathogenesis, treatment and control of bovine periodontitis.

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PRESENCE OF *PORPHYROMONAS* AND *PREVOTELLA* SPECIES IN THE ORAL MICROFLORA OF SHEEP WITH PERIODONTITIS

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In many countries sheep periodontitis is considered one of the main causes responsible for premature disposal of animals in herds because the disease causes premature loosening and loss of teeth in its natural course. With its own epidemiological characteristics and multifactorial etiology, its subgingival microbiota is compatible with that found in human, bovine periodontitis and in other animal species. Among the putative periodontal pathogens there are species that produce black pigment belonging to *Porphyromonas* and *Prevotella* genera. In conjunction with other periodontal pathogens induce, in dysbiosis, an inflammatory response resulting in destruction of periodontal tissues. The aim of this study was to detect directly by polymerase chain reaction the presence of potential periodontopathogens species of *Porphyromonas* and *Prevotella* genera in the periodontal pocket of sheep periodontitis lesions (n=14) and healthy periodontal sites (n=20). Prevalence and risk analysis were performed using Student´s T test and Spearman Correlation Test. Among the *Porphyromonas* and *Prevotella* species detected in samples of sheep periodontitis, *P. melalinogenica* (85.7%), *P. buccae* (64.3%), *P. gingivalis* (50%) and *P. endodontalis* (50%) were the most prevalent. In gingival sulcus of sheep considered periodontally healthy prevailed *P. gingivalis* (15%) and *P. oralis* (10%). *Porphyromonas gulae* and *Prevotella tannerae* were not detected in the 34 samples studied. Data evaluation by T test, enabled to verify that ocorrence of *P. asaccharolytica* (p=0.0006), *P. endodontalis* (p=0.0015), *P. gingivalis* (0.0274), *P. buccae* (0.00004), *P. intermedia* (0.0303), *P. melalinogenica* (0.00000) and *P. nigrescens* (0.0006) is associated with sheep periodontitis. The same results were obtained by Spearman correlation test. The identification of these species of *Prevotella* and *Porphyromonas* genera in the periodontal pocket of sheep is an original and important contribution to studies of the pathogenesis and control measures in sheep periodontitis.

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ISOLATION, CULTURE, AND CHARACTERIZATION OF MESENCHYMAL STEM CELLS FROM BOVINE ADIPOSE TISSUE

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The aim of this study was to isolate, process, cultivate and characterize the mesenchymal stem cells from bovine adipose tissue (MSCs-AT). For that, we used four females *Bos taurus indicus* with body condition score 3-5. After local anesthetic blockade, 4 cm below the right ischial tuberosity, adipose tissue was obtained. Samples were digested in HBSS containing 0.4% collagenase and 0.5% fetal bovine serum (FBS). The cells were then cultured at 37.5°C in 95% air and 5% CO2 using the maintenance medium (DMEM high glucose/F12 (1:1) supplemented with 20% FBS, 1% penicillin/streptomycin, 1% amphotericin B and 0.1% amikacin. Immunophenotypic analysis of MSCs was done using a flow cytometer (FC) and antibodies mouse anti-bovine CD44: FITC (clone IL-A118), rabbit anti-CD 34: FITC (polyclonal, Biorbyt®, USA), mouse anti–CD 29: AF (clone TS2/16, BioLegend®, USA), mouse anti-horse MHCII: FITC (clone CVS20, AbD Serotec®, UK) and mouse anti-Vimentin (clone v9). The results are presented as mean and mean standard error. Additionally, the differentiation assay for mesodermal lineage was performed using commercial medium (STEMPRO®, Thermo Cientific). The response was evaluated using alizarin red 2% (osteogenic assay), Oil red O 0.5% (adipogenoc assay), Alcian blue and toluidine blue (chondrogenic assay) dyes. The MSCs-AT presented adherence to plastic and fibroblastoid morphology. At FC, cells presented high expression for CD29 (100%±0), CD44 (95.47%±2.18), vimentin (95.67%±2.5), and negative/low expression of MHC-II (6.42%±2.43) and CD34 (11.12%±3.05). Besides, osteogenic differentiation was confirmed by the deposition of calcium-containing matrix, the adipogenic differentiation was demonstrated by the presence of intracytoplasmic lipid droplets and the chondrogenic differentiation by the presence of proteoglycans. The adipose tissue hosts population of MSCs with appropriate immunophenotypic profile and potential for differentiation, desirable in regenerative medicine.

