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## Fronteiras da Ciência: Explorando novos horizontes

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Title	Effects of urbanization and pollution on the heterophil/lymphocyte ratio of canaries (Serinus canaria)
Authors	Paulo Vitor Alves Ribeiro <sup>1</sup> Henrique Tsubota Manrique <sup>2</sup> Vanessa Fonseca Gonçalves <sup>3</sup> Camilla Queiroz Baesse <sup>2</sup> Vitor Carneiro Magalhães Tolentino <sup>2</sup> Celine de Melo <sup>2</sup>
Affiliations	<sup>1</sup> Instituto de Ciências Biomédicas, UFU, Uberlândia, Brazil <sup>2</sup> Instituto de Biologia, UFU, Uberlândia, Brazil. <sup>3</sup> Escola de Educação Básica (ESEBA/UFU), Uberlândia, Brazil.
Session	Biologia Ambiental, Evolução e Biologia Comparada

#### Abstract and Keywords

Stress-inducing agents can lead to morphophysiological and behavioral changes in various animals, with birds serving as excellent study models for environmental biomonitoring. The heterophile/lymphocyte ratio (H/L) is an efficient indicator of chronic stress in birds, as stress hormones typically elevate the number of heterophils while decreasing the number of lymphocytes in the blood, resulting in an increased H/L ratio. The objective of this study was to compare the H/L ratio over a 120-day period in two groups of canaries (Serinus canaria) housed in cages: one in a residence in the center of Uberlândia (urban group) and the other in a rural property far from urbanization (rural group). In both locations, we measured levels of atmospheric pollutants—carbon monoxide (CO) and particulate matter (PM)—using a manual pollutant meter. Twenty canaries were maintained in each group, and blood samples were collected every 15 days for subsequent analysis under an optical microscope. The methods performed were authorized by CEUA/UFU. Air pollution (CO and PM) and indicators of chronic stress (H/L ratio) were significantly higher in the urban environment, with the H/L ratio of urban canaries increasing with exposure time. These results suggest that canaries exposed to urbanization exhibit higher indicatives of chronic stress (H/L ratio), likely attributable to the high levels of atmospheric pollutants generated by vehicular traffic (carbon monoxide - CO and particulate matter – PM) in the urban environment. Given that these pollutants are considered harmful, the effects observed in canaries serve as a warning for both environmental and human health.

**Key-words:** Biomonitoring, stress, carbon monoxide, particulate matter.



Title	Collection and conservation of Tardigrades
Authors	Alexsandre Domingues Junior Ricardo Corbetta
Affiliations	University of Vale do Itajaí, Itajaí, Brazil
Session	Environmental Biology, Evolution and Comparative Biology

#### Abstract and Keyword s

The Phylum Tardigrada, a sister group to Arthropoda, encompasses a range of cosmopolitan micro invertebrates from fresh and salt water. There are currently more than 1400 described species, which can be distinguished into two classes, Heterotardigrada and Eutardigrada. According to Barros (2020), around 100 species occur in Brazil, 30 of them in marine environments and 70 in terrestrial and aquatic environments. Most occurrences are in the southeast and northeast of the country. In Santa Catarina the group is mentioned only once.

Specimens have been collected since August 2023 in the cities of Balneário Piçarras and Itajaí (SC), with an additional collection carried out in Blumenau. For this, a staple remover was used as a spatula to scrape the substrate with bryophytes (moss). Regarding sample storage, paper envelopes were used to store the mosses with appropriate notes about the place and date of collection, taking care not to mix different sampling points, even if they were close together, in order not to associate different species by mistake. To fix individuals on slides, they were transferred to an optical microscopy slide with a small amount of water, using an Irwin loop handcrafted with threads and fixed with Vitral Varnish. The specimens were photographed using a cell phone camera with an adapter for the Microscope eyepiece. The collection already has 9 sheets of prepared material. The observed species corresponds to the genus Echiniscus. Photos of all fixed specimens were obtained using optical microscopy at 400x magnification, where in some specimens different focus levels were photographed to later superimpose the images and obtain a single, better quality photo (focus stacking).

**Keywords:** Irwin Loop, Collection, Tardigradology, Focus Stacking



Title	Ecological corridors
Authors	Alisson Júnio Parreira Peixoto Danusa Radi Gomes Santiago Márcio José do Prado Filho Nathalia Vieira Kamimura
Affiliations	Sistema Gabarito
Session	Education

## Abstract

#### and Keywords

This work highlights the importance of ecological corridors in conservation units, exploring their role in plant connectivity through mycorrhizae, as seen in the movie "Avatar". It begins with an introduction that justifies the relevance of conservation units and the need to investigate ecological corridors. The development addresses competition among plants for resources, visual and chemical communication, mycorrhizae, and electrical signal transmission. Objectives include analysing the function of ecological corridors in biodiversity and investigating the relationship between plants and mycorrhizae. The discussion emphasises the importance of ecological corridors for biodiversity and sustainability. It concludes by reiterating the need to preserve and expand these corridors to ensure a sustainable future.

**Keywords:** ecological corridors, conservation units, connectivity, plants, mycorrhizae, movie "Avatar", competition, resources, visual communication, chemical communication, electrical signal transmission, biodiversity, sustainability.



Title	Composting of organic waste from different origins in the town called Araguari, Minas Gerais: Chemical evaluation
Authors	HR Barcelos <sup>1</sup> , IM Bicalho <sup>2</sup> , MRM Carrijo <sup>1</sup>
Affiliations	<sup>1</sup> Instituto Master de Ensino Presidente Antônio Carlos - IMEPAC/Araguari, Agronomia, Brasil; <sup>2</sup> Universidade Federal de Uberlândia, Uberlândia, Dpt Ciências Agrárias, Brasil.
Session	Environmental biology, Evolution and Comparative biology

# Abstract and Keywords

Interest in the sustainable management of organic waste contributes to environmental practices, and the use of these compounds is highlighted due to their beneficial effects on the environment. To evaluate the chemical parameters of organic compounds: coffee peel (R1), passion fruit (R2), orange peel (R3) and food residues (R4), and their action on the germination and phytotoxicity of seeds for commercial use. Waste was acquired in March and April 2023; after completion of the composting process, analyzes were carried out for chemical characterization. The germination test was in a completely randomized design with 5 treatments and 5 replications. The soil and compounds were mixed, and after 10 days, the percentage, quantification of normal seedlings, measurement of the root and aerial part, dry and fresh mass weight, and the soybean germination index were evaluated. For the phytotoxicity test, 10g of dry matter of the compounds and 100 ml of distilled water were used, after shaking and centrifugation, 5 ml of the liquid was added to germination boxes with seeds of Lepidium sativum e Lactuca sativa. The chemical results obtained for the compounds were: total organic matter 12.71%, average C/N ratio 11% and average CEC 21.25 cmolckg-1. The compounds (average: 9.05) showed higher germination than the control (6.8). Although all were statistically different from the control (4.84cm), no significant differences were identified between the compounds (average: 9.73cm) with regard to root development. All compounds analyzed showed germination rates above 80% (average: 267%) indicating the presence of growth-stimulating properties. The compounds do not present toxicity in germination, but there was a difference between the control and the other treatments. The residues form different chemical concentrations of organic fertilizer for the soil, producing effectiveness in the development of cultivated plants.

**Keywords:** Sustainability, composting process and organic fertilizer.



Title	Evaluation of the role of redox metabolism in the tolerance of tardigrades to anhydrobiosis
Authors	MEDEIROS, Felipe Diego¹; RICART, Carlos André Ornelas¹; HERMES-LIMA, Marcelo¹.
Affiliations	Department of Cellular Biology, Biology Institute, University of Brasília
Session	Anatomy

#### Abstract and Keywords

Tardigrades are highly resilient microinvertebrates that endure extreme environments through cryptobiosis. Biochemical and physiological adaptations are crucial for their survival amidst the environmental stresses they face. This study aims to comprehend the adaptive mechanisms of these organisms, especially in response to desiccation, emphasizing the potential essential role of endogenous antioxidants in surviving, recovering from anhydrobiosis, and subsequently returning to normal levels of metabolic activity. Our investigation involves the use of two inhibitors - aminotriazole (ATZ) and buthionine sulfoximine (BSO) - targeting the antioxidant activity/capacity of tardigrades from the genera Paramacrobiotus and Milnesiumm collected at the Darcy Ribeiro campus of the University of Brasília. The animals are subjected to preincubation for 24 hours in different concentrations of enzymatic inhibitors, then are induced into anhydrobiosis. They remain in this anhydrobiotic state for 24 hours and are subsequently rehydrated. The project is in an experimental phase, but previous results indicated that pre-incubation with ATZ (1 mM) causes tardigrade death after 24 hours of anhydrobiosis, with no observed effect in the control group.

Currently, we are determining the activity of antioxidant enzymes catalase and glutathione S-transferase, as well as measuring levels of lipid peroxidation and carbonylated proteins. The study highlights the importance of enzymes and antioxidants for the survival of these animals after desiccation, indicating that anhydrobiosis requires strict control of ROS production. Tardigrades employ primary strategies to deal with the danger of oxygen toxicity, exhibiting an increasing efficiency of antioxidant defenses and metabolic control of energy production and consumption processes. The work provides evidence that antioxidant defenses, such as ROS-scavenging enzymes and other molecules, represent a key group of molecules necessary for desiccation tolerance and redox balance.

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**Keywords:** Tardigrades; Anhydrobiosis; Biochemical Adaptations; Endogenous Antioxidants; Redox Metabolism



Title	Anatomy of the superficial muscles of the forearm of the hoary fox Lycalopex vetulus (Lund. 1842)
Authors	Matheus da Costa Matutino Martins <sup>1</sup> , Luísa Benke Afonso <sup>1</sup> , Marina Fernandes Arbués Mota <sup>1</sup> , Carolina Freitas da Silva <sup>2</sup> , Romeu Paulo Martins Silva <sup>1</sup> , Zenon Silva <sup>1</sup> , Roseâmely Angélica de Carvalho Barros <sup>1</sup>
	<sup>1</sup> Federal University of Catalão (UFCAT), Institute of Biotechnology, Department of Biological Sciences, Laboratory of Comparative Anatomy of Wild Animals, Catalão-GO, Brazil.
Affiliations	<sup>2</sup> Federal University of Catalão (UFCAT), Institute of Biotechnology, Medicine Course, Catalão-GO, Brazil.
Session	Anatomy

# Abstract and Keywords

Anatomy is the science that studies the structural organization of the animal body, with the Muscular System being one of its focuses. The hoary fox (Lycalopex vetulus) is the only Brazilian canid species endemic to the cerrado and one of the least studied neotropical canids in the world. Therefore, the aim of this study is to describe, comparatively, the Anatomy of the superficial muscles of the forearm in hoary fox, discussing the results with data already existing in other wild animals, domestic animals and human literature, adding possible important information for the understanding the biology of this species, in addition to the health and conservation of wild species. The dissected structures are analyzed, described and discussed with the already well-established literature on domestic dogs and other species. The data are collected from the dissection, under authorization of CEUA/UFCAT, opinion no 01/22 of carcasses of four specimens of hoary fox (Lycalopex vetulus), two males and two females, adults, but without defined age, donated by the Wildlife Research Institute - IPEVIS and the Wild Animal Triage Center - CETAS of Catalão and/or collected on the sides of highways in Goiás and Minas Gerais (Authorization - SISBIO 37072-2). The experimental protocols are carried out in accordance with the recommendations of the Brazilian College of Animal Experimentation (COBEA) guidelines, using usual techniques in Macroscopic Anatomy. The muscles observed were: extensor carpi radialis; extensor digitorum communis; extensor digitorum lateralis; extensor carpi ulnaris; extensor carpi radialis; flexor digitorum superficialis; flexor carpi ulnaris; flexor carpi radialis; pronator teres; digitorum profundus. The results indicate a muscular pattern similar to that of other mammalian species, regarding the presence and general characteristics of the muscles.

**Keywords**: Comparative Anatomy, Hoary Fox, Muscular System



Title	The unusual posterior bilateral position of the mentual foramen
Authors	Ramiro Vilela Junqueira Neto¹; Isabella Carvalho Borges Lima¹; Izadora Viana de Souza¹; Adria Rodrigues Flores¹; Letícia de Souza Castro Filice²; Fabio Franceschini Mitri³
	<sup>1</sup> Dentistry School, Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil.
	<sup>2</sup> Departmento of Medical Clinic, Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil.
Affiliations	<sup>3</sup> Department of Human Anatomy, Federal University of Uberlandia,
	Uberlandia, Minas Gerais, Brazil.
Session	Anatomy

#### Abstract and Keywords

The mentual foramen is an anatomical reference at the level of the root apex of the premolar teeth in mandible, which is a noble anatomic structure because the path to the mentual nerve and vessels. Its identification is very easily performed by imaging exams, but is difficult by intraoral palpation. It is known that the foramen is sited between the mandibular premolar teeth, at the level of them root apex. The goal of this work is to present a radiographical finding of the unusual position of the mentual foramen, i.e posterior of the premolar teeth. So, we identified the posterior unusual localization of the mentual foramen in a digital panoramic radiography of the face, in the both sides of the mandible, which is very uncommon. The mentual foramen in both sides is posterior to the second premolar tooth, at them apex root level. This condition is very rare, once that it is didactically known the most prevalence of the mentual foramen between the root apices level of the root of the first and second premolar teeth. We concluded that this radiographical finding case could warn the dentist to a thorough radiographic observation in the mentual region, which in opposite case this situation could increase the vulnerability of the mentual nerve and vessels to injury in dental procedures. CEP opinion number: 5.544.523

**Keywords:** anatomy; mental foramen; mandible.



Title	The hyoid apparatus and a radiographic finding of calcification of the stylohyoid ligament
Authors	Cynthia Moreira Miranda¹; Thiago Henrique Mendes¹; Alice Rodrigues Moreira¹; Guilherme Santos de Souza¹; Letícia de Souza Castro Filice²; Fabio Franceschini Mitri³
	<sup>2</sup> Dentistry School, Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil.
	<sup>2</sup> Departmento of Medical Clinic, Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil.
Affiliations	<sup>3</sup> Department of Human Anatomy, Federal University of Uberlandia,
	Uberlandia, Minas Gerais, Brazil.
Session	Anatomy

#### Abstract and Keywords

The styloid ligament has close proximity to some structures on neck like blood vessels, nerves, and the submandibular gland. Its calcification is a one of the most common condition of the hyoid apparatus, which characterizes the Eagle syndrome. The purpose of this work is describe the anatomy of the hyoid apparatus and to present a radiographic finding of Eagle syndrome. The hyoid apparatus is constituted by the hyoid process, stylohyoid ligament and the hyoid bone, all originate embryologically in the second brachial arch embryologicaly from the second week of gestation onwards. It has fucntion to stabilize the hyoid bone, and contributes to the tongue and pharyngeal movements. The diagnosis of this syndrome is generally trough the imaging exams trough which it can be observed the elongation of the styloid process over 3 cm in length as a result of the calcification of the stylohyoid ligament. The symptoms could include discomfort or auricular pain, temporomandibular, cervical, salivar gland compression or even neuralgia. This is a asymptomatic case of the Eagle syndrome found in a panoramic radiography of face of a male adult 48 aged. The clinical exam could include the intraoral palpation in the tonsillar fosse in a case of prior suspicion. We concluded that the health professional, mainly the dentist, should know the anatomy of the hyoid apparatus and the Eagle syndrome, because the easy and quick access to the diagnosis by imaging exams and the set of symptoms eventually reported by the patient in anamnesis.

**Keywords:** anatomy; hyoid apparatus; stylohyoid ligament.



Title	Neuroanatomical aspects of the embryonic development of the vertebrate Hemisorubim platyrhynchos
Authors	Gabriel Borges da Silva <sup>1</sup> Claudemir Kuhn Faccioli <sup>2</sup>
Affiliations	<sup>1</sup> Student, Institute of Biology, Federal University of Uberlândia, Uberlândia- MG, Brazil.
	<sup>2</sup> Professor, Institute of Biomedical Sciences, Department of Human Anatomy, Federal University of Uberlândia, Uberlândia-MG, Brazil.
Session	Anatomia

# Abstract and Keywords

Neuroanatomy is the branch of morphology dedicated to the study of the nervous system (NS). When applied to embryology, it leads to an understanding of the morphology of neurodevelopment, crucial for comprehending species habits and their applications in studies. Thus, the objectives of this work are to analyze the initial phases of neurodevelopment in the pimelodid Hemisorubim platyrhynchos. To achieve this, eggs and larvae of H. platyrhynchos were collected, fixed in modified Karnovsky, and subjected to anatomical and histological analyses of the CNS and sensory organs from fertilization to 423 hours post-fertilization (HPF). The results indicate that the development of the NS is rapid, progressing from a few dilations of the embryonic axis at 3-5 HPF to numerous dilations due to cell multiplication at 6 HPF and advanced embryonic development with well-defined cranial, encephalic, caudal, and optic vesicle regions at 9-10 HPF. At 13 HPF, increased dilations with intense cell migrations in the antero-dorsal region of the embryo and a well-developed dorso-caudal region with clearly defined somites were observed. By 17 HPF, the encephalic and sensory structures were welldeveloped, displaying noticeable eye pigmentation and more apparent divisions between the prosencephalon and mesencephalon in the anterior cranial cavity, between the mesencephalon and cerebellum in the middle region, and between the cerebellum and medulla oblongata in the posterior region. Additionally, welldefined anterior and posterior encephalic ventricles were visible. At approximately 75 HPF, the CNS development reached its final stages, with visible structures such as eyes, optic calyces, optic vesicle, and maxillary barbels showing advanced development. Therefore, it is concluded that the neurodevelopment of *H. platyrhynchos* is remarkably rapid, with significant morphological advances at 10, 17, and 21 HPF, and complete neuromorphology at approximately 75 HPF.

**Keywords:** Animal Neurodevelopment; Comparative Neuroanatomy; Embriology.



Title	Morphology of the abdominal circulation of the ocelot (Leopardus Pardalis)
Authors	Kleysson Gonçalves de Souza <sup>1</sup> ; Carolina Freitas da Silva <sup>1</sup> ; Matheus da Costa Matutino Martins <sup>2</sup> ; Eduardo Paul Chacur <sup>1</sup> ; Thiago Montes Fidale <sup>1</sup> ; Zenon Silva <sup>2</sup> ; Juan Fernando Vélez García <sup>3</sup> ; Roseâmely Angélica de Carvalho Barros <sup>2</sup> ; Romeu Paulo Martins Silva <sup>1,2</sup>
Aff iliations	<sup>1</sup> Federal University of Catalão (UFCAT), Institute of Biotechnology, Medicine Course, Catalão-GO, Brazil. <sup>2</sup> Federal University of Catalão (UFCAT), Institute of Biotechnology, Department of Biological Sciences, Laboratory of Comparative Anatomy of Wild Animals, Catalão-GO, Brazil. <sup>3</sup> Universidade del Tolima, Veterinary Medicine, Ibague, Colombia
Session	Anatomy

# Abstract and Keywords

Introduction: Anatomy is an important tool in understanding the morphology of wild species, especially those threatened, such as the Ocelot (Leopardus pardalis). Being able to propose better strategies for treating and promoting the health of these animals. Objective: To describe the abdominal arterial and venous circulation of the Ocelot (Leopardus pardalis). Methodology: Two specimens of Ocelot (Leopardus pardalis) were used, donated by the Wildlife Research Institute – IPEVIS and the Wild Animal Triage Center CETAS of Catalão (SISBIO 37072 - 2), Opinion of the ethics committee in the use of animals/CEUA Protocol 01/22. To fix the material, a 10% aqueous formaldehyde solution was used via perfusion and the vascular system was stained with latex. Macroscopic analysis was performed accurately and photodocumented. Results: The first branch of the abdominal aorta artery would be that of the celiac trunk, below the superior mesenteric artery, which is larger, heading towards the largest portion of the gastrointestinal tract. The abdominal aorta follows at a level below the vena cava, and the branch for the superior mesenteric emerges crossing over the inferior vena cava at a height above the kidneys. just below the right and left renal arteries. The inferior vena cava follows the abdominal aorta to the right, so that the phrenic-abdominal vein flowing into the inferior cava can be seen at a higher level. Soon after, the right and left renal veins flow into the vena cava. In the right renal vein, an anatomical variation was observed, so that there were 3 branches leading to the renal hilum, two of which were larger and one was smaller. In the left renal vein, it is clear that the gonadal vein flows into it at a 90° angle. Conclusion: There was a similarity with jaguars (Panthera onca). They can be correlated with domestic animals, such as cats.

**Keywords:** Leopardus pardalis; Vascular system; Anatomy.



Title	Sperm chromatin alterations identified by the chromatin dispersion method quantified through computational image analysis and their correlation with fertility and <i>in vitro</i> embryonic development
Authors	Kamila Alves Fontoura¹; Marcelo Emílio Beletti²
Affiliations	<sup>1</sup> Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil.
	<sup>2</sup> Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia,
	Minas Gerais, Brazil.
Session	Laboratory animal science

#### Abstract and Keywords

In 2020, agribusiness generated R\$1.98 trillion, accounting for 27% of Brazil's Gross Domestic Product, with livestock representing 30% of this, highlighting the importance of high zootechnical performance in livestock herds. Sperm chromatin alterations are significant contributors to male fertility issues. Routine sperm analysis does not assess chromatin decompaction; however, the "Chromatin Dispersion Analysis (ADC)" method can identify DNA fragmentation and chromatin decompaction, albeit subjectively assessed. Nine bull semen samples were used to apply the ADC technique. Subsequently, images of the slides were captured using a photomicroscope and evaluated using the "Matlab" program, which automatically identifies the number of spermatozoa with and without chromatin dispersion and calculates the dispersion area for spermatozoa with alterations. The same semen samples were used for In Vitro Embryo Production (IVEP); the ovaries were obtained from a commercial slaughterhouse. From the IVEP, cleavage and blastocyst rates were obtained, correlated with image analysis data. It was possible to identify spermatozoa with small, medium, and large halos. Subsequently, the average pixel area of the halos was computationally determined. The average rates of all IVEP were correlated with the presence, size and computationally assessed halo area. Visually assessed halo size showed no significant correlation with IVEP rates. However, the presence of a halo (r=-0.75) and computationally determined halo area (r=-95) exhibited a significant negative correlation only with the embryonic development rate. In other words, halo formation identifies chromatin alterations that do not interfere with the fertilization process but impact early embryonic development, and the larger the halo area, the greater this interference. The ADC method can identify alterations based on halo formation in spermatozoa with fragmented DNA and this alteration interferes with early embryonic development.

**Keywords:** Spermatozoa, chromatin, *In vitro* embryo production.



Title	The environmental restriction's impact on the anxiety measurement in the ethoexperimental zebrafish (danio rerio) model
Authors	Pedro Henrique Alves Martins <sup>1</sup> ; Joaquim Carlos Rossini <sup>1</sup>
Affiliations	¹Instituto de Psicologia, Universidade Federal de Uberlândia, Brasil.
Session	Ciência de Animais de Laboratório

### Abstract and Keywords

The zebrafish (Danio rerio), a small fish from South Asia, is extraordinary for laboratory scientific researches into animal behavior, since it has a low-cost rear in bioteriums, a fast reproduction rate, and a great similarity with humans in physiological aspects. This species shows, starting from its larval stage until maturity, a solid behavioral repertoire consisting of anxiety stereotypical answers, such as freezing and erratic swimming, which are marked by sudden changes in the swimming direction and speed. An important ethoexperimental test for the observation of these behaviors is the novel tank, in which the animal is put into an aquarium divided by depth zones. The time spent in the aquarium's lower section is interpreted as an anxiety behavioral indicator. This study aims to investigate the zebrafish environmental restriction effects in this novel tank, confining it through the beaker protocol. It used 60 mature animals of both genders, divided in two groups (control and experimental). The control group was submitted just to the novel tank test, and the experimental group was submitted first to a 15 minutes restriction period in a beaker (beaker protocol), and then exposed to this novel tank. The experimental group results show a relevant raise in its environment exploration, and swimming speed and acceleration in the novel tank test. These parameters can be associated with anxiety stereotypical behaviors in this species, such as erratic swimming. However, it did not show statistically relevant differences between the groups in the freezing and in the time spent in the novel tank test zones. A possible explanation for this is the 'ceiling effect', in which direct contact with the animals may have caused this resemblance.

**Keywords:** zebrafish, ethoexperimental test, anxiety.



Title	Standardization of an experimental protocol for gestational diabetes in animal model
Authors	Fernanda Naves Araújo do Prado Mascarenhas¹; Maria Júlia Neves Ribeiro¹; Natália Ferreira Silva¹; Rener Mateus Francisco Duarte²; Foued Salmen Espindola²; Renata Graciele Zanon¹
Affiliations	<sup>1</sup> Institute of Biomedical Sciences, Federal University of Uberlandia (UFU), Uberlandia, MG, Brazil <sup>2</sup> Institute of Biotechnology, Federal University of Uberlandia (UFU), Uberlandia, MG, Brazil.
Session	Ciência de Animais de Laboratório

#### Abstract and Keywords

Diabetes mellitus (DM) is a metabolic syndrome characterized by a dysfunction in insulin production, secretion and/or action, causing hyperglycemia. When the disease occurs during pregnancy, it is called gestational diabetes mellitus (GDM), and the hyperglycemic state can affect the fetus's development. The present work aimed to standardize an experimental model of efficient GDM induction in Wistar rats using streptozotocin (STZ). Therefore, the animals obtained by REBIR-UFU, CEUA 056/18, were placed for mating overnight, and the following morning the presence of spermatozoa was verified in the females' vaginal wash, an indication of first gestational day (G1), and after five days (G5), pregnant rats were induced to diabetes, having previously fasted for 24 hours, intraperitoneally, using STZ, at a dose of 50 mg/kg. The data evaluated were the mothers' weight (g) and blood glucose (mg/dL), during pre-mating, G7 and G17. They were considered diabetic when blood glucose  $\geq 200$  mg/dL. After birth, the puppies were evaluated for the number of individuals, sex and stillbirths. And after the weaning period, the mothers' blood was collected to evaluate markers of oxidative damage present in plasma serum. We achieved 55.6% success in obtaining the experimental model of GDM, as we were able to implement a sequence of essential care (mating, vaginal wash, fasting, dose), with proof of the establishment of the pathology characterized by evolution weight gain, hyperglycemia and oxidative damage in maternal blood, in addition to changes in the quality of the litter (that characterize the hyperglycemic environment as hostile to the development of the fetus). It is considered an effective protocol, superior to those commonly observed by researchers in the DMG study area, making it possible to avoid the excessive use of animals in this research model.

**Keywords:** diabetes, gestation, methods, streptozotocin



Title	Lactobacillus lactis, a little explored probiotic, can colonize animals in embryonic life
Authors	Maria Vitoria Mendes Felix Costa <sup>1</sup> , Lara Reis Gomes <sup>1</sup> , Vasco Ariston de Carvalho Azevedo <sup>2</sup> , Simone Sommerfeld <sup>1</sup> , Gabriela Zangari Cardoso <sup>1</sup> , Thais Fernanda Martins dos Reis <sup>1</sup> , Belchiolina Beatriz Fonseca <sup>1,3</sup>
Affiliations	<sup>1</sup> Faculty of Veterinary Medicine, Federal University of Uberlandia, Uberlândia - MG, Brazil; <sup>2</sup> Department of Genetics, Ecology, and Evolution, Federal University of Minas Gerais, Belo Horizonte-MG, Brazil/ Postgraduate Program in Animal Science in the Tropics at the Federal University of Bahia; <sup>3</sup> Postgraduate Program in Genetics and Biochemistry, Federal University of Uberlandia, Uberlândia - MG, Brazil.
Session	Laboratory Animal Science

# Abstract and

This study aimed to evaluate if the Bacillus velezensis (BV) and Lactococcus lactis subsp. lactis (LL), two bacteria with probiotic properties but little studied, can colonize the intestine of the chicken embryo (CE) until birth and potentially protect them from diseases. We selected two strains of BV and LL that inhibit Salmonella Heidelberg (SH) and extra-intestinal E. coli (EC). The selected strains showed immunomodulatory effects in cell culture. After the ethical comittee approved the experiment (certificate number: 23117.022979/2023-60), we tested the strains in CE, which serve as a wonderful vivo model. Then we inoculated 3 log UFC/CE of BV and LL alone or in association in CE at 18 days of incubation via allantoic fluid and when the broiler was born, we counted the probiotic bacteria in the intestines. The number of CE was 5 (per group) and we tested negative control in paralell. Both probiotic bacteria multiplied and colonized the intestines of CE however wille LL colonized intestine of all CE tested, BV colonized just one chicken. The mean of LL was 5.76-6.09 log UFC/g of intestine and BV was 3.39 UFC/q of intestine. Our research team was able to find beneficial bacterial strains that successfully colonized the gastrointestinal tracts of birds, resulting in control of two notorious pathogens, SH and EC with immunomodulatory effects and improvement in intestinal health. The success of this intervention makes CE and chicks an excellent model system for testing the efficacy of probiotics in controlling pathogenic microorganisms in other animals and humans. Additionally, considering that chicken meat can transmit SH and EC to humans, we proved that BV and LL can colonize the intestine of broilers resulting in an inocuos food for human consumption. The findings provide a solid foundation for future studies to build upon, ultimately contributing to the development of novel preventive measures against foodborne illnesses and promoting public health worldwide.

Ethics Committee: Certificate number 23117.022979/2023-60

**Keywords:** inhibition, probiotics, chicks, chicken embryo, model systems.



Title	Laboratory animals, the protagonists of science!
Authors	Ludmilla S. Mendes <sup>1</sup> ; Sandra G. Klein <sup>1</sup> ; Isabela D. S. Rocha <sup>1</sup> , Milene C. O. Ferreira <sup>1</sup> ; Pedro H. R. Marcelino <sup>1</sup> ; Isabela L. Lima <sup>1</sup> ; Flávia B. Ferreira <sup>1</sup> ; Luan Freitas Ferreira <sup>2</sup> ; Matheus Morais Neves <sup>1</sup> ; Thomas Santos Arrais <sup>1</sup> ; Giovana Magalhães Ferreira <sup>1</sup> ; Ray César Silva <sup>1</sup> , Murilo Vieira da Silva <sup>1</sup> .
	<sup>1</sup> Biotechnology Laboratory in Experimental Models - LABME, Federal University of Uberlândia, Uberlândia - MG, Brazil.
Affiliations	<sup>2</sup> Undergraduate Course in Literature (English), Federal University of Uberlândia, Uberlândia- MG, Brazil.
Session	Laboratory Animals Science

#### Abstract and Keywords

The use of laboratory animals as experimental models has been essential to boost scientific progress, covering the validation of medicines, treatments, development and physiology studies, among others. However, although they play a crucial role in science, the public has little knowledge about the subject, leading to several doubts regarding their purpose, which species are most used, what management is carried out and whether there are welfare protocols applied to the use of these animals.

The initiative "Laboratory Animals, the protagonists of science!" presented at the "Brincando e Aprendendo" event held at the Dica Museum, was created with the aim of presenting what is carried out within the biotechnology laboratory and experimental models, aiming to resolve the doubts created by misinformation. To this end, different dynamics were presented at the event, such as educational videos, a display of mouse histological slides under an electron microscope, a presentation guided by members on animal experimentation, together with an exhibition of banners and interactive banners,

where the public, after absorbing the content taught by employees, can train

their knowledge using games present on the interactive banner. In conjunction with the presentation and public interaction, gifts were also offered with the aim of encouraging visitor participation in the event.

In total, the meeting reached approximately 500 people of different age groups per presentation day during its three days, welcoming families and students at primary, secondary and higher education levels, where it promoted awareness and interest in an academic career. Understanding the relevance of laboratory animals in science helps to clarify doubts, demystify misconceptions and encourage interest, in addition to cultivating due respect for both animals and the science that involves them.

**Keywords:** experimental model, scientific divulgation, laboratory animals



Title	The role of iron accumulation in the neurodegenerative disease development in chronic experimental infection by toxoplasma gondii
Authors	Lucas Mauricio Lopes de Navasquez, <sup>1</sup> Anna Laura de Jesus Gomes, M.Sc, <sup>1</sup> Iasmin Aparecida Cunha Araújo, Ph.D, <sup>1</sup> Flavia Batista Ferreira, Ph.D, <sup>1</sup> Allison Benatti Justino, Ph.D <sup>2</sup> Ana Luiza Silva Borges, M.Sc <sup>2</sup> Vinicius Prado Bittar, M.Sc <sup>2</sup> Foued Salmen Spindola, Ph.D, <sup>2</sup> Neide Maria da Silva, Ph.D, <sup>1</sup>
Affiliations	All authours: <sup>1</sup> Institute of Biomedical Science, Federal University of Uberlandia, Uberlandia, Brazil. <sup>2</sup> Institute of Biotechnology, Federal University of Uberlandia, Uberlandia, Brazil.
Session	Laboratory Animal Science

# Abstract and Keywords

Toxoplasma gondii is an obligate intracellular parasite, worldwide disseminated, infecting numerous animals, capable of affecting the central nervous system, generating neuroinflammation, neurotransmitter imbalance, and recently has been proposed as a risk factor in the pathophysiology of neuropsychiatric disorders. There is a kind of cell death that is the iron-dependent. Nowadays, it is implicated in the pathological cell death associated with degenerative diseases (i.e. Alzheimer's, Huntington's and Parkinson's). This study aimed to analyze the effect of the chronic infection in the development of neurodegenerative disease dependent of iron accumulation in the brain. For this, BALB/c and C57BL/6 mice were orally infected with cysts of ME49 T. gondii strain and the behavioral, histological, and immunological parameters were analyzed in chronic phase. It was observed that C57BL/6 mice were more susceptible than BALB/c mice. The parasitism in the brain analyzed by immunohistochemistry was assessed by counting the number of parasitophorous vacuoles and cysts in 40 microscopic fields, using a 40x objective. C57BL/6 mice showed an average of 14.0, 25.3 and 10.0 cysts and, 480, 220 and 170 tachyzoites on days 30, 60 and 90, respectively, showing a decrease in tachyzoites numbers over the time. Iron deposition was identified by Perls staining and analyzed using Aperio ImageScope Software with 20x magnification. An average of 87.6, 60.0 and 280.25 iron marked points was observed, while in the control group it was 19.2. In spatial memory test at 30, 60 and 90 days post-infection, it was detected cognitive deficit, with an alternation rate below 60%. In conclusion, chronic T. gondii infection may represent a risk factor for the development of neurodegenerative disorders, dependent of iron accumulation in the brain.

**Keywords:** *Toxoplasma gondii*, chronic, memory, iron, neurodegenerative disorder.



Title	Effect of chronic <i>Toxoplasma gondii</i> infection on depressive-like behavior in mice exposed to chronic stress condition
Authors	Anna Laura de Jesus Gomes, M.Sc. <sup>1</sup> Flávia Batista Ferreira, Ph.D. <sup>1</sup> Ana Paula Navarro Gonçalves, M.Sc <sup>1</sup> Tiago Wilson Patriarca Mineo, Ph.D. <sup>1</sup> Fernanda Maria Santiago,Ph.D. <sup>1</sup> José Roberto Mineo, Ph.D <sup>1</sup>
Affiliations	All authours: <sup>1</sup> Institute of Biomedical Science, Federal University of Uberlandia, Uberlandia, Brazil.
Session	Laboratory Animal Science

### Abstract and Keywords

Toxoplasma gondii is an obligatory intracellular parasite that infects approximately one-third of the global human population, serving as the etiological agent of toxoplasmosis. Studies have demonstrated its capacity to infect a diverse range of species, establishing latent infections in the brain and implicating its role in the pathogenesis of various psychiatric disorders, including schizophrenia, anxiety, obsessive-compulsive disorder, impulsivity disorders, and depression. The present study sought to investigate whether chronically infected mice with T.gondii, subjected to a chronic stress condition through movement restriction, displayed altered behavior. BALB/c mice were categorized into four groups: control: without stress and chronic *T.gondii* infection; stressed: animals subjected to stress without chronic *T.gondii* infection; infected: animals without stress but with *T.gondii* infection; infected/stressed: animals experiencing both condition. Animals received 500 tachyzoites via intraperitoneal. The stress lasted 15 days. All groups were submitted to forced swim test and analyzed after 52 days. The impact on mice subjected to chronic T.gondii infection revealed an increase in depressive-like behavior compared to groups under stressed or non-stressed conditions, as well as to the both groups of uninfected animals. Notably, the immobility time of infected mice was higher, they spent less time swimming than the uninfected animals. No significant differences were observed in anti-T.qondii levels or parasite load in the brains of both groups infected mice. Furthermore, akin to the group of stressed animals alone, the infected/stressed animals exhibited lower body weight gain compared to the control or only infected groups. In conclusion, our findings unveil new perspectives on understanding the consequences of chronic infection, specifically in the presence of parasite cysts within the central nervous system.

**Keywords:** *Toxoplasma gondii*; Chronic infection; Stress condition; Depressive-like behavior



Title	Understanding the chicken embryo as a model of Campylobacter jejuni infection beyond death
Authors	Paula Fernanda de Souza Braga¹; Emília Rezende Vaz²; Simone Sommerfeld¹; Fabiana de Almeida Araujo Santos²; Alessandra Aparecida Medeiros-Ronchi¹; Belchiolina Beatriz Fonseca¹²
Affiliations	<sup>1</sup> Graduate Program in Animal Science, Federal University of Uberlândia, Uberlândia, MG, Brasil
	<sup>2</sup> Graduate Program in Genetics and Biochemistry, Federal University of
	Uberlândia, Uberlândia, MG, Brazil.
Session	Laboratory Animal Science

# Abstract and Keywords

Campylobacter jejuni (CJ) is one of the main agents related to foodborne infections. Although CJ typically causes a self-limited immunocompetent humans, vulnerable populations may manifest arthritis or Guillain-Barré syndrome. A better understanding of infection mechanisms and the response of a host organism make in vivo models essential for preliminary studies. Chicken embryos (CE) offer a simple method to study complex biological systems with high reproducibility and lower cost. Therefore, we aimed to evaluate the virulence and infection of CJ strains isolated from chickens (C030/30, C046/10, and C092/6) and humans (IAL 2383) to better understand the pathogen-host relationship (Ethics Committee 10/2022/CEUA/PROPP/REITO). To understand the changes caused by CJ, we inoculated low infective doses (2.5 log CFU/CE). Positive control group (PC) was inoculated with Salmonella Typhimurium (ST), and two negative controls inoculated with saline 0.9% and a probiotic strain of Bacillus subtilis. Then, we evaluated embryo mortality, weight, gross and microscopic lesions and the counts of macrophages and lymphocytes (TCD4 and TCD8) using flow cytometry analysis. At low doses, all CJ strains resulted in low mortality. We found no statistical difference among the groups when comparing the percentage of monocytes and lymphocytes TCD8+. There was an increase in TCD4+ in the positive control (ST). CE infected by CJ strains IAL 2383 and C030/3 had significant differences from the PC. The strains CJ IAL, C030/3, and C092/6 induced an discreet inflammatory change in the liver of CE similar to the PC. In conclusion, at a low dose, CJ generates lesions in chicken embryos, and some strains can stimulate the immune system without a defined response pattern, emphasizing that the host-pathogen relationship is straindependent. These findings reinforce the importance of studying the virulence, infection, and immune response of various strains of CJ and the CE model's utility.