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DOPPLER ULTRASOUND MAMMARY ARTERY EVALUATION ON CROSSBRED MURRAH BUFFALOES DURING THE GROWTH PHASE

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The objective of this study was to evaluate the hemodynamic indices of the mammary artery and its relationship with buffalo calves body weight. We used six crossbred Murrah females, clinically healthy, with initial age of zero months and reared extensively. Ultrasound examinations were performed (Doppler) of the cranial and caudal mammary arteries, with an interval of 28 days each, for a year, to determine the resistivity index (RI) and pulsatility index (PI) and internal vessel diameter (ID). Furthermore, weighing was performed and body weight (BW) was given in kilograms (kg). We used analysis of variance with repeated measures over the months, Spearman correlation coefficient and non-linear multiple regression considering a 0.05 significance level. There was a decrease of RI and PI and increased ID from the first to the last month of the study (P<0.0001), however, there was no statistical difference in hemodynamic parameters between the cranial and caudal mammary arteries (P>0.05). There was correlation between IR and IP (r= 0.94; P<0.0001), and the ID (r= -0.98; P<0.0001), and IP to the ID (r= -0 98; P<0.0001). Linear effect of BW on IR (R2= 0.97; P= 0.0054) and on ID (R2= 0.97; P= 0.0069) was detected. The results are related to the reduction of vascular resistance, the continuous increase in blood perfusion of arterial lumen and the metabolic activity resulting from the increased blood supply and the nutrient demand in response to mammary development. Similarly, the variation of BW was matched to increased blood perfusion and arterial caliber, being, therefore, adaptive mechanisms that probably contributed to the maintenance of the gland dynamic state. Thus, the variation of the hemodynamic indices of mammary arteries besides reflecting body growth can be considered predictive of mammary development progression incrossbred Murrah buffaloes during the first year of life.

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EVALUATION OF LACTATE CONCENTRATION, BODY WEIGHT AND MEAT TRAITS IN NELLORE CATTLE SUBMITTED TO HIGH-GRAIN RATION

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The aim of this study was to determine blood lactate concentration during the feedlot (FE) period and after slaughter and to check its influence on body weight and meat quality traits in Nellore cattle finished at feedlot. Ninety uncastrated males were submitted to the feedlot. Animals had average weight of 390 ± 37 kg and about 24 months of age. Lactate measurements was measured by spectrophotometry on the day 1 (L1), day 14 (L14), day 27 (L27), day 69 (L69) and day 96 (L96). Muscle meat samples were collected from *Longissimus thoracis* and laboratory analysis of lightness (L*), redness color intensity (a*) and yellowness (b*), shear force (SF) and pH of meat samples without aged (_0) and aged for seven days (_7) were performed. The lactate variables were analyzed by least squares with the use of PROC MIXED of SAS, considering a model with effects of pen and day of blood sample collection. Analysis of body weight at day 69 (W69) and meat quality traits were performed with a model including pen and lactate effects. Phenotypic correlations (r^2) were performed. Lactate remained stable during the FE period with means of 4.83±0.30 mMol/L, 5.68±0.29 mMol/L, 5.03±0.29 mMol/L and 4.15±0.31 mMol/L for L1, L14, L21 and L69, respectively. Higher mean was obtained for L96 (10.77±0.27 mMol/L) compared with the previous four times ($P<0.001$). No significant association for W69 and L69 was found. For meat quality traits, the lactate effect was significant for a* and b* ($P<0.10$) just for _0. The r^2 estimated for lactate and a* of _0 was negative, $\rho = -0.19$ ($P<0.10$). For lactate, r^2 was statistically significant just for measures performed during the FE period ($P<0.05$). The results of this study showed that the lactate measured during the FE period was stable and had no relation to the measurement made after slaughter and also the lactate at post slaughter can influence a* and b* in _0. It is important to avoid stress factors at this stage to ensure better color of the meat product.