**Keywords:** Foodborne pathogens, zoonotic bacteria, innate immune response, human campylobacteriosis



Title	Comparative evaluation of dry and liquid biochemical analyses in murine models
Authors	Milene Caroline de Oliveira Ferreira; Renan Faria Guerra; Flavia Batista Ferreira França; Isabela Lemos de Lima; Thomas Santos Arrais; Matheus Morais Neves; Ludmilla Silva Mendes; Pedro Henrique Ribeiro Marcelino; Sandra Gabriela Klein; Murilo Vieira Silva.
Affiliations	Laboratório de Biotecnologia em Modelos Experimentais - LABME, Universidade Federal de Uberlândia, Uberlândia 38.405-330, Brasil
Session	Ciência de Animais de Laboratório

# and Kevwords

The in vitro analysis of biochemical analytes is crucial for animal health assessment, allowing for the quantification of biological markers in bodily fluids, thereby contributing to effective clinical management. Dry Biochemistry conducts automated analyses for quantifying analytes through photoelectric colorimetry, differing from liquid biochemistry by requiring minimal biological material and eliminating the use of reagents, providing automation and rapid results. This study aimed to assess the feasibility of analytical validation of Dry Biochemistry, available for use in dogs and cats, for murine biochemical parameter analysis. Groups of male and female mice (n=25) were formed, with 500-600µl of blood collected via retro-orbital puncture from C57BL/6 and BALB/c mice aged 3-4 weeks in an SPF system. Fifteen microliters of serum samples were pipetted and subjected to biochemical and statistical analyses using the unpaired t-student test with significance defined at p<0.05 in Origin 8.0 software. During the analysis of Total Protein, GGT, and AST tests, the existence of discrepant values within the established reference range for dogs was observed, although there were no outliers within the linear range of the methods used. Validation of analyte reference intervals is necessary, considering the different strains of murine models analyzed, the utility of calibration curves, the availability in the kits of the tested analytes, and the lack of statistically significant difference at p<0.05 between the groups for Total Protein and GGT. However, the presence of outliers at the upper limit of the reference range resulted in a significant difference for AST. Therefore, proceeding with equipment validation tests in murine models is viable, following regulations from the Ministry of Agriculture, Livestock, and Supply (MAPA) and the company responsible for national production and distribution.



Title	Doxorubicin reduces heart rate variability in rats
Authors	Guimarães L.C. <sup>1</sup> ; Fidale TM. <sup>2</sup> ; Pereira TCR <sup>1</sup> ; Deconte SR <sup>3</sup> ; Lopes PR <sup>4</sup> ; Ferreira- Junior MD <sup>5</sup> ; Ferreira-Neto ML <sup>6</sup> ; Brito WS <sup>7</sup> ; Gomes RM <sup>5</sup> ; de Souza FR <sup>1</sup> ; Cavalcante KVN <sup>6</sup> ; Herrera GC <sup>8</sup> ; Armond BM <sup>9</sup> ; Resende ES <sup>10</sup>
Affiliations	<ol> <li>Experimental Medicine Laboratory, Federal University of Uberlândia (UFU), PhD candidate in the Health Sciences Department - PGCS, Faculty of Medicine, Uberlândia, MG, Brazil;</li> <li>Professor at the Dept of Medicine, Federal University of Catalão (UFCAT), Catalão, Goiás, Brazil;</li> <li>Laboratory Technician/Biology, Institute of Biomedical Sciences, Dept of Physiology, Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil;</li> <li>Faculty of Dentistry - FOAr, Department of Physiology and Pathology, São Paulo State University "Júlio de Mesquita Filho" – UNESP, Araraquara, SP, Brazil;</li> <li>Dept of Physiological Sciences, ICB, Federal University of Goiás (UFG), Goiânia, GO, Brazil;</li> <li>Dept of Physioloy, Biomedical Science Institute, Federal University of Uberlândia, Uberlândia, MG, Brazil;</li> <li>Statistician, Federal University of Piauí (UFPI), Teresina, PI, Brazil;</li> <li>Laboratory Technician, Veterinary Hospital, Federal University of Uberlândia, MG, Brazil;</li> <li>Professor, Dentistry Course, Salgado de Oliveira University – UNIVERSO, Rio de Janeiro, RJ, Brazil;</li> <li>PPG in Health Sciences, Faculty of Medicine, Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil.</li> </ol>
Session	Poster Session

## Abstract and Keywords

Doxorubicin is a widely used chemotherapeutic agent that, despite its efficacy in treating various types of malignant tumors, has limitations due to its cardiotoxic effects. This study aimed to verify the cardiac actions generated by the administration of doxorubicin in rats, using heart rate variability (HRV) analysis. Twenty Wistar rats were divided into two groups: Control (C) and Doxorubicin (D), with animals in the D group receiving six intraperitoneal injections of doxorubicin totaling a cumulative dose of 75 mg/kg. The HRV analysis showed that doxorubicin induced an increase in sympathetic activity, evidenced by an increase in heart rate and systolic blood pressure. Specifically, a significant increase in mean arterial pressure (MAP) from 105.82 to 115.31 mmHg, in heart rate (HR) from 363.67 to 401.43 bpm, and in systolic blood pressure (SBP) from 127.07 to 141.04 mmHg was observed in the D group compared to the C group. Furthermore, statistically significant differences were found in the spectral analysis of the frequency domain, with the LF component (low frequency) increasing from 8.1429 to 14.7143 ms^2 and the LF/HF ratio (indicative of the sympathetic/vagal balance) increasing from 0.2457 to 0.7957, suggesting increased sympathetic activity. The HF component (high frequency), on the other hand, showed higher values in the C group compared to the D group, reflecting a reduced parasympathetic activity induced by doxorubicin. In the time domain, the R-R interval showed higher values in the C group, indicating a reduction in heart rate variability in rats treated with doxorubicin. The symbolic analysis also revealed significant differences, particularly in the OV component, with increased values in the D group, reinforcing the predominance of sympathetic modulation. This study, approved by the Ethics Committee on Animal Use (CEUA) under number 045/19, demonstrates that doxorubicin compromises the autonomic modulation of the heart, favoring sympathetic activity and reducing heart rate variability, which may contribute to the risk of developing cardiotoxicity.

**Keywords:** Heart rate variability, Doxorubicin, Cardiotoxicity



Title	Pre-Clinical evaluation of cardiovascular alterations induced by the extract of erythroxylum suberosum a. St. Hilaire
Authors	Ingrid França Matos¹; Stefanne Madalena Marques¹;Giuliana Muniz Vila Verde²; James Oluwagbamigbe Fajemiroye³;Gustavo Rodrigues Pedrino¹
Affiliations	<sup>1</sup> Center of Neuroscience and Cardiovascular Research - Federal University of Goiás, Goiânia-GO, Brazil <sup>2</sup> Natural Products Laboratory - State University of Goiás, Anápolis-GO, Brazil <sup>3</sup> Department of Pharmacology - Federal University of Goiás, Goiânia-GO, Brazil
Session	Biologia e Doenças Cardiovasculares

## Abstract and

Hypertension (HA) represents a significant public health issue, characterized by persistent elevation of blood pressure (BP). HA is often asymptomatic and constitutes a primary risk factor for the development of cardiovascular, cerebrovascular, and renal diseases. The Brazilian cerrado biome harbors a rich diversity of plant species, among which the genus Erythroxylum stands out. This genus encompasses several species with biomedical potential, exemplified by plants of the species *Erythroxylum suberosum* A. St. Hilaire (*E. suberosum*). However, few studies have been conducted to validate the antihypertensive potential of these plants. Therefore, the objective of this study was to evaluate the antihypertensive potential of the crude extract of E. suberosum. To this end, Wistar rats (n=6) and spontaneously hypertensive rats (SHR) (n=6), weighing between 250-300 grams. All experimental procedures were submitted to the Ethics Committee for Animal Use of UFG (process number: 061/17). The animals were underwent to surgical procedures for the unanesthetized recording of mean arterial pressure (MAP) and heart rate (HR) following bolus administration of three doses of E. suberosum extract (0.08 mg/kg; 0.16 mg/kg; 0.25 mg/kg). Our results demonstrate that the extract did not induce alterations in MAP in normotensive rats ( $\Delta$  vehicle: 4,04 ± 2,4 mmHg;  $\Delta$  0,25 mg/kg: 1,5 ± 0,95 mmHg). In SHRs, the highest dose significantly reduced MAP ( $\Delta$  vehicle: -2,14  $\pm$ 2,85 mmHg vs.  $\Delta$  0,25 mg/kg: - 13,62 ± 3,80 mmHg; p<0,05). The extract did not induce changes in HR in normotensive ( $\Delta$  vehicle: -11,07 ± 5,49 bpm vs.  $\Delta$ 0,25 mg/kg: 7,15  $\pm$  7,28 bpm) or SHR ( $\Delta$  vehicle: 1,60  $\pm$  9,90 bpm vs.  $\Delta$  0,25 mg/kg:  $-16,32 \pm 22,29$ bpm). Collectively, our data corroborate the antihypertensive potential of E. suberosum extract. However, new studies must be carried out to define possible mechanisms of action.

**KEYWORDS:** Hypertension; Cardiovascular Disease; Plant Extract; *E. Suberosum* 



Title	Assessment of cardiotoxic potential and risk stratification for pesticides used in Brazilian agro-industrial practice
Authors	Alessandra Silva Teles <sup>1</sup> , Gabriel Rugai Cunha Lima <sup>1</sup> , Luís Fernando Freire Figueiredo <sup>1</sup> , Prof. Dr. Artur Santos Miranda <sup>1</sup>
Affiliations	<sup>1</sup> Biophysics and Physiology Department, UFMG, Belo Horizonte – MG, Brazil
Session	15/05, 16/05

# and Keywords

Brazil stands as a pivotal agro-exporter while also ranking among the largest consumers of pesticides in the world. There's widespread agreement that the intensive use of these chemicals leads to soil, food, and water contamination, with cardiovascular issues also linked to pesticides exposure. However, current regulations do not mandate cardio-toxicological analysis for their monitoring and risk management. As so, our understanding of the extent of pesticide exposure in relation to cardiovascular disease development remains limited, both regarding chronic exposure effects and its underlying cellular mechanisms. In this study, we delve into the primary pesticides known for their cardiotoxic effects, aiming to establish a risk stratification for their potential cardiotoxicity. For the cell-based assays, H9c2 cell line myoblasts were subjected to 24 hours of exposure to the pesticides Mancozeb and Endosulfan. Cytotoxicity was assessed through Annexin V/Propidium Iodide fluorescence using flow cytometry. Production of reactive species was compared via fluorescent probes imaging. Cell membrane potential was evaluated using the whole cell patch-clamp technique. Afterwards, statistical analysis involved Shapiro-Wilk normality tests, unpaired t-tests, variance analysis and Tukey's test.

High cytotoxicity was observed in flow cytometry assays following exposure to Mancozeb (95,6% cell viability in control group vs. 69,9% [1 $\mu$ M] vs. 1,96% [1 $\mu$ M], R² = 0.9616, IC50 = 1.957 $\mu$ M). Among the fluorescence imaging assays there was a notable increase in total reactive oxygen species production in cells exposed to both Mancozeb (1,46 RFU in control group vs 9,26 RFU [10 $\mu$ M], p<0,001) and Endosulfan (3,92 RFU control group vs 6,57 RFU [10 $\mu$ M], p=0,02189).

We conclude that the evaluated pesticides show cardiotoxic potential, as shown by cytotoxicity at low doses and alterations in reactive species production and membrane potential, key aspects of cardiac signaling.

**Keywords:** pesticides, Mancozeb, Endosulfan, cardiovascular, cardiotoxicity, reactive species, cytotoxicity.



Title	Changes in cardiac function in an experimental model of intestinal ulcerative colitis
Authors	Sofia Evelyn Veloso Gomes <sup>1,</sup> Ivan Lobo de Souza Marques <sup>2</sup> , Raquel da Silva Ferreira <sup>3</sup> , Mariana de Almeida Oliveira <sup>1</sup> , Ana Maria Caetano de Faria <sup>1</sup> , Jader dos Santos Cruz <sup>1</sup>
Affiliations	1Departamento de Bioquímica e Imunologia, Universidade Federal de Minas Gerais, UFMG 2Departamento de Fisiologia e Farmacologia, Universidade Federal de Minas Gerais, UFMG 3Departamento de Clínica e Cirurgia Veterinárias, Universidade Federal de Minas Gerais, UFMG
Session	pôster

# and Keywords

Inflammatory bowel diseases (IBD) consist of a systemic state of inflammation, intestinal ulcerative colitis (IUC) and Crohn's disease are included within these. IBD has been identified as influencing the incidence of cardio diseases and changes. However, only recently have studies on the aforementioned influence and inflammatory disease states been carried out in more depth. Still, the association between inflammatory bowel diseases and cardio diseases (CD) has not yet been widely discussed. Male mice of the C57BL/6 lineage were used and divided into two distinct groups: treated (IUC) and controls. UC was induced through the administration of a 2% aqueous solution of sodium dextran sulfate (DSS) for 21 days, with a week's break between day 7 and day 14. 30 ml of DSS (IUC ) and 30ml of water for controls. Electrocardiograms were performed on days 0, 7, 14 and 21. On day 21 the mice were euthanized to prepare cardiomyocytes. Treated mice showed left bundle branch block and decreased QRS complex amplitude and QTc interval. Cardiomyocytes from the control group stimulated at a frequency of 1 Hz showed:  $6.7 \pm 3.1$  shortening fraction, 0.1 ms  $\pm$  0 .01 time to peak, 0.03 ms  $\pm$  0.01 contraction speed, and 0.06 ms  $\pm$  0.02 relaxation speed. Cardiomyocytes from the treated group stimulated at a frequency of 1 Hz present:  $5.5 \pm 3.7$  shortening fraction, 0.1 ms  $\pm 0.02$  time to peak,  $0.03 \text{ ms} \pm 0.006 \text{ contraction speed}$ , and  $0.08 \text{ ms} \pm 0.02 \text{ relaxation speed}$ . Cardiomyocytes from the control group stimulated at a frequency of 5Hz resent:  $6.6 \pm 4$  shortening fraction, 0.06 ms  $\pm 0.01$  time to peak t, 0.01 ms  $\pm 0.002$ contraction speed, and 0. 04 ms ± 0.009 relaxation speed. Cardiomyocytes from the treated group stimulated at a frequency of 5Hz show:  $4.7 \pm 1.7$  shortening fraction, 0.06 ms  $\pm$  0.01 time to peak t, 0.02 ms  $\pm$  0.01 contraction speed cell, and 7 ms ± 32 cell relaxation speed. The study findings indicate that ulcerative colitis affects heart function. Compared to the control group, the ulcerative colitis group showed increased QRS duration and relaxation velocity, along with decreased QT interval and fractional shortening.

**Keywords:** Inflammatory bowel diseases, intestinal ulcerative colitis, cardio diseases

Aprovação na Comissão de ética: 123/20

Apoio financeiro: CNPq, FAPEMIG



Title	Detection of C-reactive protein based on silver nanoparticles: An optical diagnosis for an indicator of cardiovascular risk
Authors	Nicole. A. B. Madurro¹; Aquilla C. Souza²; Yara C. P. Maia¹
Affiliations	<sup>1</sup> School of Medicine, Federal University of Uberlandia, Uberlandia, Brazil <sup>2</sup> Biotechnology Institute, Federal University of Uberlandia, Uberlandia, Brazil
Session	Biologia e Doenças Cardiovasculares

## Abstract and Keywords

C-reactive Protein (CRP) levels correlate with the risk of cardiovascular diseases, as well as other infectious and inflammatory conditions. Rapid and straightforward diagnosis at the point-of-care is crucial for timely intervention by healthcare professionals, thereby enhancing the chances of successful treatment. Convential methods for detecting CRP in blood, such as immunoassays, immunoturbidimetry, latex agglutination, and nephelometry, necessitate skilled labor, are time-intensive, and relatively expensive. Optical and colorimetric approaches on nanoparticles have attracted attention due to their simplicity of analysis, cost-effectiveness, and better response time. In this study, we propose an optical method imploying silver nanoparticles (AgNP). Specifically, silver nitrate (10 mL, 1.0 mmol  $L^{-1}$ ) was added dropwise to a solution of sodium borohydride (30 mL, 2.0 mmol L<sup>-1</sup>), while cooled in an ice bath. The resulting mixture was vigorously stirred on magnetic plate until the colloidal suspension turned yellow. Subsequently, the obtained colloidal suspension was subjected to nanospectrophotometry, revealing a peak at 413 nm. Following this, anti-CRP antibody (0.1 mol  $L^{-1}$  in phosphate buffer, 5  $\mu L$ ) was added to the AgNPs suspension and allowed to incubate for 20 minutes. spectrophotometric analysis displayed a peak at 462 nm, indicating antibody binding to the nanoparticles surface, resulting in a bathochromic effect due to the increased nanoparticles size. The CRP antigen (0.1 mol  $L^{-1}$  in phosphate buffer,  $5~\mu L)$  was them added to the AgNP:anti-CRP complex and maintened at room temperature for 20 minutes. Further analysis using nanospectrophotometry revealed a peak at 471 nm, suggesting interaction between the anti-PCR probe and the target CRP. All experiments were conducted in triplicate. These findings hold promise for the rapid testing of samples obtained from patients with cardiovascular diseases as well as healthy individuals.

**Keywords:** diagnosis; silver nanoparticles; cardiovascular diseases.

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Title	Evaluation of the renal effects of nanoparticles loaded with essential oil from <i>Citrus sinensis</i> in spontaneously hypertensive rats
Authors	Karla de Aleluia Batista <sup>1</sup> ; Cassio Nazareno Silva da Silva <sup>2</sup> ; Juliana Vila Verde Ribeiro <sup>2</sup> ; Guilherme Ananias Cardoso <sup>2</sup> ; Júlio César Lima Silva <sup>2</sup> ; Kátia Flávia Fernandes <sup>2</sup> ; Pâmela Yasmin de Oliveira Ferreira <sup>2</sup> ; Paulo César Ghedini <sup>2</sup> ; Carlos Henrique Xavier Custódio <sup>2</sup> .
Affiliations	<sup>1</sup> Instituto Federal de Goiás, Goiânia, Goiás <sup>2</sup> Universidade Federal de Goiás, Goiânia, Goiás
Session	Biologia e Doenças Cardiovasculares

### Abstract and Keywords

The essential oil from Citrus sinensis (OE) can have important protective effects against the oxidative and inflammatory processes related to hypertension. However, some drawbacks related to its stability reduce its biological potential. In this sense, this study aimed to evaluate the renal protective effects of nanoparticles of acetylated cashew gum polysaccharide loaded with OE (acCGP@OE) in spontaneously hypertensive rats. The acCGP@OE nanoparticles were characterized by dynamic ligth scattering and transmission electron microscopy. The renal effects of the OE and acCGP@OE were evaluated using Wistar (normotensive control) and SH rats (CEUA-UFG nº049/22). OE or acCGP@OE were daily orally administrated for 30 days in a dosage of 75 mg Kg-1. After treatment the metabolic parameters and antioxidative effects in kidney tissue were evaluated, and the e renal protectant effect was studied. The data were analyzed by two-way ANOVA followed by Tukey's post hoc test. Results evidenced OE loading of 75.18% into the acCGP@OE, who presented a smooth and spherical morphology, with hydrodynamic diameter of 161nm, polydispersity of 0.141 and Zeta potential of -19.17mV. The treatment with acCGP@OE reduced the weight of the SH rats (17.4%, p<0.05), without interference with food and water ingestion, urinary volume, and feces content (p<0.05). The treatment also caused reduction of urinary sodium (21.1%, p<0.05 vs SHR group), serum creatinine (50%, p<0.05 vs SHR group), and urea (68.4% p<0.0001 vs SHR group); increases in urinary excretion of creatinine (37.3%, p<0.05 vs SHR group) and urea (49.1%, p<0.05 vs SHR group); and increases in creatinine (2fold increase, p<0.05 vs SHR group) and urea clearance (59.5%, p<0.01 vs SHR group). The results evidence that acCGP@OE was able to recover the glomerular filtration in SH rats. Additionally, the treatment with acCGP@OE led to the reduction in the TBARS levels (13.5%, p<0.05 vs SHR group) and increases in tissue. Altogether, these results show that acCGP@OE are promising treatment against the nephropathy associated with hypertension.

**Keywords:** *d*-limonene; cashew gum; glomerular filtration; nephopathy.



Title	Antihypertensive and cardioprotective effects of nanoparticles loaded with essential oil from <i>Citrus sinensis</i>
Authors	Cassio Nazareno Silva da Silva¹; Juliana Vila Verde Ribeiro¹; Guilherme Ananias Cardoso¹; Isac Cardoso dos Santos¹; Júlio César Lima Silva¹; Kátia Flávia Fernandes¹; Pâmela Yasmin de Oliveira Ferreira¹; Paulo César Ghedini¹; Carlos Henrique Xavier Custódio¹; Carla de Aleluia Batista²
Affiliations	<sup>1</sup> Universidade Federal de Goiás, Goiânia, Goiás <sup>2</sup> Instituto Federal de Goiás, Goiânia, Goiás
Session	Biologia e Doenças Cardiovasculares

## Abstract and Keywords

Considering that the essential oil from C.sinensis is very volatile as well as is affected by temperature, sunlight, moisture, and pH alterations, alternative technologies must be employed to preserve its biological potential. In this study, nanoparticles of acetylated cashew gum polysaccharide (acCGP) were used as carriers for EO. The produced acGCP@EO nanoparticles were characterized by TEM, XDR, DSC and TG and their antihypertensive and cardioprotective effects were evaluated in spontaneously hypertensive rats (CEUA-UFG nº049/22). Wistar (normotensive control) and SH rats were treated daily with EO or acCGP@EO for 30 days, in a dose of 75 mg.Kg-1. The systolic blood pressure was recorder during the treatment and the metabolic parameters were accessed at the end of the treatment. The data were analyzed by two-way ANOVA followed by Tukey's post hoc test. acCGP@EO nanoparticles presented a spherical morphology with size about 161.1 nm and Zeta Potential of -19.1 mV. Results from XDR, DSC and TG revealed a good compatibility between acCGP and the EO during the nanoparticle formation. The essential oil and the nanoparticles showed an antihypertensive effect, being observed a delta of reduction about 51 mmHg for EO and 42 mmHg for acCGP@EO (p<0.05, vs SHR group). Regarding the metabolic parameters, it was found that treatment did not interfere with food intake, water ingestion and feces content, neither interfered with the content of serum and urinary protein content and the sodium concentration at plasma (p>0.05 vs WS and SHR groups). However, the content of urinary sodium was reduced (23.41% for OE; 20.92% for acCGP@OE, p<0.05), indicating a possible amelioration in the natriuretic behavior of SH rats. Additionally, it was observed a reduction in the TBARS levels (26.06%, p<0.05) and an increase in the activity of superoxide dismutase enzyme on cardiac tissue (58.82%, p<0.05) in the SH rats treated with acCGP@EO. Altogether, results indicated that acCGP@EO has evoke potent antihypertensive and cardioprotective effects.

**Keywords:** Cashew gum; Blood pressure; TBARS; superoxide dismutase.



Title	Arterial and intracreanial pressure changes induce by intracerebroventricular infusion in renovascular hypertensive rats renovascular hypertension
Authors	<sup>1</sup> Fernanda Campos de Medeiros; <sup>1</sup> Pedro Lourenço Katayama; <sup>2</sup> Rodrigo de Alburquerque Pacheco Andrade; <sup>1</sup> Eduardo Colombari; <sup>1</sup> Debora Simões de Almeida Colombari
Affiliations	¹School of Dentistry- UNESP, Araraquara, Brazil; ²Brain4Care
Session	Biology and Cardiovascular Diseases

## Abstract and Keywords

Renovascular hypertension is a secondary hypertension, which has an identifiable cause. Two-kidneys-1-clip (2K1C) rat model of renovascular hypertensionon, present increases in arterial and intracranial pressure (AP and ICP, respectively) and changes in the morphology of ICP wave. Therefore, the aim of the present study was to compare the effect of intracerebroventricular (ICV) saline infusion in normotensive and 2K1C rats. Male Hotlzman rats (250-280 g) were randomly separated to be sham or with 2K1C, which was done by partial left renal artery occlusion. Six weeks after surgery, the animals were anesthetized and had the femoral vein and artery cannulated. In the following day, the animals had their basal blood pressure recorded and subsequently received intravenous anesthesia with urethane (1.2g/kg), were tracheostomized, placed under artificial ventilation and receive the ICP sensor implant and a needle for infusion into the right lateral ventricle (100 µL/min for 10 minutes). From the analysis of the recordings, it was observed that there are significant differences between normotensive and hypertensive animals. The initial mean compliance for normotensive animals is  $42.43 \,\mu\text{L/mmHg}$  and  $4.85 \,\mu\text{L/mmHg}$  for 2K1C, and the differences remain during infusion. Throughout, the average maximum ICP is 40.5mmHg for normotensives and 114.8 for 2K1C animals. There are also differences in the mean P2/P1 ratio during the infusion, which varies from 0.6466 and 9.391 for normotensive rats and from 1,50 to 2.1684 for hypertensive rats. Finally, the wave amplitude of the ICP also varies between groups, being 2.1556 for normotensive and 9.3274 for hypertensive. All results considered relevant took P<0.05 into account. Our findings showed that in renovascular hypertension there are changes in the intracranial pressure pattern, including its wave morphology during icv infusion.

**Keywords:** intracranial pressure; hypertension; brain compliance; wave morphology; challenge of increased intracranial pressure.

CEUA: 16/2023



Title	Effects of polyphenols-enriched fraction from fruit peel of <i>Annona crassiflora</i> Mart. on oxidation and glycation of human low density lipoprotein (LDL)
Authors	Nycollas Bruno Oliveira Fernandes¹; Douglas Carvalho Caixeta²; Allisson Benatti Justino¹; Robinson Sabino Silva²; Foued Salmen Espindola¹; Françoise Vasconcelos Botelho¹
Affiliations	<sup>1</sup> Institute of Biotechnology, UFU, Brazil <sup>2</sup> PGP Health Sciences, Faculty of Medicine, UFU, Brazil.
Session	Biologia e Doenças Cardiovasculares

# and Keywords

Reactions involving LDL oxidation and glycation prevent LDL receptor-mediated uptake and promote the uptake of lipoprotein modified by scavenger receptors in macrophages leading to the formation and progression of atherosclerosis. Annona crassiflora Mart. it is a plant widely used in folk medicine with several clinical properties, among them are protection against oxidative stress, inflammation and glycation. The aim of our study was to evaluate the effect of crude extract and ethyl acetate fraction of Annona crassiflora Mart fruit peel on oxidation and glycation of LDL in vitro. Its oxidation was monitored through the formation of conjugated dienes at 234nm, production of thiobarbituric acid reactive species (TBARS), fluorescence analysis of tryptophan residues and through structural changes by infrared spectroscopy. For the glycation assays, LDL was incubated with methylglyoxal and different concentrations of the crude extract of the ethyl acetate fraction of Annona crassiflora Mart. The fluorescence intensity, as a measure of LDL glycation products, was analyzed at 340nm excitation and 380nm of emission and the results were expressed in fluorescence units. The results showed that the fruit peel extract and its fraction slowed or inhibited the oxidation of LDL induced by Cu II ions in vitro. Fourier Transform Infrared Spectroscopy (FTIR) revealed that specific regions of Apolipoprotein B-100 that have been modified and treatments reduced such changes. However, the increase in the levels of advanced glycation products observed in LDL glycated by methylglyoxal were not reversed by the crude extract or by the ethyl acetate fraction, only by quercetin. Whereas treatments with Annona crassiflora Mart. extracts slowed or inhibited LDL oxidation in vitro, it is interesting to evaluate the effects on the development of atherosclerotic lesions.

**Keywords**: Atherosclerosis, LDL, oxidation, glycation and *Annona crassiflora* Mart



Title	Possible mechanism of arterial pressure maintenance in obese ovariectomized female rats
Authors	Helena de Oliveira Deróbio; Sara de Moraes Leite; Gabriela Maria Lucera; José Vanderlei Menani; Eduardo Colombari; Debora Simões de Almeida Colombari.
Affiliations	School of Dentistry, Araraquara, São Paulo State University
Session	Biology and Cardiovascular Diseases

## and Keywords

Obesity is a major health problem and affects men and woman. We have demonstrated before that female obese rats (ovary intact) did not have a difference in baseline arterial pressure compared to euthrophic female. However, we still do not know if obesity will cause change in baseline arterial pressure in ovariectomized obese rats and if the estrogen replacement will do any change. Holtzman female rats (280 - 300 g) were fed with standard diet (SD, 11% calories from fat) or high fat diet (HFD, 45% calories from fat) for 6 weeks. In the same week of the commecing of the diets regiment, the rats were bilaterally ovariectomized (OVX) and at the end of the 6th week, 17-□estradiol (40 □g of body weight, sc) was injected for 7 days. The groups were then: SD-OVX (n = 4), SD-OVX/E2 (n = 3), HFD-OVX (n = 5), HFD-OVX/E2 (n = 3), HFD-OV =3). In the 7th day, the femoral artery and vein were canulated for mean arterial pressure (MAP) recording and drugs injection. In the next day, in freely moving rats, we observed that baseline MAP was not different between all groups (SD-OVX; SD-OVX+E2; HFD-OVX; HFD-OVX+E2; respectively: 119 ± 5;  $125 \pm 10$ ;  $123 \pm 2$  and  $118 \pm$ 

0,2 mmHg). After 30 min, losartan administration (10 mg/kg, iv) produced very sutle changes in MAP in all group ( $\square$  MAP, -6 ± 2; -7 ± 9; - 7 ± 2 and -7 ± 2, respecvively). After 30 min of losartan injection, hexametonium (20 mg/kg, iv) produced a substantial decrease in MAP, which was also similar between groups ( $\square$  MAP, -61 ± 5, -68± 8 , -64 ± 4, -69 ± 4 mmHg, respectively). From the preliminary data, it seems that at euhydrated conditions, there is no difference in baseline and in the mechanisms controlling MAP in euthrophic and obese OVX female rats. We will next challenge these females to a water deprivation that increases the renin-angiotensin system (RAS). Since RAS is overactiveated in obese rats, a difference in controlling MAP under this circumstance might appear.

**Keywords:** obesity, female, menopause, angiotensin II, sympathetic nervous system.

CEUA: 10/2023



Title	Antimicrobial effect of MWCNTs in the poultry industry
Authors	Arthur Franco Demétrio; Phelipe Augusto Borba Martins Peres; Natieli Saito; Paula Cristina Batista de Faria.
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brasil.
Session	Biologia e Doenças Gatrointestinais.

## Abstract and Keywords

Poultry farming is substantial for Brazil's economy, as the country is the largest exporter and second-largest chicken meat producer globally. However, zoonotic species such as *Escherichia coli*, *Salmonella* spp, and *Campylobacter* spp pose significant health risks, forming biofilms in industrial environments and causing gastrointestinal illnesses worldwide.

Nanoparticles have gained attention due to their potential applications in various fields, including food packaging, water treatment, sanitizing solutions, etc., due to their antimicrobial activity. This pilot study aims to evaluate the bactericidal potential of the multi-walled carbon nanotubes (MWCNTs) against a reference strain of *E. coli* (ATCC25922).

The microorganisms were refreshed in Mueller Hilton Broth, then the cultures were subjected to a minimum inhibitory concentration (MIC) assay using MWCNTs to inhibit bacterial growth. Both plates containing the experimental samples and the positive and negative controls, were incubated at 37°C for 24h, one under agitation (240 rpm) and the other at rest. After the incubation period, resazurin assay was used to monitor the viability of bacteria exposed to different concentrations of MWCNTs. One-way ANOVA was used to compare cell viability between treatment and control groups.

Under agitation, E. coli viability was only 10,64% at the highest concentration (1mg/ml) of MWCNTs, representing an 89,36% (p< 0,0001) reduction when compared to the positive control. In the plate that remained at rest, we observed a smaller (45,62%), but still significant (p< 0,0001), reduction in cell viability compared to the control.

These preliminary results show an interesting antimicrobial activity of carbon nanotubes, indicating their potential use in synergy with sanitizers, which will be tested in future stages of these studies, and could represent an important tool in reducing the incidence of foodborne diseases.

**Keywords:** Gastrointestinal illnesses, public health, nanobiotechnology.



Title	Synergistic effects of caffeine and universal tyrosine kinase inhibitor Sorafenib on hepatocellular carcinoma C3A/LX2 spheroids
Authors	Luana Riechelmann Casarin¹; Letícia Cardoso Valente¹; Julia Stocco da Silva¹; Luís Fernando Barbisan²; Guilherme Ribeiro Romualdo¹.
Affiliations	<sup>1</sup> Experimental Research Unit (UNIPEX), Botucatu Medical School, São Paulo State University(UNESP), Brazil. <sup>2</sup> Department of Structural and Functional Biology Botucatu Biosciences Institute, São Paulo State University (UNESP), Brazil
Session	Poster session 1

#### Abstract and Keywords

Hepatocellular Carcinoma (HCC) represents a significant global epidemiological challenge. Despite advancements in management through systemic therapies such as Sorafenib (SOR), clinical outcomes remain modest. Thus, exploring bioactive compounds that can enhance the antitumor effects of therapies is encouraged. In this sense, the alkaloid caffeine (CAF) – available in coffee beans and derived beverages - had already showed beneficial properties in in vitro models of HCC, and chemopreventive effects on chemically-induced models of hepatocarcinogenesis, including data of our own group. Then, this in vitro study evaluated if caffeine (CAF) can enhance the antitumor response of SOR. Human LX2 hepatic stellate cells, C3A HCC cells (two-dimensional - 2D), and C3A/LX2 spheroids (tridimensional - 3D) were individually treated with CAF and SOR for 24 and 48 hours. The viability assay (MTT) was performed to determine the half- maximal effective concentration (EC50) for SOR and CAF, individually. Data were analyzed by one-way ANOVA followed by Tukey's test and the Combinatory Index (CI) was calculated using CompuSyn software. Then, spheroids were treated with the combination of SOR+CAF, with concentrations at 1/5 and 1/10 of the EC50. Due to increased model complexity, spheroids held increased IC50 values for SOR (24h:4.1±1.7mM, R<sup>2</sup>:0.77; 48h:391±117μM, R<sup>2</sup>:0.74) or CAF (24h:38±21mM, R<sup>2</sup>:0.50; 48h:31±00mM, R<sup>2</sup>:0.84) compared to C3A (SOR:33.0 $\pm$ 0.0 $\mu$ M, R<sup>2</sup>:0.90; 48h, 13.0 $\pm$ 0.0 $\mu$ M, R<sup>2</sup>:0.93; CAF:24h, 10.0±0.0mM, R<sup>2</sup>:0.84; 48h, 2.2±0.0mM, R<sup>2</sup>:0.89) or LX2 monolayers  $(SOR:24h; 11.0\pm4.2\mu M, R^2:0.84-0.91; 48h; 0.08\pm0.10\mu M, R^2:0.89; CAF:24h;$ 7.5±2.1mM, R<sup>2</sup>:0.85; 48h; 7.9±2.6mM, R<sup>2</sup>:0.90). The SOR+CAF combination in 1/5 (6200 $\mu$ M+78 $\mu$ M) and 1/10 IC50 (3100 $\mu$ M+39 $\mu$ M) promoted the effects of CAF or SOR alone in 24h (1/5 IC50:67.1±1.32 vs antitumoral 87.8±7.09 vs 89.3±5.01 cell viability, p=0.0002; 1/10 IC50: 76.2±6.96 vs 86.2±4.59 vs 87.2±8.52 cell viability, p=0.024) and 48h (1/5 IC50: 34.7±0.00 vs 79.7±1.53 vs 50.3±4.12 cell viability, p<0.0001; 1/10 IC50: 54.8±6.10 vs  $89.7\pm2.03$  vs  $81.0\pm5.25$  cell viability, p<0.0001), promoting the decrease in the cell viability of C3A/LX2 spheroids. The CI analysis revealed a synergistic effect (CI<1) of CAF+SOR combination in 48h for both 1/5 (CI=0.94) and 1/10 (CI=0.74) IC50 combinations. The data indicate that CAF+SOR combination synergistically promotes SOR antitumoral response in HCC, and future clinical studies should be considered.