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IMPUTATION GENOTYPING OF LOW-DENSITY FOR HIGH DENSITY BY THE PROGRAM FIMPUTE IN BREED GYR

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The main objective of imputation is genotyping individual's references with high density marker panel (HD), and candidate selection with low density panels (LD), which have lower cost, and then predict the genotypes of locos missing with information from the reference population to standardize the genome. The programs most used in inferring genotypes in animal production and have good accuracy imputation are FIMPUTE and BEAGLE, which are based on population DL, family information. For imputation of genotypes was held with a population of Gir, 18 were genotyped animals with Bovine HD containing 777,962 markers, and 155 animals with Bovine LD contains 33,000 markers, as well as pedigree information: Nº individuals in pedigree: 4052, Nº bulls: 236, Nº individuals with offspring: 1266, Nº individuals without descent: 278, considering the autosomes, for the allocation of commercial LD panels for the HD chip. We used the FIMPUTE program because it is efficient in exploring long haplotypes, usually close relatives. Imputation was simulated assuming that the validation set of animal genotypes were available for markers that were present in HD. The results of the imputation were: Nº polymorphisms Single Nucleotide Base (SNPs) was 734,848, errors Nº SNPs: 1353, Mendelian errors: 0.0032% (4421 SNPs), heterozygous loci: 3.1211%, Nº individual genotypes: 173 animals. The allocation has increased the number of animals for genomic analysis, increased SNPs to HD, with small errors, but to have the reliability of imputation should be made genotypes blocks analysis to determine the percentage of accuracy, a process that is still in analysis. Concluding that the imputation is a great option to reduce the cost of genotyping, in addition to being a useful tool to enlarge the genome information to make genomic selection (GS) and Analyses of genomic association (GWAS).

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GENE EXPRESSION OF HEAT SHOCK PROTEIN AND MEAT QUALITY OF NELORE CATTLE

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The study of genes of the heat shock proteins (HSPs) has an important role in the meat science. The involvement of HSP27 in the tenderness of the beef is still considered highly controversial. The aim of this study was to evaluate the gene expression of the heat shock protein (HSP27) with different tenderness traits in Nellore cattle. Ninety Nellore animals were used. The animals were pasture-fed until approximately 24 months of age, when they were moved to feedlot. The feedlot period was 95 days with average body weight of approximately 550 kg. The animals were weighed at the beginning of the feedlot period and every 28 days until slaughter. Immediately after slaughters, samples for gene expression analysis were collected. After slaughter, samples of the *Longissimus thoracis* muscle between 12 - 13th ribs were designed the tenderness analysis through shear force (SF). Two extreme groups were separated by means shear force (SF): (1) animals with tender meat (n = 15 animals) and (2) animals with tough meat (n = 15 animals). Total RNA was extracted from skeletal muscle samples and was used for quantitative real-time PCR (qRT-PCR) reactions by Taqman probes. As reference genes were used the glyceraldehydes 3-phosphate dehydrogenase (GAPDH), TATA box binding protein (TBP) and Beta Actin (ACTB) and as target genes of HSP27. The statistical linear mixed models for the qRT-PCR data were used, as proposed by literature. The statistical linear mixed models for the qRT-PCR data were used, as proposed by literature. The animal group named with tender and tough meat presented average of 3.9 ± 0.9 kg and 7.9 ± 1.3 kg, respectively. Expression of HSP27 (HSPB1) gene did not differ significantly (P> 0.05) between the two groups. Therefore, it can be inferred that the gene expression of the tender meat group approaches of the tough meat group, that is, occurs the same gene expression of the HSP27 in both groups of Nelore cattle.