**Keywords:** Caffeine, Sorafenib, HCC, Synergistic effects



Title	Synergistic effects of caffeine and Atezolizumab plus Bevacizumab immunotherapy on hepatocellular carcinoma C3A/LX2 spheroids
Authors	Leticia Cardoso Valente¹; Luana Riechelmann Casari¹; Julia Stocco da Silva¹; Luís Fernando Barbisan²; Guilherme Ribeiro Romualdo¹.
Affiliations	<sup>1</sup> Experimental Research Unit (UNIPEX), Botucatu Medical School, São Paulo State University(UNESP), Brazil. <sup>2</sup> Department of Structural and Functional Biology, Botucatu Biosciences Institute,
	São Paulo State University (UNESP), Brazil
Session	Poster Session 1

# Abstract and

Hepatocellular Carcinoma (HCC) exhibits a high global incidence and mortality, along with an unfavorable prognosis. However, currently available treatments for advanced HCC, while promising, do not achieve entirely satisfactory results. Consequently, there is a growing impetus for innovative strategies, such as the development of therapies combining different bioactive food compounds with other treatments for advanced-stage HCC. In this context, the alkaloid caffeine (CAF) had already showed chemopreventive effects against HCC in vivo, also featuring antiproliferative, pro-apoptotic and anti-inflammatory properties in vitro. Thus, we assessed whether CAF enhances the antitumor response of immunotherapy with Atezolizumab (ATZ) and Bevacizumab (BVZ) in a threedimensional (3D) model. Human LX2 hepatic stellate cells and primary C3A HCC cells (3D) were seeded in ultra-low attachment U-shaped plates and individually treated with different concentrations of CAF, ATZ, and BVZ for 24 and 48h. Cell viability assay (MTT) was used to determine the half-maximal effective concentration (EC50) for CAF, ATZ, and BVZ, individually. Next, spheroids were treated with the combination of CAF+ATZ+BVZ, with concentrations at 1/6 and 1/10 of the EC50. Data were analyzed by one-way ANOVA followed by Tukey's test and the Combinatory Index (CI) was calculated using CompuSyn software. The immunotherapies (ATZ, 24h: 321±127μM, R2:0.89; 48h: 294±24μM,  $R^2:0.89$ ; BVZ, 24h:112±15 $\mu$ M,  $R^2:0.92$ ; 48h:94±22 $\mu$ M,  $R^2:0.83$ ) featured decreased IC50 values compared to CAF (24h:38±21mM, R2:0.50; 48h:31±00mM, R<sup>2</sup>:0.84) in C3A/LX2 spheroids. The CAF+ATZ+BVZ combination in 1/6 IC50 ( $5127\mu M + 49\mu M + 16\mu M$ ) increased the antitumoral effects of CAF or immunotherapy alone in both 24h (79.0±5.30 vs 92.9±0.42 vs 100.1±1.47 cell viability,p=0.0068) and 48h (70.9 $\pm$ 2.67 vs 82.1 $\pm$ 3.87 vs 82.0 $\pm$ 3.08 cell The CAF+ATZ+BVZ combination of 1/10 IC50 viability,p=0.0038).  $(3100\mu M + 29\mu M + 9\mu M)$  promoted the immunotherapy response only at 48h of exposure  $(74.11\pm4.40 \text{ vs } 92.9\pm2.65 \text{ vs } 89.6\pm2.68 \text{ cell viability,p}=0.0081)$ . The CI analysis revealed a synergistic effect (CI<1) of CAF+ATZ+BVZ combination in 48h for both 1/6 (CI=0.94) and 1/10 (CI=0.74) IC50 combinations. These preliminary findings indicate that CAF and immunotherapy combination synergistically promotes the immunotherapy response to HCC, and should be considered in further clinical studies.

**Keywords:** Caffeine, Atezolizumab, Bevacizumab, HCC, Synergistic effects



Title	Evaluation of urinary indole, and plasma d-ROMs and BAP test in cancer patients – A preliminary study
Authors	Eugenio Luigi Iorio <sup>2</sup> ; Clarissa Aires de Oliveira <sup>1,2</sup> ; Lara Ferreira Paraiso <sup>1</sup> ; Foued Salmen Espíndola <sup>1</sup>
Affiliations	<sup>1</sup> Institute of Biotechnology, Federal University of Uberlandia, Uberlandia, MG, Brazil.
	<sup>2</sup> International Observatory of Oxidative Stress. 84127, Salerno. Italy.
Session	Biologia e Doenças Gastrointestinais

## and Keywords

Cancer is a multifactorial condition characterized by inflammation and oxidative distress, an emerging health risk factor exerting deleterious effects on various organs and systems, including gut microbiota. The use of diagnostic kits serves as an invaluable tool in clinical settings, offering simplicity and reliability in disease detection and monitoring. Among these, indole test evaluates the concentration of indole in a urine sample. Indole is the product of tryptophanase, that catalyzes the breakdown of L-tryptophane to pyruvate and indole. The reduced bioavailability of L-tryptophane, can impair the gut-brain axis, with mood and/or sleep disturbances and possible loss of immune function. Moreover, dysbiosis may promote the entry of indoles in the plasma with possible unwanted systemic side-effects. Moreover, due the role of redox system in the relationships between gut bacteria, immune system and cancer, we can argue that indole metabolism can impact redox system and vice-versa. The present study was aimed to evaluate urine indole (UI) levels and oxidative stress (OS) in patients with different neoplasms. The UI concentration was measured by a kit based on a modified Kovac's reagent (reference value under 20 mg/L). The OS was assessed via d-ROMs test, normal range 250 - 300 Oxidising Units (O.U), and via BAP test, optimal value ≥2200 µmol/L on a capillary blood plasma sample. The study was retrospective and analyzed the database of thirty-two patients diagnosed with cancers, including breast (n=10), intestinal (n=6), prostate (n=3), and other malignancies (n=13), underwent examination. Results indicated a median indole concentration of 23.9 mg/L (interquartile deviation 19.5), with mean ( $\pm$  SD) d-ROMs and BAP test values of 475  $\pm$  122 O. U and  $2368 \pm 307 \mu mol/L$ , respectively. Preliminary data from the study revealed elevated levels of indole, which could potentially be linked to dysbiosis. Additionally, the high levels of d-ROMs and low levels of the BAP test confirmed a condition of oxidative distress.

**Keywords:** cancer, gut microbiota, indole, d-ROMs test, BAP test, oxidative stress.



Title	Effect of a leukotriene receptor antagonist in peptic ulcer
Authors	Wygny Araújo Macedo; Sheila da Silva Crethon; Yure Tavares Furtado; Gabriel Pires de Jesus; Maria Eduarda de Sousa Bento; Karla Graziella Moreira; Anderson Luiz-Ferreira.
Affiliations	Inflammatory Bowel Disease Research Laboratory, Department of Biological Sciences, Institute of Biotechnology, Federal University of Catalão (UFCAT), Catalão, Brazil
Session	Pôster

### Abstract and Keywords

Peptic ulcer (PU) is a common disease of the gastrointestinal tract that affects up to 20% of the world's population. Treatment is challenging due to the limited efficacy and serious adverse effects of the currently available drugs. Considering the high costs and the time needed to develop new drugs, drug repositioning, which consists of using drugs already established to treat other diseases, has been an alternative approach to speed up the process of validating new therapies. Seeking to provide drug alternatives for the treatment of PU, this work consists of the investigation of the gastroprotective effects of montelukast (MTK), a leukotriene D4 receptor antagonist. The gastroprotective action of MTK (5, 10, 15 mg/kg; p.o.) was evaluated in two experimental models using rats (ethanol and a non-steroidal anti-inflammatory drug - NSAID) mimicking the disease in humans. To elucidate the mechanisms of action, the involvement of sulfhydryl groups (G-SH) was also assessed in animals pretreated with a G-SH blocker. All experimental protocols were approved by the Animal Use Ethics Committee of the UFCAT (Protocol 04/21). Data are expressed as mean  $\pm$  S.D. of seven rats. In the ethanol-induced ulceration protocol, MTK (15 mg/kg, p.o.) promoted gastroprotection with 76% inhibition of the ulcerative lesion area (A.L.U.) compared to the negative control group (707.4  $\pm$  121.8 versus 168.2  $\pm$  145.7; p< 0.001). When ulcers were induced with a NSAID (indomethacin), MTK (15 mg/kg, p.o.) reduced the ALU by 59% compared to the negative control group  $(823.4 \pm 140.5 \text{ versus } 482.8 \pm 247.4; p < 0.01)$ . Previous administration of G-SH blocker significantly reduced the gastroprotection exerted by MTK, allowing us to infer the participation of this pathway in the gastroprotective activity of MTK (1473.0  $\pm$  277.3 *versus* 2590.0  $\pm$  177.8). The results point to the effectiveness of MTK in combating gastric ulcers, suggesting that MTK could be a good therapeutic agent for the treatment of gastric ulcers.

Keywords: montelukast, gastric ulcer, ethanol, gastric mucosa



Title	Mechanism of action of cannabidiol in neurodegenerative processes associated with aging: an integrative review
Authors	Marina Border Martim <sup>1</sup> ; Me. Aline Gavioli <sup>2</sup> ; PhD. Gilberto Laurentino <sup>2</sup> ; Dra. Sandra Regina Mota Ortiz <sup>2</sup> ; Ana Beatriz Cadeu <sup>1</sup>
Affiliations	<sup>1</sup> UNICID, São Paulo, Brazil <sup>2</sup> USJT, São Paulo, Brazil
Session	Neurobiology

### Abstract and Keywords

Alzheimer's Disease is characterized by the accumulation of  $\beta$ -amyloid plaques, a possible administration of cannabidiol may decrease the accumulation of  $\beta$ amyloid plaques and may improve neuroplasticity during aging. Cannabidiol extracted from Cannabis sativa (without psychoactive effects) has emerged as a possible strategy interacting with astrocytes, decreasing pro-inflammatory functions and studying neuronal cell death induced by β-amyloid, due to its ability to eliminate reactive oxygen species and reduce lipid peroxidation, which decreases the neurodegeneration process, being particularly interesting when considering the aging of the nervous system, an important serious alteration to neurodegeneration, present in diseases such as Parkinson's Disease and Alzheimer's Disease. In view of this possible form of treatment, the present study aimed to verify the mechanisms that cannabidiol lead to neuroplasticity in Alzheimer's disease. This integrative review was elaborated using specific descriptors defined according to "the PICOS search strategy", with the following descriptors: P (population) - Alzheimer's disease; I (intervention) - Use of Cannabidiol; C (comparator) Mechanism of action and neuroplasticity; plus, Boolean "AND" operators. After applying the inclusion/exclusion criteria and eligibility criteria, 14 articles were included in the study. There are 2 in vitro / in cyto studies, 3 molecular studies and 8 studies in transgenic animals. The results obtained show that the administration of cannabidiol has a positive role in neurodegenerative conditions, especially linked to the aging process, acting through pathways of inhibition of oxidative stress and inflammatory process, as well as the pathways of Wnt / b-catenin, GSK-3 $\beta$  and PC12.

**Keywords:** Cannabidiol; Alzheimer's; Neuroplasticity; Cannabinoid; Aging.



Title	MASLD-associated hepatocarcinogenesis in mice: influence of an obesogenic western diet on the tumoral metabolomic landscape
Authors	Gabriel P. Bacil <sup>1</sup> ; Maria Leticia O. Lyra <sup>1</sup> ; Keila N. Cavalcante <sup>2</sup> ; Daniel R. Cardoso <sup>2</sup> ; Guilherme R. Romualdo <sup>3</sup> ; Luís Fernando Barbisan <sup>1</sup>
Affiliations	<sup>1</sup> Department of Structural and Functional Biology – Biosciences Institute of Botucatu - UNESP, Botucatu, Brazil; <sup>2</sup> Department of Molecular Chemistry and Physics – São Carlos Institute of Chemistry - USP, São Carlos, Brazil; <sup>3</sup> Experimental Research Unit (UNIPEX) – Botucatu Medical School (FMB) - UNESP, Botucatu, Brazil.
Session	Poster session III.

#### Abstract and Sevwords

Hepatocellular carcinoma (HCC) is the 3rd deadliest cancer, and metabolic dysfunction-associated steatotic liver disease (MASLD) has become the major cause of chronic liver disease and HCC worldwide. Nevertheless, still urgent to unravel the molecular landscape of hepatocarcinogenesis by using preclinical models. Thus, this study assessed the tumoral metabolomic profile in a murine model of MASLD-associated hepatocarcinogenesis. Male C57BL/6J mice received intraperitoneal doses of diethylnitrosamine (DEN, 25 mg/Kg of body weight, 1x/week) or vehicle, for 4 weeks. At the 6th week of life, mice were fed a Western diet (WD, 30% fat and 0.2% cholesterol-added chow, and 45/55% glucose/fructose-water diluted solution) or basal diet, for 6 months (CEUA 1410/2022). Hepatic samples were collected for histopathological and collagen deposition analyses, and immunostaining for CD68, Ki67, and a-smooth muscle actin (a-SMA). Tumoral samples were collected for histopathological diagnosis and metabolomic profiling. WD-fed mice presented increased final body weight (p<0.0001), relative liver weight (p=0.0001), and adiposity index (p<0.0001). A higher tumoral multiplicity was observed in the WD+DEN group, when compared to the DEN group. Extensive micro/macrovesicular steatosis (p<0.0001), hepatocellular hypertrophy (p<0.0001), and lobular inflammation (p=0.0111) were observed in non-tumoral hepatic samples of WD-fed groups, which also featured increased hepatic collagen content (p=0.0011), α-SMA levels (p<0.0001), CD68+ cells (p<0.0001) and Ki67+ hepatocytes (p<0.0001). The tumoral metabolomic profile from WD-derived tumors presented higher glucose and lower ATP and lactate levels, when compared to DEN-derived tumors. The enrichment analysis targeted both aminoacyl-tRNA and valine, leucine, and isoleucine biosynthesis as drivers of MASLD-associated hepatocarcinogenesis. Thus, the WD-induced milieu modulates hepatocarcinogenesis by modifying the tumoral metabolomic profile in mice.

**Keywords:** MASLD; hepatocarcinogenesis; Western diet; metabolomics.



Title	Optimization of fluorescence assay for detection of Advance glycation end products (ages) and evaluation of antiglycoxidation agents and their efficacy
Authors	Tarcisio Paiva Mendonça; Rener Mateus Francisco Duarte; Vinicius Prado Bittar; Ana Luiza Silva Borges; Pedro Figueiredo Freitas Costa; Otávio Silveira Rizzi; Annelise Arantes Rocha; Renata Roland Teixeira; Foued Salmen Espindola.
Affiliations	Institute of Biotechnology – Federal University of Uberlandia, Uberlandia, MG 38400-319
Session	6. Endocrine system - biochemistry

#### Abstract and Keywords

Fluorescent advanced glycation end products (fAGEs) are linked to chronic diseases like diabetes. This study aims to assess the glycoxidation of proteins caused by fAGEs, focusing on some of the wavelengths of emission and excitation (em/ex) fluorescence like Pentosidine (335/385nm) and Argpyrimidine (335/385nm). Bovine Serum Albumin (BSA) in the presence of Methylglyoxal (MGO) and Fructose (glycating agents) were incubated, adding the following antiglycants, Aminoguanidine (NH-G) (positive control) and the antioxidants Nacetylcysteine (NAC) and Quercetin (QUERC). The incubation was carried out in NaN3 (Azide) presence at 37C for 3, 7, and 14 days (3d, 7d, and 14d) and read in a fluorimeter. For Pentosidines reads caused by Fructose, NH-G inhibited by 88% (d3), 92%(d7), and 97%(d14), while NAC showed reductions of 75% (d3), 74%(d7), and 58% (d14). QUERC inhibited 100% over the three days. For Pentosidines with MGO, NH-G achieved 98% (d3), 99% (d3 and d14), and NAC 98% (d3), 99% (d7 and d14), with QUERC maintaining 100%. Regarding Argpyrimidine reads, NH-G inhibited by 89% (d3), 96% (d7), and 98% (d14), NAC recorded percentages of 72% (d3), 73% (d7) and 55% (d14), while QUERC remained at 100% over the three days. With MGO associated with Argpyrimidine, NH-G and NAC achieved inhibitions of 97% (d3), 99% (d7) and 99 (d14), and QUERC maintained 100%. Results confirm the consistent effectiveness of the assay. β-amyloid inhibition detection by Thioflavin T-test showed for MGO, NAC, and NH-G, inhibitions were 63% and 70%, respectively, after 14 days, while QUERC demonstrated superior efficacy with 66% (d14). These results showed the consistent effectiveness of the anti-fAGEs antioxidant compounds NAC and QUERC. Furthermore, Quercetin, a main flavonoid in many vegetables, including those used as foods, showed its potential to inhibit fAGEs and the consequent protein aggregation, highlighting its role as an adjuvant therapeutic approach for chronic diseases like diabetes.

**Keywords:** Advanced glycation end products (AGEs); Fluorescence assay; Antiglycating agents; Glycoxidation; Protein aggregation inhibition.



Title	Intermittent fasting modulates kidney function in female rats
Authors	Arthur Fornazari Iost <sup>1</sup> ; Olívia Ramos Costa Val Pedrosa <sup>1</sup> ; Leonardo Oliveira Guarnieri <sup>1</sup> ; Maria Aparecida Ribeiro Vieira <sup>1</sup> ; Diogo Barros Peruchetti <sup>1,2</sup>
Affiliations	<sup>1</sup> Department of Physiology and Biophysics, Institute of Biological Sciences, Federal University of Minas Gerais, Belo Horizonte, Brazil <sup>2</sup> INCT-Nanobiofar, Belo Horizonte, Brazil
Session	Nutrição e Metabolismo

## and Keywords

Dietary restrictions have been gaining interest among large populations worldwide. In particular, some reports have shown the beneficial effects of intermittent fasting (IF) protocols on different neurological and cardiovascular diseases. However, the effects on the kidneys are still poorly understood. We aimed to verify the potential impact of IF on renal function. Female Wistar rats (8-10 weeks old) were randomly divided into two experimental groups: 1) CONT (control group, n=4) – with free access to chow; and 2) IF (intermittent fasting group, n=5) - rats subjected to the IF protocol [fasting: 18h (from "zeitgeber time" (ZT)=8 up to ZT=24)/ feeding: 6h (from ZT=1 up to ZT=7)] for 4 weeks. During the feeding period, the rats had free access to chow (CEUA-UFMG#13-2023). Once a week, the rats were housed in metabolic cages (during the feeding period) to measure chow and water intakes, as well as to collect urine for biochemical analysis. Compared to the CONT group, the IF group showed a reduction in body weight (BW, 15%), which was observed despite an increase in water intake (3.0-fold) and food intake (6.0-fold). The IF group also presented a reduction in urinary volume (64%, mL/6h/kg BW) after 2 weeks of diet restriction. No significant alterations were observed in urinary creatinine concentrations. Interestingly, urinary glucose mass was reduced (44%) in the IF group. However, proteinuria and UPCr (the ratio between urinary proteins and creatinine) were increased (1.8-fold and 1.6-fold, respectively). A positive Pearson correlation was found between BW and  $(r=0.4906/R^2=0.2407/P=0.001)$ , glycosuria while an inverse Pearson correlation found between BW and was proteinuria  $0.7169/R^2=0.5140/P<0.0001$ ). Our preliminary findings suggest that the 4week IF protocol modulates renal function, especially the mechanisms involved in renal proteins and glucose handling.

Keywords: intermittent fasting, kidney, glycosuria, proteinuria



Title	Chemical characteristics of synthetic and plant-based drugs from a magistrative formula used in the treatment of obesity
Authors	Nathalia Cristina Gouvêa de Souza; Luciana Karen Calábria
Affiliations	Universidade Federal de Uberlândia - Campus Pontal, Ituiutaba - MG, Brasil
Session	Nutrição e Metabolismo

#### Abstract and Keywords

Obesity is a multifactorial chronic disease that may require drug therapy. Magistrative formulas, with mixed composition, have been used for weight loss, but have been causing health problems. This exploratory and descriptive study provides a survey of the main chemical characteristics of drugs (fenproporex, amitriptyline, fluoxetine, sibutramine, furosemide, cascara and senna) found in a formula recommended for weight loss by local doctors. Its clinical applications, pharmacokinetics and pharmacodynamics, doses, adverse effects and interactions were described. For literature review used PubMed, SciELO, Periódicos Capes and Google Scholar; official documents from ANVISA and manufacturers; and PubChem and DrugBank. The data revealed the potential of each of the drugs, however, it is highlighted how unsafe it is to combine all drugs in a single formula due to interactions and physiological damage, considering that sibutramine, furosemide, fenproporex and cascara have more than 1600, 1500, 700 and 390 interactions, respectively. These interactions can result in the most different physiological damages. It is important to highlight that, although only one active metabolite of each herbal medicine has been found with antiobesity action, scientific information about the other active ingredients present in these drugs has not been completely elucidated. There is a need to raise awareness among health professionals in the technical and specialized dispensing of formulas, and the promotion of actions to sensibilizing the population about safe use and only under medical guidance.

**Keywords:** Weight loss; Drug interaction; Pharmacological treatment.



Title	Mimetic peptides of beta-casein proteins A1 and A2 for diagnosis in bovine milk
Authors	Phelipe Augusto Borba Martins; Peres; Fabiana de Almeida Araújo Santos; Emilia Rezende Vaz; Arthur Franco Demétrio; Natieli Saito; Vivian Alonso- Goulart.
Affiliations	Federal University of Uberlandia (UFU), Uberlândia, Minas Gerais
Session	Nutrition and Metabolism

### Abstract and Keywords

Cow's milk is a globally consumed food rich in essential nutrients. One of its protein components is beta-caseins, which vary according to the genotype of the animals. The A1 and A2 types of beta-casein differ by a single amino acid at position 67. Type A1, in gastric digestion, releases an opioid (BCM-7) described in cases of lactose intolerance, heart problems, diabetes, and even autism in children. In opposite of that, type A2 milk has become the focus of dairy production, to quarantee quality and food safety. Detecting such specific fragments of proteins makes it difficult to develop rapid and cost-effective field tests. Therefore, this study aimed to develop mimetic peptides of A1 and A2 beta-caseins as an experimental model for the development of diagnostic platforms in the field. Phage Display technology was applied on commercial anti-A1 and anti-A2 beta-casein antibodies, for two biopanning in parallel, followed by incubation with the phage library with 7aa peptides, washing steps, and negative selections using non-binding agents. An antibody against the reverse was utilized, and ligands were collected by competitive elution with the recombinant target protein. Subsequent steps of selection, amplification, DNA extraction, and sequencing were made, and validations were carried out using the ELISA technique. After analyzing the ELISA results and sequence alignments with the recombinant protein, the following clones were selected: 01 clone (A) mimetic of A1 beta-casein with a ratio of 2.24 and 02 clones (D and E) mimetic of beta-casein A2 casein with a ratio of 2.73 and 3.26, respectively. These selected clones can efficiently be used as targets for the differentiation of A1 and A2 beta-caseins, enabling the progression of other studies aimed at the development of commercial products with high global relevance in veterinary medicine and public health.

**Keywords:** Phage Display, genotyping, milk, BCM-7.



Title	To evaluate the hypoglycemic effect of bioactive peptides from common bean (PV3) in mice submitted to a high-fat diet
Authors	Barros, C <sup>1</sup> ; Martins, J <sup>1</sup> ; Torsoni, M <sup>2</sup> ; Chaves, W <sup>2</sup> ; Lopes, P <sup>2</sup> ; Costa, C <sup>2</sup> ; Ribeiro, J <sup>1</sup> ; Xavier, C <sup>1</sup> ; Batista, K <sup>3</sup>
Affiliations	<sup>1</sup> Universidade Federal de Goiás; <sup>2</sup> Unicamp- Campus Limeira; <sup>3</sup> Instituto Federal de Goiás
Session	7- Nutrition and Metabolism

# and Keywords

The number of cases of people with diabetes, obesity and cardiovascular diseases is increasing every day, due to the lifestyle of society Bean peptides come as a way out for the treatment or prevention of cardiometabolic diseases. This aimed study to evaluate a hypoglycemic effect of hard-to-cook bean peptides in C57 BL/6 mice. The present study carried out chronic treatment in C57 BL/6 mice and administered daily doses of hardened common bean peptides (called PV3), through pseudo-gavage for 60 days, and the experimental protocol was approved by CEUA (5857-1/2021). Thus, 4 groups were obtained: standard diet (SD), SD + PV3, HFD, and HFD + PV3. In these groups were measured: Body weight weekly, Fasting blood glucose in the 4th and 9th week using a glucometer, Glucose, Insulin tolerance. The animals were euthanized, and your liver was dissected for western blothing analysis. The results show that PV3 prevented obsessiveness in animals with a high-fat diet, reduced blood glucose in animals treated with PV3 as can be seen in glucose and insulin tolerance tests, and in WB the results show that it increased insulin receptor expression. Thus, we can conclude that PV3 is an excellent alternative for the prevention of cardiometabolic diseases.

**Keywords:** PV3, diabetes, hypoglycemic, biological activity, bioactive peptides.



Title	Influence of social jet lag in food intake in bariatric patients: a one-year follow-up study
Authors	Aline Cunha Carvalho <sup>1,2</sup> , Luisa Pereira Marot <sup>1</sup> , Luiz Augusto Mattar <sup>2</sup> , José Américo Gomides de Souza <sup>2</sup> , Ana Cristina Tomáz Araújo <sup>2</sup> , Camila Thais da Costa Assis <sup>3</sup> , Maria Carliana Mota <sup>1</sup> , Cibele Aparecida Crispim <sup>1</sup>
Affiliations	<ul> <li>Chrononutrition Research Group, Faculty of Medicine, Federal University of Uberlandia</li> <li>LEV Clinic – Treatment of Obesity, Uberlandia, Brazil,</li> <li>Triangulo Mineiro university Center</li> </ul>
Session	Oral presentation

#### Abstract and Keywords

Circadian misalignment indicates a failure in the synchronization between endogenous factors (central and peripheral clocks) and environmental signals, causing changes in the physiological circadian rhythm. Social jet lag (SJL) is a measure that indicates the degree of this misalignment which has been associated with an increased risk of overweight and obesity, as well as changes in food consumption. However, the scientific literature is still scarce studies with these associations in patients undergoing bariatric surgery.

#### Objective

Evaluate the association between SJL with food consumption outcomes one year after bariatric surgery.

#### **Methods**

A total of 122 bariatric patients were included (77% female, aged 33 years (range 28 - 41); 79.5% underwent Roux-en-Y gastric bypass). Food consumption and SJL were evaluated in the preoperative evaluation and in the third and sixth months and one year after surgery. SJL was calculated based on the absolute difference between the mid-sleep time on weekends and weekdays. Food consumption was evaluated through the application of 2 dietary recalls 24H in each assessment according to the multiple pass method (Totalling - 976 Recalls). Generalised estimating equations were performed to evaluate effect of time, isolated effect of SJL and the interaction of these variables in food consumption during first year of surgery.

#### Results

The group more exposed of SJL (Large SJL) had higher intakes of calories (p = 0.001), carbohydrate (p = 0.038), proteins (p= 0.001) and total (p = 0.002) and polyunsaturated (p = 0.011) fat when compared with the group less exposed of SJL (Small SJL).

#### Conclusions

SJL was negatively associated with food consumption outcomes 1 year after bariatric surgery. Future studies with longer follow-up are needed to confirm these findings.

**Keywords:** Bariatric surgery, Food consumption, Social jet lag



Title	Association between plasma omega-3 fatty acids and muscle strength according to protein intake: NHANES 2011 - 2012
Authors	Rafaela Nehme¹; Larissa S. Limirio¹; Jaqueline L. Pereira ²; Flavia M.S. de Branco¹; Erick P. de Oliveira ¹
Affiliations	<sup>1</sup> Laboratory of Nutrition, Exercise and Health (LaNES), School of Medicine, Federal University of Uberlandia (UFU), Uberlandia, Minas Gerais, Brazil;
	<sup>2</sup> Department of Nutrition, School of Public Health, University of São Paulo (USP),
	São Paulo, SP, Brazil
Session	Oral

# and Keywords

Muscle strength is an important predictor of quality of life and mortality. Several studies have evaluated the association between omega-3 ( $\omega$ -3) intake and muscle strength, but the evidence is still limited. In addition, no study has investigated whether this association is dependent on protein intake. The aim of the study was to evaluate the association between plasma  $\omega$ -3 and muscle strength according to protein intake. A cross-sectional study was performed evaluating 1708 individuals aged between 20-80 years old from the National Health and Nutrition Examination Survey (NHANES) 2011-2012. Total plasma  $\omega$ -3 fatty acids and their subtypes (alpha-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA)) were evaluated using chromatographymass spectrometry. Muscle strength was evaluated through handgrip strength, with the combined grip strength of both hands considered. Food intake was assessed through two 24-hour recalls. All the procedures was approved by Ethics Review Board (ERB) (protocol #2011-17). Linear regression analysis was used to evaluate the association between plasma ω-3 fatty acids and muscle strength according to protein intake (< 0.8g/kg;  $\ge 0.8 - < 1.2g/kg$ ;  $\ge 1.2 g/kg$ ) adjusting for potential confounders (energy intake, alcohol intake, waist circumference, smoking, familiar annual income, race, arthritis, physical activity, age and sex). These analyses were conducted for the entire sample, as well as stratified by age groups (< 60 years old;  $\geq$  60 years old). For total sample, total plasma  $\omega$ -3 (< 0.8g/kg:  $\beta$ =-0.0008; p-value= 0.834;  $\geq 0.8 - < 1.2 g/kg/d$ :  $\beta$ = 0.003; p-value= 0.507;  $\geq 1.2$  g/kg/d:  $\beta$ = -0.002; p-value= 0.949) ALA, EPA, and DHA were not associated with muscle strength. The lack of association persisted even after stratification by age group. Plasma  $\omega$ -3 fatty acids were not associated with muscle strength in older and young people regardless of protein intake.

**Keywords:** ω-3, Muscle strength, Protein



Title	Handgrip strength is not correlated with the five times sit-to- stand test in kidney transplant patients
Authors	Maria Eduarda de F. Mendonça, Giovanna de M. Ribeiro, Rafaela Nehme, Débora M. Morais, Larissa S. Limirio, Erick P. de Oliveira
Affiliations	Laboratory of Nutrition, Exercise and Health (LaNES), School of Medicine, Federal University of Uberlandia (UFU), Uberlandia, Minas Gerais 38400-902, Brazil
Session	Poster presentation

## Abstract and Keywords

Muscle strength is an important predictor of quality of life and low muscle strenght is associated with negative outcomes and clinical conditions, such as risk of falls, sarcopenia and mortality. The European Working Group on Sarcopenia in Older People (EWGSOP) determines the strength as the first component to diagnose sarcopenia. To measure muscle strength the consensus suggests some tests such as handgrip strength and the five-times-sit-to-stand test. However, to the best of our knowledge, no study has evaluated whether these tests are correlated in clinical populations, such as individuals with renal disease. Therefore, our aim was to evaluate the correlation between handgrip strength and the five-times-sit-to-stand test in kidney transplant patients. A cross-sectional study was performed evaluating 127 kidney transplant patients. Muscle strength was evaluated using handgrip strength (HGS) and five times sit to stand (5STS). A simple and partial Pearson correlation was performed in two statistical models. Model 1 was unadjusted for confounders. Model 2 was adjusted for sex, age and body mass index (BMI). P value <0.05 was considered statistically significant. The study protocol was approved by the Human Research Ethics Committees of the Federal University of Uberlandia (number: 1688246). All subjects signed an informed consent term. No correlation was found between the handgrip strength and the five-times-sit-to-stand test in any of models (Model 1: r= 0.093 p= 0.295; Model 2: r=0.035; p=0.693). In conclusion, our findings indicate a lack of correlation between the hand grip strength test and the five-times-sit-to-stand test in kidney transplant patients.

**Keywords:** Stength; Kidney transplant; Handgrip; Five times sit to stand test;



Title	Chrononutritional behaviors: a longer eating duration is not necessarily related to a higher daily caloric intake. Data from NHANES 2015-2018
Authors	Nayara Bernardes da Cunha; Gabriela Pereira Teixeira; Catarina Machado Azeredo; Ana Elisa Madalena Rinaldi; Cibele Aparecida Crispim.
Affiliations	Graduate Program in Health Sciences, Faculty of Medicine, Federal University of Uberlândia, Minas Gerais, Brazil.
Session	Nutrição e Metabolismo

## Abstract and

Chrononutrition, which bridges the circadian system with metabolism and nutrition, has emerged as a promising avenue for enhancing obesity management strategies. Some studies have linked eating within a longer window and, consequently, at later times, with higher caloric intake. The aim of the present study was to investigate the association of chrononutrition variables related to the eating duration (ED) and meal timing with energy intake in American adults and elderly (NHANES 2015-2018). A total of 5,777 adults and elderly individuals were included in the analysis. Meal timing data were collected through two 24-hour dietary recalls in both cycles (2015-2016 and 2017-2018). Multiple linear regression adjusted for confounders was used to evaluate the association between quartiles of chrononutrition variables and dietary intake variables. Eating duration was positively associated with meal frequency in adults (β 0.54; p < 0.001) and elderly (β 0.50; p < 0.001). A positive association between ED and caloric midpoint was found only in the elderly (\$ 0.19; p =0.021). No associations were found between eating duration and/or caloric midpoint with total energy intake. In conclusion, our study shows that a longer eating duration and later caloric midpoint are not associated with higher energy in the American population. Further studies considering other possible mechanisms related to meal timing are necessary to address the effects of chrononutrition pattern on metabolic health in free-living human subjects.

**Keywords:** Dietary intake; Chrononutrition; Meal timing; Survey.



Title	Is self-reported short sleep duration associated with obesity? A systematic review and meta-analysis of cohort Studies
Authors	Guimarães KG¹; Silva CM¹; Latorraca COC²; Oliveira RA³; Crispim CA¹
Affiliations	<sup>1</sup> UFU PPG Ciências da Saúde, Brasil; <sup>2</sup> Cochrane Brazil; <sup>3</sup> IMEPAC Araguari, Brasil
Session	Apresentação Pôster

### Abstract and Keywords

#### Context

A possible association between self-reported short sleep duration and risk of obesity has been studied.

#### Objective

To analyze the association between sleep duration and obesity.

#### Methods

The LILACS, Medline, Central, Embase, and OpenGrey databases were searched from inception until July 2020. Two authors screened the studies independently according to the PECO strategy, as follows: participants: > 18 years old; exposure: short sleep duration; control: regular sleep; outcome: obesity). Only cohort studies were included. A total of 3286 studies were retrieved with the search strategy, but only 36 were included. Disagreements were resolved by a third author. The quality of studies was assessed with Newcastle-Ottawa Quality Assessment Form for Cohort Studies. The certainty of the evidence was assessed using Grading of Recommendations Assessment, Development, and Evaluation. Results

#### Results

Thirty-six studies were included, and 22 contributed quantitative data. Most of the studies (n = 27) assessed sleep by self-report. The meta-analysis showed a significant association between self-reported short sleep (RR, 1.18; 95%CI; 1.15-1.26, I2 = 31%; P for heterogeneity = 0.18) and development of obesity, and the chances of developing obesity increased when self-reported sleep duration decreased.

#### Conclusions

Self-reported short sleep was significantly associated with a higher incidence of obesity, with moderate quality of evidence.

**Keywords:** sleep duration, obesity, meta-analysis



Title	Metabolism effects of a supplemented diet on the kidney and liver in a doxorubicin toxicity model
Authors	Poliana Rodrigues Alves Duarte <sup>1</sup> ; Rodrigo Rodrigues Franco <sup>1</sup> ; Danielle Diniz Vilela <sup>2</sup> ; Douglas Carvalho Caixeta <sup>2</sup> ; Adriele Vieira de Souza <sup>2</sup> ; Simone Ramos Deconte <sup>2</sup> ; Clesnan Mendes-Rodrigues <sup>2</sup> ; Thiago Montes Fidale <sup>1</sup> ; Foued Salmen Espindola <sup>2</sup> ; Renata Roland Teixeira <sup>2</sup> ; Elmiro Santos Resende <sup>2</sup> .
Affiliations	<sup>1</sup> Universidade Federal de Catalão; <sup>2</sup> Universidade Federal de Uberlândia.
Session	NUTRIÇÃO E METABOLISMO

## Abstract and Keywords

Supplements and diets containing L-leucine, a branched-chain amino acid, have been considered beneficial for controlling oxidative stress and maintaining cardiac tissue in toxicity models using doxorubicin, a drug widely used in cancer treatment. However, there is a lack of studies in the literature that assess the effects of this diet on other organs and tissues, such as the liver and kidneys. Therefore, this study aimed to evaluate the effects metabolism of a leucine-rich diet on the liver and kidneys of healthy Wistar rats submitted to the doxorubicin toxicity model by analyzing biomarkers of oxidative stress and histological parameters. The animals were divided into four groups: naive, doxorubicin, L-leucine, and doxorubicin + L-leucine, and the diet was standardized with 5% L-leucine and a dose of 7.5 mg/kg of doxorubicin. We evaluated tissue injury parameters and biomarkers of oxidative stress, including enzymes, antioxidant profile, and oxidized molecules, in the liver and kidneys. Although some studies have indicated benefits of a diet rich in L-leucine for the muscle tissue of animals that received doxorubicin, our results showed that the liver was the most affected organ by the L-leucine-rich diet since the diet reduced its antioxidant defenses and increased the deposit of collagen and fat in the hepatic tissue. In the kidneys, the main alteration was the reduction in the number of glomeruli. These results contribute to the scientific literature and encourage further studies to evaluate the effects of an L-leucine-rich diet or its supplementation, alone or combined with doxorubicin using an animal model of cancer. Therefore, our study concludes that the leucine-rich diet itself was harmful and, when co-administered with doxorubicin, was not able to maintain the antioxidant defenses and tissue structure of the evaluated organs.