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BIOMARKER IDENTIFICATION IN BLOOD *BUBALUS BUBALIS* BUFFALO BY SHOTGUN PROTEOMICS*

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Significant scientific breakthroughs have occurred in recent years involving basic experimental research, which has affected the biotechnology sector, regarding the development of new bioactive compounds using animal bioisumes. Accordingly, *Bubalus bubalis* have gained considerably ground in the world economy as biological material and devices, as the heterologous fibrin sealant from CEVAP. This sealant is composed by a serine protease from *Crotalus durissus terrificus* snake venom and a fibrinogen-rich cryoprecipitate from blood plasma of domestic buffaloes and it has been used as a scaffold for stem cells, biological glue in surgical procedures and as an aid for treating chronic venous ulcers in humans. This study aimed to characterize the differentially expressed proteins in the serum of buffalos affected by brucella through proteomic approach in order to highlight candidates for molecular biomarkers. The development of diagnostic tests and/or clinical prognostic more effective and with greater specificity for serum-buffalos can away the incidence of false negative and false positive tests.The serum of forty buffaloes were analyzed, in which 20 control buffalos and 20 brucella-infected buffaloes. All samples were subjected to a sequential depletion protocol involving two protein precipitation steps: depletion of proteins will be performed in the presence of DTT and then subjected to a second stage of decomplexation with acetonitrile. A tryptic digestion protocol was realized and the samples were analyzed by LC-MS/MS using MicroQ-ToF III mass spectrometry (Bruker Daltonics) coupled to LC-20AT chromatograph liquid (Shimadzu). Mascot software (Matrix Sciences) was used to identify the differentially expressed proteins and a statistical routine were standardized.As results, five potential biomarkers of brucella in water buffalo were evidenced in this study, in which are alpha-1B-glycoprotein precursor protein, inter-alpha-trypsin inhibitor, beta-tubulin and inter-alpha-trypsin inhibitor. These proteins participate in the inflammatory response pathway.This protein description can provide a better understanding of brucellosis mechanisms in water buffaloes. In the future, this study will support the developing of a specific diagnostic platform for brucella diasease in *Bubalus bubalis*.

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SERUM AMYLOID A (SAA) AND HAPTOGLOBIN (HP) CONCENTRATIONS OF HEALTHY HORSES SUBJECTED TO EXPERIMENTAL SMALL COLON ENTEROTOMY AND TREATED WITH SODIC HEPARIN

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The acute phase response (APP) is part of a defense mechanism to injury and is correlated with the severity of the underlying condition or intensity of surgical trauma. Heparin is routinely used in horses to avoid abdominal adhesions; however no information is available about its effect on inflammation. This study aimed to evaluate the inflammatory response in horses subjected to small colon enterotomy and treated with systemic heparin. Ten adult horses were subjected to surgery and divided into 2 groups: the control group (CG) and treated group (TG). TG immediately after surgery and at every 12 hours for 5 days, received heparin at a dose of 150 IU/kg subcutaneously. The animals underwent WBC count, determination of SAA and Hp; peritoneal fluid evaluation prior to enterotomy; 12 hours; 24h; 48h; 72h; 6; 10, and 14 days after enterotomy. No differences were observed between groups or time points for WBC count or peritoneal fluid features. SAA levels were undetectable before and 12 h after surgery, but started to increase 24 h after the surgical procedure, reaching a peak at 48 h for both groups. Serum Hp markedly increased at 24 h and a peak was reached at 48 h for the control group, and at 72 h for the treated group. No significant difference was observed between groups for serum Hp or SAA concentrations; however the CG showed significantly higher Hp concentration at 48 h (2.13g/L) when compared to before surgery (0.978g/L). The TG demonstrated significant different concentrations for SAA at this same time point. The absence of differences between 48 and 72 h after surgery for s Hp concentrations suggests that heparin influences Hp values. SAA is a more sensitive APP than Hp for evaluating inflammation in horses; however when evaluating the role of heparin in these animals Hp showed higher sensibility. Possibly this response is associated with the biological functions of Hp but the pathway that results in this action is not clear.