Keywords: doxorubicin; L-leucine; Liver and Kidneys



Title	The association between adductor pollicis muscle thickness and physical function, as well as nutrition status, in patients on hemodialysis
Authors	Débora Moreira Morais; Isadora Cordeiro Trombim; Bárbara Perez Vogt
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	Poster presentation

## and Keywords

The adductor pollicis muscle thickness (APMT) may be associated with the muscle strength in patients on hemodialysis. Moreover, because it is considered a good nutritional indicator and not influenced by body water, the APMT may be associated with the nutritional status of these patients. Therefore, the objective was to assess the association of APMT with physical function and nutritional status in patients on hemodialysis. This study was approved by the Ethics and Research Committee of the Federal University of Uberlândia (CAAE:59193822.3.0000.5152). The APMT was measured using a caliper between pollicis and index finger. Physical function was evaluated by handgrip strength (HGS), Short Physical Performance Battery (SPPB), the sit-to-stand test, walking speed test, timed up and go (TUG). Appendicular muscle mass index (AMMI) was estimated by bioelectrical impedance data. The nutritional status was evaluated by the Malnutrition Inflammation Score (MIS). Fifty-one patients were included, 60.8% men, mean age 58.4±12.6 years. Significant correlations were not found between APMT and the following physical function tests: walking speed test (r=-0.091; p=0.523), SPPB (r=-0.028; p=0.847), TUG (r=0.107; p=0.456), and the sit-to-stand test (r=0.043; p=0.770). The HGS and AMMI correlation was tested according to sex, as these variables exhibited differences for men and women: HGS for men (r= 0.133; p= 0.476) and for women (r=-0.34; p=0.888) and AMMI for men (r=0.296; p=0.105) and for women (r=0.245; p=0.298). There were no correlations between APMT and MIS (r=-0.067; p=0.643). Values of AMPT were not different between the groups according to adequate physical function. In conclusion, there were no association of APMT with physical function and nutritional status in patients on hemodialysis. Therefore, we suggest AMPT may not be a good indicator of physical function for patients on hemodialysis.

**Keywords:** Physical Function; Nutritional Assessment; Hemodialysis; Adductor Pollicis Muscle Thickness.



Title	Kombucha promotes increased survival in <i>Drosophila Melanogaster</i>
Authors	Ferreira RB; Malta SM; Marquez AS; Ueira-Vieira C.
Affiliations	Universidade Federal de Uberlândia, Instituto de Biotecnologia, Uberlândia, Brasil.
Session	Poster presentation.

### Abstract and Keywords

Kombucha is a probiotic beverage obtained by fermenting sugar-added Camellia sinensis extract by a symbiotic culture of bacteria and yeast. It is commonly homemade and traditionally associated with a series of functional claims. Although there are few studies regarding its properties, it is known that Kombucha has a series of antioxidant compounds and several beneficial microorganisms. Thanks to this, Kombucha is an interesting target for studies on aging. Therefore, animal models can be used, and the w(1118) strain of Drosophila melanogaster was preferred due to its aging-associated neurological changes. A survival test was carried out in order to evaluate the effect of Kombucha on the survival rates of *D. melanogaster*, in which adult flies of up to 3 days post eclosion were separated by sex, distributed and treated in quintuplicate. Every 48 hours, when changing the treatment medium, the dead individuals were manually counted, and the numbers obtained were recorded as absolute mortality until no living specimens remained. It was observed that groups treated with Kombucha had increased survival rates compared to control. Kombucha can and should, therefore, be better studied in order to better characterize detail its anti-aging activity.

**Keywords:** Kombucha; aging; survival; *Drosophila melanogaster*.



Title	Elevated systemic levels of angiotensin-(1-7) modulate renal dysfunction observed in the early phase of Diabetic Kidney
Authors	Laura Barroso Ferreira de Oliveira¹; Mariana Rodrigues Campos¹; Vitória Luiza Lacerda¹; Kamylle Silva Ferraz¹; Letícia Cristine Cardoso dos Santos²; Paula Peixoto Campos Lopes²; Maria Aparecida Ribeiro Vieira¹; Celso Caruso-Neves³,⁴,⁵; Maria José Campagnole-Santos¹,⁶; Robson Augusto Souza dos Santos¹,⁶; Diogo de Barros Peruchetti¹,⁶.
Affiliations	¹Department of Physiology and Biophysics, Institute of Biological Science, Federal University of Minas Gerais, Belo Horizonte, Brazil; ²Department of Pathology, Institute of Biological Science, Federal University of Minas Gerais, Belo Horizonte, Brazil; ³Carlos Chagas Filho Biophysics Institute, Federal University of Rio de Janeiro, Rio de Janeiro, Brazil; ⁴Rio de Janeiro Innovation Network in Nanosystems for Health Nano SAUDE/FAPERJ, Rio de Janeiro, Brazil ⁵INCT-Regenera, Rio de Janeiro, Brazil; ⁶INCT-Nanobiofar, Belo Horizonte, Brazil
Session	Biologia e Doenças Renais

## Abstract and Keywords

The deleterious effects of hyperglycemia on glomerular and tubular cells are crucial in the development of Diabetic Kidney Disease (DKD). However, the mechanisms involved in this process are still elusive. In this present work, we aimed to study the potential modulatory effect of angiotensin-(1-7) [Ang-(1-7)], a component of the Renin-Angiotensin System, in DKD. We used 16-week- old male L3292 transgenic rats, which present high systemic levels of Ang-(1-7), and Sprague Dawley (SD) rats as the respective controls. Type 1 diabetes mellitus was induced by a single injection of streptozotocin (STZ, 55 mg/kg). Four groups were generated: 1) SD-ND, normoglycemic SD rats (control, n=8); 2) SD-D, diabetic SD rats (n=8); 3) L3292-ND, normoglycemic L3292 rats (n=9); 4) L3292-D, diabetic L3292 rats (n=9) (CEUA-UFMG#100/2023). All analyses were performed after 3 weeks of diabetes. Diabetic groups presented similar levels of hyperglycemia (4.0-fold) as well as similar levels of increased glycosuria and urinary volumes (4.0-fold). Creatinine clearance (a marker of glomerular function) only increased (2.5-fold) in the L3292-D group. Regarding renal protein handling, despite a similar increase in proteinuria and renal clearance of proteins (3.0-fold and 2.81-fold, respectively) in both SD-D and L3292-D groups, the protein filtration was only increased (1.5-fold) in the L3292-D group. In addition, we observed a 1.35-fold increase in FEproteins associated with a 40% reduction in cortical albumin-FITC uptake in the SD-D group. Interestingly, these deleterious effects were not observed in the L3292- D group. Furthermore, a significant increase (8.4-fold) in urinary  $\gamma$ -glutamyl transferase, a proximal tubule injury marker, was observed only in the SD-D group. Altogether, our preliminary data suggest that higher systemic levels of Ang-(1-7) promote dual effects on diabetic kidneys; it contributes to glomerular hyperfiltration while attenuating tubular dysfunction at early stages of DKD.

**Keywords:** Diabetic Kidney Disease, proteinuria, tubular injury, angiotensin-(1-7)



Title	Activation of Ang II/AT1R axis mediates the tubular injury associated with proteinuria observed in the early stage of diabetic kidney disease
Authors	Mariana Rodrigues Campos <sup>1</sup> ; Laura Barroso Ferreira de Oliveira <sup>1</sup> ; Vitoria Luiza Lacerda <sup>1</sup> ; Letícia Cristine Cardoso dos Santos <sup>2</sup> ; Paula Peixoto Campos Lopes <sup>2</sup> ; Maria Aparecida Ribeiro Vieira <sup>1</sup> ; Celso Caruso-Neves <sup>3,4,5</sup> ; Diogo de Barros Peruchetti <sup>1,6</sup>
Affiliations	¹Department of Physiology and Biophysics, Institute of Biological Science, Federal University of Minas Gerais, Belo Horizonte, Brazil; ²Department of Pathology, Institute of Biological Science, Federal University of Minas Gerais, Belo Horizonte, Brazil; ³Carlos Chagas Filho Biophysics Institute, Federal University of Rio de Janeiro, Rio de Janeiro, Brazil; ⁴Rio de Janeiro Innovation Network in Nanosystems for Health – NanoSAUDE/FAPERJ, Rio de Janeiro, Brazil; ⁵INCT-Regenera, Rio de Janeiro, Brazil; 6INCT-Nanobiofar, Belo Horizonte, Brazil.
Sassion	Biologia e Doenças Renais

#### and Keywords

Diabetic kidney disease (DKD) pathogenesis involves tubular dysfunction associated with tubular proteinuria. The mechanism involved in this process is still poorly understood. Clues come from reports showing the renoprotective effects of losartan, a specific antagonist of the angiotensin type 1 receptor (AT1R), on different renal diseases. Herein, we investigated the potential role of AT1R activation in tubular proteinuria and injury at the early stage of DKD. Ten-week-old male Wistar rats were used to develop DKD using streptozotocin (STZ, 55 mg/kg i.v.). When indicated, the animals were daily treated with 30 mg/kg/day of losartan via gavage. Four groups were generated: 1) CONT, normoglycemic rats (n=5); 2) STZ, diabetic rats (n=4); 3) STZ+LOS, diabetic rats treated with losartan (n=5); 4) LOS, normoglycemic rats treated with losartan (n=3). All analyses were performed after 8 weeks (CEUA-UFMG #100/2023). There were no changes in plasma creatinine, plasma urea, creatinine clearance, or glomerular structure parameters. Compared with the CONT group, the STZ group presented: 1) increased blood glucose (3-fold) and urinary flow (6-fold); 2) an increase in proteinuria (mg/48h) and renal protein clearance (2.5-fold and 12.4-fold, respectively) associated with an increased fractional excretion of proteins (4.3-fold, FEproteins, a marker of renal protein reabsorption); 3) a 3.4-fold increase in urinary γ-glutamyl transferase (a marker of tubular injury); 4) an increased kidney injury score (8.0±0.89 arbitrary units) associated with a 2.45-fold increase in interstitial cell infiltration. Interestingly, except for blood glucose and urinary flow, the treatment with losartan attenuated all tubular proteinuria and injury parameters in the STZ+LOS group. No difference was observed between the CONT and LOS groups. Altogether, our preliminary data indicate that the Activation of AT1R mediates hyperglycemia-induced tubular injury and proteinuria observed at the early stages of DKD.

Keywords: diabetes, kidney disease, proteinuria, angiotensin II



Title	Oral treatment with angiotensin-(1-7) ameliorates proteinuria in subclinical acute kidney injury animal model
Authors	Vitória Luiza Lacerda <sup>1</sup> ; Mariana Rodrigues Campos <sup>1</sup> ; Laura Barroso Ferreira de Oliveira <sup>1</sup> ; Maria Aparecida Ribeiro Vieira <sup>1</sup> ; Robson Augusto Souza dos Santos <sup>1,2</sup> ; Celso Caruso-Neves <sup>3,4,5</sup> ; Diogo Barros Peruchetti <sup>1,2</sup>
Affiliations	¹Department of Physiology and Biophysics, Institute of Biological Science, Federal University of Minas Gerais, Belo Horizonte, Brazil; ²INCT-Nanobiofar, Belo Horizonte, Brazil; ³Carlos Chagas Filho Biophysics Institute, Federal University of Rio de Janeiro, Rio de Janeiro, Brazil; ⁴Rio de Janeiro Innovation Network in Nanosystems for Health – NanoSAUDE/FAPERJ, Rio de Janeiro, Brazil; ⁵INCT-Regenera, Rio de Janeiro, Brazil.
Session	Biologia e Doenças Renais

## Abstract and Keywords

Subclinical Acute Kidney Injury (SubAKI) is a clinically silent syndrome associated with tubular injury and proteinuria without changes in glomerular function. Previous works point out that overactivation of the intrarenal Renin-Angiotensin System mediates these processes in subAKI. However, the mechanism involved in this process still needs to be elucidated. In this present work, we studied the potential effects of Ang-(1-7) on development of proteinuria observed in a subAKI animal model. Eight-week-old female C57Bl6 mice were used to generate the following groups: 1) CONT (n=5), control; 2) subAKI (n=5), mice treated with i.p. injection of 10g/kg/day bovine serum albumin (BSA) and oral doses of hydroxypropyl β-cyclodextrin (HPβCD) for 7 consecutive days; 3) subAKI+Ang-(1-7) (n=5), mice treated with BSA and 30µg/kg/day Ang-(1-7) included in HPβCD [HPβCD-Ang-(1-7)] via gavage; and 4) Ang-(1-7) (n=5), mice treated with HPβCD-Ang-(1-7) alone (CEUA-UFMG#100/2023). Then, the animals were housed in metabolic cages for collection of 24h urine for biochemical analysis. No changes were observed in urinary volume, urinary flow, or the glomerular function parameters assessed. Compared to the CONT group, the subAKI group presented an increase in: 1) proteinuria and urinary protein mass (mg/24h) by 10-fold and 8-fold, respectively; and 2) urinary γ-glutamyl transferase activity (U/L), a specific tubular injury marker, by 8-fold. Co-treatment with HPβCD-Ang-(1-7) attenuated these alterations observed in subAKI (subAKI+Ang-(1-7) group). Our preliminary data allow us to suggest that the oral formulation of Ang-(1-7) ameliorates the development of proteinuria and tubular injury observed in subAKI.

**Keywords:** subclinical acute kidney injury, proteinuria, angiotensin-(1-7)



Title	The absence of interferon-gamma protects animals with experimental cerebral malaria from tubular damage during acute kidney injury
Authors	Fernanda de Souza Freitas <sup>1</sup> ; Eduardo Ferreira da Silva <sup>1</sup> ; Gabriela Rosa da Silva Costa <sup>1</sup> ; Giovanna Bertolino Vieira <sup>1</sup> ; Kleber Simônio Parreira <sup>1,2</sup> ; Wânia Rezende Lima <sup>1</sup> .
Affiliations	<sup>1</sup> Universidade Federal de Catalão, Catalão, Brasil <sup>2</sup> Centro Universitário do Triângulo-UNITRI, Uberlândia, Brasil
Session	Biologia e doenças renais

## and Keywords

Acute kidney injury (AKI) is one of the main complications of severe malaria that causes fatalities in patients infected with Plasmodium falciparum. Falciparum, in the kidneys, leads to glomerular filtration loss, tubular necrosis, and glomerulonephritis. It is believed that sequestration of infected erythrocytes, blockage of renal microcirculation, immune-mediated glomerular damage, and blood volume depletion are responsible for renal injury. However, the contribution of the immune response to AKI during severe malaria is poorly understood. Therefore, the aim of this research was to investigate the pathogenesis of AKI associated with experimental cerebral malaria model (ECM) in interferon-gamma-deficient (IFN-KO) mice. Animal models, C57BL/6J and IFN-KO were infected with P. berghei Anka (PbA), and morphometric analysis was conducted using the ImageJ image software and histopathological analyses were performed. Animals were used in accordance with the approval of the Animal Ethics Committee-UFU number 002/21. Morphometric results revealed that IFN-KO animals infected with PbA had a significant increase in both Bowman's capsule space measurements (p≤0.0017) and glomerular area (p≤0.0086) compared to the control group using ANOVA and the Tukey test. Periodic acid-Schiff staining findings showed that infected C57BL/6J animals exhibited leukocytic migration within blood vessels, tubular damage, renal corpuscle damage with collapse of glomerular tuft, and sclerosis. In contrast, infected IFN-KO animals showed leukocytic migration within blood vessels and collapse of glomerular tuft, but no evident tubular damage was observed. The proximal convoluted tubules showed intact cells with preserved brush border. In summary, our research revealed that the absence of IFN-gamma altered the morphometry of the renal corpuscle but may protect against tubular damage during AKI in ECM complications, thereby contributing to the role of IFN-gamma in the pathogenesis of AKI in malaria.

Keywords: P. berghei Anka; acute kidney injury; Interferon gamma



Title	Gene ontology analysis of salivary proteomics in chronic kidney disease
Authors	Bianca Uliana Picolo <sup>1</sup> ; Nathalia Rabello Silva <sup>2</sup> ; Luiz Ricardo Goulart Filho ( <i>in memorian</i> ) <sup>1</sup> ; Mario Machado Martins <sup>1</sup> ; Hebréia Oliveira Almeida Souza <sup>1</sup> ; Letícia Cristina Machado de Sousa <sup>2</sup> ; Ana Luiza Barbosa Ferreira <sup>2</sup> ; Richard Costa Polveiro <sup>3</sup> ; Robinson Sabino da Silva <sup>4</sup> ; Vivian Alonso-Goulart <sup>1</sup> ; Luciana Saraiva da Silva <sup>2</sup> .
Affiliations	<sup>1</sup> Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart, Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, MG, Brazil. <sup>2</sup> Faculty of Medicine, Federal University of Uberlândia, Uberlândia, MG, Brazil. <sup>3</sup> Laboratory of Bacterial Diseases, Sector of Preventive Veterinary Medicine and Public Health, Department of Veterinary Medicine, Federal University of Viçosa, Viçosa, MG, Brazil. <sup>4</sup> Innovation Center in Salivary Diagnostic and Nanobiotechnology, Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, MG, Brazil.
Session	Biologia e Doenças Renais

# and Keywords

Chronic kidney disease (CKD) is a public health challenge due to the lack of reliable markers for diagnosis and monitoring. Salivary proteomics emerges as a promising approach to identify non-invasive biomarkers and allows the study of pathways through which these biomarkers may act in CKD. This study aims to carry out the gene ontology of the proteins found in the salivary proteomic profile. This study received ethical approval by the Human Research Ethics Committee of the Federal University of Uberlândia (approval number 4.430.315). Saliva was collected from ten individuals with chronic kidney disease undergoing hemodialysis and ten healthy individuals, which were analyzed using liquid chromatography coupled to a mass spectrometer (LC-MS/MS). The spectra obtained were processed by SpectrumMill software. Statistical analyzes were performed using STATA 14.2 and MetaboAnalyst 5.0 software. The enrichment analysis was performed using gene ontology (GO) for selected proteins, by the PANTHER software. From these analyses, it was possible to observe that the most abundant proteins, present in more than 50% of participants in each group, showed molecular functions with structural and catalytic activities predominant in the CKD group, while the binding function is prominent in the healthy group. In relation to biological processes, signaling processes, response to stimuli and metabolic processes are more prevalent in CKD. The classes phospholipase and protease inhibitor were evident in the healthy group. In the differential proteins between the groups, candidates for salivary biomarkers, the binding functions, catalytic activity and molecular regulation were highlighted, as well as cellular process and biological regulation. From these analyses, it is possible to conclude that CKD involves many mechanisms and pathways that need to be further studied at the molecular level, contributing to future applications in the diagnostic and prognostic field.

**Keywords:** CKD,LC-MS/MS,gene ontology



Title	Gene ontology analysis of urinary proteomics in chronic kidney disease
Authors	<sup>1</sup> Letícia Cristina Machado de Sousa; <sup>1</sup> Nathalia Rabello Silva; <sup>2</sup> Bianca Uliana Picolo; <sup>1</sup> Marta Silva dos Santos; <sup>3</sup> Richard Costa Polveiro; <sup>2</sup> Luiz Ricardo Goulart Filho (in memoriam); <sup>2</sup> Hebréia Oliveira Almeida Souza; <sup>1</sup> Michelly Yorrana Miranda de Araujo; <sup>2</sup> Mario Machado Martins; <sup>1</sup> Luciana Saraiva da Silva.
	<sup>1</sup> Faculty of Medicine, Federal University of Uberlândia, Uberlândia, MG, Brazil.
Affiliations	<sup>2</sup> Nanobiotechnology Laboratory Prof. Luiz Ricardo Goulart, Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, MG, Brazil.
	<sup>3</sup> Laboratory of Bacterial Diseases, Sector of Preventive Veterinary Medicine and Public Health, Department of Veterinary Medicine, Federal University of Viçosa, Viçosa, MG, Brazil.
Session	Biologia e Doenças Renais

### Abstract and Keywords

The identification of new biomarkers that make it possible to monitor the progression of chronic kidney disease (CKD) is necessary due to the growing number of people with the disease caused mainly by changes in lifestyle and population aging. This study aims to carry out the gene ontology of the proteins found in the urinary proteomic profile. For proteomic analysis, urine samples were collected from 20 individuals, 10 healthy and 10 with terminal CKD. These samples were analyzed using liquid chromatography equipment coupled to a tandem mass spectrometer and then bioinformatics, gene ontology and protein interaction analyzes were carried out. The study was approved by the Human Research Ethics Committee (CEP) of the Federal University of Uberlândia (UFU), under opinion number 4,430,315. Among the differential proteins between the groups, it is possible to observe the characterization of the gene ontology for different functions and processes. In relation to molecular function and biological process, proteins had greater association with binding and cellular process functions, respectively. In relation to the cellular component, there is a greater participation of proteins in the cellular anatomical entity. Finally, regarding the class of proteins, it is worth highlighting that these were mainly related to the transfer/transport protein class. Among the proteins that showed a significant difference, five (hemopexin, beta-2-microglobulin, retinol-binding protein 4, transthyretin and factor D) were considered potential biomarkers for CKD, because the mechanisms of action of these proteins are involved with the pathophysiology of the disease.