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SERUM PROTEIN PROFILE AND GGT ACTIVITY OF PIGLETS BEFORE AND AFTER COLOSTRUM INTAKE

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Onida, PT[1]

The aim of this study was to evaluate the serum protein profile and gama glutamyltransferase (GGT) activity of 40 Penarlan piglets before and after colostrum intake. Blood samples were taken at two different time points: until six hours after birth and 48h after colostrum intake. Protein fractions in serum were determined by means of sodium dodecyl sulphate-polyacrylamide (SDS-PAGE) gel electrophoresis, total serum protein concentration by the Biuret method, and GGT activity was determined with commercial kits (Intertek – Katal) in an automatized spectrophotometer (Cobas Mira Plus; Roche Diagnostic Systems). As the data did not meet the normal distribution criteria, the nonparametric Wilcoxon-signed rank test ($P<0.05$) was performed to compare quantitative variables between before and after colostrum intake. Total serum protein concentration was significantly ($P<0.0001$) increased after colostrum ingestion, and the concentrations of IgG heavy and light chains were also significantly increased ($P<0.05$) at the same time point. Gama glutamyltransferase serum activity was significantly decreased ($P=0.0012$) after colostrum ingestion. The ceruloplasmin fraction was evident before and after colostrum intake and a 23kDa protein was observed in all piglets and was significantly increased ($P<0.05$) 48h after colostrum intake. Although in general, GGT activity could present wide inter-individual variations, apparently, the sow´s colostrum has limited activity of this enzyme when compared to ruminants species.

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ASSOCIATION BETWEEN BODY MEASUREMENTS AND EQUINE REPETITIVE ELEMENT 1 (ERE1) IN BRAZILIAN CUTTING LINE QUARTER HORSES BREED

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CM Marchiori[1]
LC Trevisan[1]
LEC Santos[1]
AMT Ospina[1]
BPM Silva[1]
RA Curi[1]

Within the Cutting line Quarter Horses, the animals are usually destined to the functional competition, exploring skills as agility and obedience, traits that are considered of the great importance in the management of cattle in a field. The selection directed to different goals (conformation, racing or cutting lines) has also promoted significant changes in the body traits of the Quarter Horses. Cutting line horses are more compact and have advantaged rear muscles when compared to the racing lines. The aim of this study was to analyze an association between Equine Repetitive Element 1 (ERE1) and Body Measurements in Cutting line Quarter Horses. Were collected blood samples and body measurements of height at withers (HW), body length (BL) and heart girth (HG) of 69 units of Cutting Line Quarter Horses. The analysis was performed through the association between the genotypes of the ERE1 marker obtained by PCR and the estimate breeding values of Body Measurements using the blupf90 and Plink v.1.07 softwares. The HG has high correlation with body weight and was suggestively associated with ERE1 (p-value = 0.08647), HW (p-value = 0.1947) and BL (p = 0.4602) were not significant. The SINE ERE1 appears in a promoter region of the gene MSTN that regulates negatively the muscle growth, the Insert of this element inhibits the expression of MSTN taking to the greater muscle development. A major sample could confirm the suggestive result found here. This study may help the understanding of the relations between the performance and the morphology of the animals within their breed.

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ANALYSIS OF THE EQUINE REPETITIVE ELEMENT 1 (ERE1) ON RACING AND CUTTING LINES OF QUARTER HORSES

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With different objectives of selection, Quarter Horses have lines or segments with different skills or abilities, namely the racing, cutting and conformation lines (ABQM, 2013). Studies on the effect of a short interspersed element (SINE) called Equine Repetitive Element 1 (ERE1) in the promoter region of the myostatin gene (MSTN) showed that the insert of it causes a reduction in gene expression and thus increasing muscle mass in horses. The aim of this study was to analyze the occurrence of Equine Repetitive Element 1 (ERE1) of MSTN gene in racing and cutting lines of Quarter Horses breed. Blood samples were collected from 450 registered equines Quarter Horses of sexes, 69 of cutting and 381 of racing horses. To analyze the ERE1 insertion polymorphism at the myostatin promoter, we amplified genomic DNAs using primers from the sequences flanking the insertion site by simple PCR method. The analysis of frequencies and association were performed by using the R software from the package genetics. The ERE1 was polymorphic in the cutting line with allelic frequency of 0.9 and 0.1 of the allele ERE+ and ERE- and genotypic 0.8 and 0.2 of ERE+/ERE+ and ERE+/ ERE- respectively. Only one animal of racing strain showed heterozygous genotype (ERE+/ ERE-), in this case ERE1 is practically fixed in the racing-line. The difference between allelic frequency ERE1 was significant by Fisher's test (p-value = 1.567e-11; OD = 0.010, CI = 0.0002 – 0.0716). The ERE1 set in the racing line can be explained by a possible indirect selection for strong animals, once they dispute short and high-speed races.

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COMPARED IDENTIFICATION OF MAMMALS OF WILD BRAZILIAN FAUNA THROUGH HAIR*

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The wild animal's trafficking is classified as the third biggest in the world. Veterinary forensic medicine appeared with the goal of conciliate the knowledge with the justice, supporting at environmental issues. The taxonomic identification of wildlife is a routine process of forensics expert. The aim was to identify animals of Brazilian wildlife through microscopic analysis of bristle. Samples of hair of 17 species were analyzed. After being washed with ethyl alcohol¬, one layer of incolor nail polish was applied on a glass microscope slide, the hair was placed on top of the nail polish, and other glass slide was pressed over the hair. In the case of marrow samples, the hair remained in a clarifying mixture, composed of hydrogen peroxide and bleaching powder. The glass slides were analyzed by optical microscopy, using optical and photographs were taken with the software "AxioVision". With the program "Image J", the following measurements were obtained: marrow diameter, overall diameter, ratio marrow/cuticle, ratio diameter overall of marrow/cuticle and amount of scales in 100μm and descriptive statistical analysis was made by "Graph Pad Prism". The photographed images used for measurement were clear and the measures were easily made, it's possible to notice distinction between the hairs, and based on reference materials, it was noted similarity to the assessments made. At descriptive statistical analysis, the size of hair between the species had a mean of 2,99±1,47mm and a non-parametrical distribution, the comparative test between the measures had a statistical significance ($P<0.05$) to the overall diameter and the ratio marrow/cuticle. However, there's a difference between these measurements in the hair of different species. It's plausible the realization of more studies for certificate values in other species and bigger amount of individuals.

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GENETIC CHARACTERIZATION OF SPECIMENS FROM GENUS *ALOUATTA*

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The Alouattas, known as howler monkeys, are distributed from Mexico to southern Argentina, occupying different habitats. In Brazil, occur in southern Bahia to Rio Grande do Sul. They can be easily recognized, because of their anatomy, prehensile tail, and longer hair on the face and quite distinctive roar. The species configures the list of IBAMA as "endangered and vulnerable" being, deforestation and consequently the destruction of their habitats and isolation of the population into small fragments, the main causes of this picture. The use of tools in the field of molecular biology has assisted in data which can be used in species conservation programs. In order to genetically characterize a population of nine animals from different localities of the State of São Paulo, but currently kept in captivity, this research analyzed DNA samples obtained from blood using microsatellite markers. Five pairs of primers were tested where the respective annealing temperatures were determined in the temperature gradient thermocycler program. After standardization of the reaction, for each primer was conducted a new reaction for incorporation of fluorescence and the products were genotyped. The results analyzed using the GeneAlEx program. Four of them were satisfactory for analysis (Ab04, AB10, Ab13 and Ab19). Genotyping products identified heterozygous and homozygous animals for each of the loci studied identifying 19 different alleles. From the analysis of chi-square, the observed heterozygosity was different from the expected indicating that the gene frequencies are changing over the generations. The positive results of amplifications enable the use of these primers in further studies as individual identification and parentage analysis, this data can contribute in further implementations of species management and conservation programs.

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CHARACTERIZATION OF FORMATION AND MAINTENANCE OF PAIRS OF PARROTS (*AMAZONA AESTIVA*) KEPT IN CAPTIVITY

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SA Castro[1]

Allogrooming behavior is a social interaction that has hygiene function and guarantees the establishment and maintenance of affective bonds between the group members. In the nature parrots are monogamous that remain paired up the end of life. In captivity, despites the formation of couples, it is also described isossexual pair formation[1]. The objective of this study was monitoring the time duration and frequency of *allogrooming* and *autogrooming* behavior, and evaluate if the relation in the social pair is symmetric or not. Forty-one adult parrots (23 males, 18 females) kept at a collective aviary were individually monitored for 40 days. Based on social behaviors (affiliative and agonistics interactions) it was identified 7 social pairs [3 isosexual female, 2 isosexual male and 2 heterosexual]. The pairs were monitored during 2 months, once a week (from 8 to 11a.m. - 90 min/pair) and the social interactions were recorded. The behaviors data were expressed in duration (seconds) and frequency. During the *autogrooming*, the parrots repeatedly nibbled its own skin or preen its own feathers with its beak slightly open and with the tongue in the body regions that they could reach. During the *allogrooming*, the partner made grooming on the unreachable body area like the cephalic and cervical regions. There was a positive correlation between the total duration and frequency of the *allogrooming* behavior ($r = 0.8$; $p < 0.001$). The strongest social pair invested 33% of time in *allogrooming* while the weakest only 3%. Within the pair, the time invested in *allogrooming* was asymmetric, and a member of the pair invested more time doing than receiving the *allogrooming* ($p<0.02$), despite the par being iso or heterosexual. No pair displayed copulation attempts. Our results confirm that blue fronted parrots also establish isossexual pairs. This find suggests that these birds could establish stable friendship relation links and opens perspectives to elaborate and test hypothesis for the biological meaning.[1]Queiroz, C. Master Dissertation, 2015.

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EFFECT OF FOOD ENRICHMENT ON THE BEHAVIOR AND WELFARE OF PARROTS (*AMAZONA AESTIVA*) KEPT IN CAPTIVITY – PRELIMINARY RESULTS

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Parrots kept in captivity often exhibit behavioral disorders like stereotypies and feather plucking. To deal with these problems and improve animal welfare, several environmental enrichment protocols are indicated, including food enrichment procedures. The objective of this study was evaluate the adaptive capacity of long lasting captive parrots (*Amazona aestiva*), used to eat processed fruits to the consumption of fruits in their natural state and the effect of this food enrichment on the bird's behavior. Forty-one parrots, 23 males and 18 female, weighing 444±37.7g and 391±21.3g respectively, kept in a large collective aviary, had their body condition score (1-3 points - BCS) recorded, according to the pectoral muscle condition, and were monitored during 41 days for to the presence of stereotypies. After identification of the birds possessed stereotypic behavior, the feeding management was changed offering the birds fruits in their natural state [papaya (*Carica papaya*); orange (*Citrus sp*); yellow cavendish banana (*Musa sp*); palm-jerivá (*Syagrus romanzoffiana*); chinaberry (*Melia azedarach*) and leucaena (*Leucaena leucocephala*)]. It was defined as acceptance criteria the consumption of at least 50%. The mean BCS was 2.2 and the stereotypies were detected in 12% (5/41) of the birds; the most observed abnormal behaviors were (a) remain hanging by the open legs in aviary corners, (b) remain hanging on the ceiling by the legs with the head contact with the ceiling, and (c) to move the head from right to left repeatedly in sinuous movements. Banana and chinaberry were the most accepted fruits (89% and 82% of consumption, respectively). Data are now being analyzed to evaluate the effect of food enrichment on the bird's behaviors. Based in the preliminary results the birds have adapted promptly to the fruits in their natural state, showing behavioral flexibility to obtain food.

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