**Keywords:** Biomarkers, Chronic kidney disease, Proteomics.



Title	Salivary proteomic characterization in chronic kidney disease
Authors	Ana Laura Fonseca Orlandi <sup>2</sup> ; Bianca Uliana Picolo <sup>1</sup> ; Nathalia Rabello Silva <sup>2</sup> ; Luiz Ricardo Goulart Filho ( <i>in memorian</i> ) <sup>1</sup> ; Mario Machado Martins <sup>1</sup> ; Hebréia Oliveira Almeida Souza <sup>1</sup> ; Letícia Cristina Machado de Sousa <sup>2</sup> ; Richard Costa Polveiro <sup>3</sup> ; Robinson Sabino da Silva <sup>4</sup> ; Vivian Alonso-Goulart <sup>1</sup> ; Luciana Saraiva da Silva <sup>2</sup> .
Affiliations	<sup>1</sup> Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart, Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, MG, Brazil; <sup>2</sup> Faculty of Medicine, Federal University of Uberlândia, Uberlândia, MG, Brazil; <sup>3</sup> Laboratory of Bacterial Diseases, Sector of Preventive Veterinary Medicine and Public Health, Department of Veterinary Medicine, Federal University of Viçosa, Viçosa, MG, Brazil; <sup>4</sup> Innovation Center in Salivary Diagnostic and Nanobiotechnology, Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, MG, Brazil.
Session	Biologia e Doenças Renais

### Abstract and Keywords

Chronic kidney disease (CKD) affects several individuals, which is due to the lack of reliable, accurate and low-cost markers for early diagnosis and monitoring of the disease, negatively impacting quality of life and generating additional costs for the public health system cover treatment expenses. To identify potential CKD biomarkers contributing to early diagnosis, a non-invasive alternative is salivary proteomics. The aim of this study was to analyze salivary proteomics to identify potential biomarker candidates for CKD. This study received ethical approval from the Human Research Ethics Committee of the Federal University of Uberlândia (approval number 4.430.315). Ten individuals with CKD undergoing hemodialysis and ten healthy individuals participated in this study, from whom sociodemographic variables and saliva were collected. Fifty micrograms of saliva protein samples were quantified for protein solution digestion by trypsinization, and subsequently analyzed using liquid chromatography coupled with mass spectrometry (LC-MS/MS), with the obtained spectra processed using SpectrumMill software. Statistical analyses were performed using Agilent Mass Profiler Professional (MPP) v.B.13.1.1, STATA 14.2, and MetaboAnalyst 5.0 softwares. The following tests were performed to differentiate the groups: t-test corrected by the FDR criterion, orthogonal discriminant analysis by partial least squares (OPLS-DA), volcano graph, heat map and ROC curve. Regarding salivary proteomics, 431 proteins were found, with three proteins (API-5, SGSM2, and PI-PLC) showing statistical differences between the analyzed groups, with high sensitivity and specificity. Thus, salivary proteomic characterization revealed differential proteins between the analyzed groups, which could be considered potential biomarkers for CKD, with application in the theranostic field.

**Keywords:** Proteomics, LC-MS/MS, kidney disease, biomarkers.



Title	Urinary proteomic profile in end-stage chronic kidney disease
Authors	Pamella Arrais Vilela <sup>1</sup> ; Nathalia Rabello Silva <sup>1</sup> ; Bianca Uliana Picolo <sup>2</sup> ; Letícia Cristina Machado de Sousa <sup>1</sup> ; Marta Silva dos Santos <sup>1</sup> ; Richard Costa Polveiro <sup>3</sup> ; Luiz Ricardo Goulart Filho (in memoriam) <sup>2</sup> ; Hebréia Oliveira Almeida Souza <sup>2</sup> ; Mario Machado Martins <sup>2</sup> ; Luciana Saraiva da Silva <sup>1</sup> .
Affiliations	<sup>1</sup> Faculty of Medicine, Federal University of Uberlândia, Uberlândia, MG, Brazil; <sup>2</sup> Nanobiotechnology Laboratory Prof. Luiz Ricardo Goulart, Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, MG, Brazil; <sup>3</sup> Laboratory of Bacterial Diseases, Sector of Preventive Veterinary Medicine and Public Health, Department of Veterinary Medicine, Federal University of Viçosa, Viçosa, MG, Brazil.
Session	Biologia e Doenças Renais

## Abstract and Keywords

Chronic kidney disease (CKD) is characterized by a progressive loss of kidney function associated with a consequent reduction in blood filtration capacity and maintenance of homeostasis. This disease is considered a serious public health problem as it is associated with high morbidity and mortality rates. Therefore, it is necessary to identify new biomarkers for the diagnosis and monitoring of CKD progression. This study aimed to describe the urinary proteomic profile of healthy individuals and those with CKD on hemodialysis to identify potential biomarkers for the disease. This study received ethical approval from the Human Research Ethics Committee of the Federal University of Uberlândia (approval number 4,430,315). Ten individuals with CKD on hemodialysis and ten healthy individuals participated in the study. Sociodemographic data and urine samples were collected. The samples were analyzed by liquid chromatography coupled to a mass spectrometer (LC-MS/MS) and the spectra obtained were processed by SpectrumMill software. Statistical analyzes were performed using Agilent Mass Profiler Professional (MPP) v.B.13.1.1, STATA 14.2 and MetaboAnalyst 5.0 software. The following tests were carried out to differentiate the groups: t-test corrected by the FDR criterion, orthogonal discriminant analysis by partial least squares (OPLS-DA), volcano plot and heatmap. Regarding urinary proteomics, 416 proteins were identified in the proteomic profile, with 19 proteins showing statistical differences between the groups analyzed. These proteins can be considered biomarker candidates due to their high sensitivity and specificity. It is concluded that the proteins found were capable of characterizing and differentiating the urinary proteomic profiles of the two groups.

**Keywords:** DRC, LC-MS/MS, proteomics.



Title	Evaluation of pulmonary compliance in mice contaminated with particulate matter (PM) and in the asthmatic model
Authors	Thalia Giovana Guimarães Ribeiro¹; Luciano Rivaroli¹
Affiliations	<sup>1</sup> Departament of Natural Sciences /Federal University of São João Del-Rei, São João del Rei, MG, Brasil.
Session	Biology and Respiratory Diseases, oral poster.

## and Keywords

A pollutant that gains attention on environmental contamination is Particulate Matter (PM), that affects urban air quality and directly increase the incidence of respiratory diseases. Assessing the impact of PM exposure is critical to understand and protect exposed populations. 32 adult male Balb/C mice were used (CEUA 4277220523/UFSJ protocol). They were divided into four groups (n=8): Control (C), Asthma Induced (AI), PM Contaminated (PM) and AI and PM Contaminated (AIPM). After asthma induction (i.p. injection of egg albumin (EA) with subsequent instillation of EA) and exposure to PM (nasal instillation), the animals had their left lungs collected and placed in an organ bath coupled to a force transducer for tension analysis. Stretches of 1 mm were performed with a micromanipulator while the tension was recorded. The one-way ANOVA test was used to evaluate the feasibility of the data and the Kolmogorov-Smirnov test to evaluate normality, the Student's t-test was to analyze the difference between the groups, being considered significant when p < 0.05. All groups showed progressive increases in tensions with the stretches. The mean tensions in each stretch point were significantly higher in the order C<AI<PM<AIPM, in the maximum stretch point, supported by all groups (7mm), presented respectively, the tensions:  $10.35\pm7.26$ ;  $17.47\pm9.73$ ;  $35.24\pm19.89$ ;  $63.62\pm37.10$  gf.g <sup>1</sup>(gram-force per gram of tissue). The slope coefficient of the trend lines corroborates these data: 2.6; 2,53; 6,40; 6.71. The data indicate that animals exposed to particulate matter have lower tissue compliance. The data also show that the asthma condition potentiates the reduction of complacency, indicating alterations in tissue architecture, probably due to the immune response to the contaminant in addition to the sensitization of the asthmatic model. Ongoing histological analyses may help to understand these initial results.

**Keywords:** Lung, Particulate Matter, Tissue Mechanical Properties, Compliance



Title	Respiratory muscle strength and quality of life in post- COVID-19 patients
Authors	Luís Fillipe Maciel da Silveira; Carlos Fernando Ronchi; Juliana de Fátima Cruz; Angelo Piva Biagini.
Affiliations	UFU, Uberlândia, Brasil
Session	Biologia e Doenças Respiratórias

# Abstract and Keywords

Infection with the new coronavirus (SARS-CoV-2), known as COVID-19, results in severe acute impairment of the respiratory system. COVID-19 has many negative effects among infected individuals. This study evaluated respiratory muscle strength (RMS), quality of life (QoL) and possible correlations between the RMS variables and the physical functioning score evaluated by the Short Form Health Survey (SF-36) in a group previously diagnosed with COVID-19, being a quantitative, cross-sectional, observational group study, with convenience sampling, dependent on age and independent of sex, which was approved by Human Ethics Committee - Federal University of Uberlândia (protocol 1.182.730). The 28 volunteers included responded to SF-36, which comprises the scores: Physical Functioning (PF), Role Physical (RP), Bodily Pain (BP), General Health (GH), Vitality (VT), Social Functioning (SF), Role Emotional (RE) and Mental Health (MH) and performed the FMR test using maximum respiratory pressures (maximum inspiratory pressure (MIP) and maximum expiratory pressure (MEP)) with the manovacuometer. Our findings indicate that the mean RMF was lower than predicted, women MIP and MEP of 58,23 and 64,11 cmH2O, predicted value 79,87 and 111,7 cmH2O; men MIP and MEP of 71,36 and 87,27 cmH2O, predicted value 102,28 and 111,61 cmH2O. The average SF-36 performance in all domains was greater than 50 points (PF = 63,92; AF = 59,82; DR = 53,60; EGS = 64,28; VT = 53,39; AS = 56,69; AE = 58,33; SM = 58,42) and there was a positive correlation between FMS and the PF score (r = 0.884, p  $\leq 0.05$ ). It is possible to conclude that post-COVID-19 patients have reduced RMS. The QoL results were normal, making it possible to establish a positive correlation between RMS and PF score, showing that higher RMS was correlated with better PF.

**Keywords:** Post-COVID-19, COVID-19, respiratory muscle strength, quality of life, physical functioning, Sf-36.



Title	Resistance exercise effects on obstructive sleep apnea (OSA)
Authors	Alinny Cristiny de Araujo Peres, Ana Carolina Ferreira Fernandes, Wanessa Silva de Oliveira, Alice Menezes Batista, Eduardo Henrique Rosa Santos, Angelo Piva Biagini, Carlos Fernando Ronchi.
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brasil.
Session	Biologia e Doenças Respiratórias

### Abstract and Kevwords

Obstructive Sleep Apnea (OSA) is characterized by repetitive upper airway collapse during sleep, which causes impairment in cardiac, metabolic, and neurocognitive function due to sleep superficialization and fragmentation. Exercise may represent a key resource in treatment as it improves sleep architecture and venous return, however there is still no consensus regarding it. This study aimed to evaluate resistance exercise impact on sleep symptoms, as well as in pulmonary, anthropometric, and neurocognitive repercussions in OSA patients. Protocol (nº65269122.2.0000.5152) had 24 sessions (12 weeks) using 8 resistance exercises for arms, legs, and abdomen. Seventeen volunteers (9 men and 8 women) who were at high risk of OSA based in Berlin questionnaire were included. Mean age was 49.29 ± 9.37 years, and body mass index (BMI)  $29.17 \pm 3.82 \text{ kg/m}^2$ . Comparing pre and post intervention, we had significant results in sleep apnea risk score (Berlin) decreasing from 2.29±0.47 to  $1.76\pm0.75$  (p=0.014), and sleep quality score (PSQI) from  $9.06\pm3.15$  to 7.18±2.81 (p=0.012). Regarding respiratory muscle strength, we had an improvement with Pimax from 82.06±25.17 to 93.64±24.23 cmH2O (p=0.002), and Pemax 93.18±26.13 to 101.23±22.12 cmH2O (p=0.04). In addition, we also found a reduction in waist circumference from 95.99±10.01 to  $94.16\pm8.95$ cm (p=0.012). There was no significant improvement in sleepiness, hip, calf and neck circumferences, BMI, body mass and cognitive function. Therefore, based in our finding resistance exercise can be used in OSA treatment to improve sleep quality, and respiratory strength, as well as to reduce cardiovascular risks related to abdominal fat. Due to its low cost and easy applicability, it can be applied in primary care. In addition, it can be used simultaneously with CPAP in severe OSA patients.

**Keywords:** Obstructive Sleep Apnea, Exercise, Sleep quality.



Title	New In Vitro therapeutic approaches of anti-activin a mimetic peptide in lung cells in cancer disease
Authors	Natieli Saito; Maria Eduarda Costa Mundim; Victor Alexandre Felix Bastos; Phelipe Augusto Borba Martins Peres; Thulio Marquez Cunha
Affiliations	Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart Filho, Institute of Biotechnology (IBTEC), Federal University of Uberlândia, Uberlândia, Brazil.
Session	Biology and Respiratory Diseases

### Abstract and Keywords

Lung cancer is a severe disease affecting a significant portion of the population. However, current treatments have limited efficacy in substantially increasing patients' life expectancy. Consequently, this study aimed to explore an alternative approach in cancer therapy by utilizing a peptide antagonist of Activin A, a molecule implicated in lung tumorigenesis. The E10 peptide was selected for initial experiments utilizing the phage display technique. It was used as a treatment in immortalized lung cells: Beas-2B (ATCC CRL-9609), a normal bronchial epithelium, and A549 (CRM-CCL-185), a human lung carcinoma. In vitro, assays assessing cell cytotoxicity, proliferation, and wound healing demonstrated the peptide's efficacy across different concentrations. The methodology employed for cell cytotoxicity and proliferation involved the reduction of diphenyltetrazolium bromide (MTT), a yellow tetrazolium dye, converted by viable cells into purple formazan crystals. The cells were treated with 1, 10, and 50 µM of the E10 peptide, with and without the addition of pmTGF-β. After 24 hours, the peptide significantly reduced proliferation without inducing cytotoxicity, as observed in the control comparison. Cells were cultured for the wound healing assay, and the cell monolayer was scratched with a p200 pipet tip. The cells were treated with the peptide at concentrations of 1,10, and 50 μM for 24h, with additional groups treated with and without pmTGF-β1. The assay measured cellular migration through the occupied area using Image J software, confirming the peptide's ability to inhibit cellular migration in A549 cells, irrespective of pmTGF-β1 presence. In conclusion, these preliminary findings indicate that the E10 peptide exhibits promising results in proliferation and cytotoxicity, suggesting its potential to act on specific receptors and modify cell migration capacity, crucial characteristics in lung cancer cells.

**Keywords:** lung cancer; Activin A; peptide



Title	Evaluation of TNF-alpha role in occupational disease silicosis using murine model
Authors	Matheus Morais Neves; Sandra Gabriela Klein; Ray César Silva; Flávia Ferreira Batista; Isabela Lemos de Lima; Wânia Rezende Lima; Murilo Vieira da Silva.
Affiliations	Laboratory of Biotecnology in Experimental Model - LABME, Federal University of Uberlândia, Uberlândia 38.405-330, Brazil.
Session	Biologia e Doenças Respiratórias Apresentação Pôster

### Abstract and Keywords

Among occupational diseases, respiratory diseases have a high incidence, causing various health damages to workers, such as silicosis, the focus of the present summary. This pathology, caused by inhalation of silica particles, is characterized by an important inflammatory process. In this context, we sought to understand the pathological evolution of the disease as well as the impact of TNF-alpha molecules, of paramount importance in inflammation, through a reliable and human-like model, in order to seek ways to suppress the pathology. This study aimed to evaluate the role of the cytokine TNF-alfa in histological and biochemical changes related to oxidative damage, in the face of silicosis-induced inflammation. To this end, silica at a concentration of 200mg/kg body weight was instilled intratracheally in C57BL/6 and receptor TNF-alpha knockout animals, with the approval of the ethics committee, protocol number 041/21, receptor knockout animals, which were followed for 28 days, followed by euthanasia, lung collection, and histological and biochemical analysis. There was a significant increase in pulmonary septal thickness in all instilled groups compared to controls; however, there was no significant increase in molecules marking oxidative damage correlating with inflammation in lung tissue homogenates. Therefore, histological damage was the main marker of the pathological progression of silicosis, with minimal involvement of biochemical inflammation markers, and no apparent decisive impact of the TNF-alpha molecule in this context, as evidenced by its similarity between the TNF-alpha receptor knockout and wild-type animals.

**KeyWords:** Occupacional disease, Oxidative-stress, Inflammation, Silicosis, Animal model.



Title	Cardiovascular responses produced by the angiotensinergic dipeptide – angiotensin- (1-2) in the rostral ventrolateral medulla of normotensive rats
Authors	Andréa Moreira dos Santos <sup>1</sup> ; Lara Marques Naves <sup>1</sup> ; Stéfanne Madalena Marques <sup>1</sup> ; Clos Henrique de Castro <sup>2</sup> ; Gustavo Rodrigues Pedrino <sup>1</sup> ; Aline Andrade Mourão <sup>1</sup> ; Daniel Alves Rosa <sup>1</sup>
Affiliations	<sup>1</sup> Center of Neuroscience and Cardiovascular Research - Federal University of Goiás, Goiânia-GO, Brazil.
	<sup>2</sup> Integrated Laboratory of Cardiovascular and Neurological Pathophysiology – Federal University of Goiás, Goiânia-GO, Brazil.
Session	Neurobiologia

## and Keywords

The Renin-Angiotensin System (RAS) is responsible to the maintenance of cardiovascular homeostasis, the regulation of hydroelectrolytic balance, the physiological control of blood pressure, extracellular volume, and tissue perfusion, among other functions. The dipeptide Aspartate<sup>1</sup>-Arginine<sup>2</sup> [Angiotensin-(1-2)] exhibits vasodilatory effects and a reduction in blood pressure via intravenous infusion. However, the central effects of Ang-(1-2) on cardiovascular function remain elusive. In light of this, the aim of this study was to explore the potential cardiovascular effects elicited by the nanoinjection of Ang-(1-2) into the rostroventrolateral medulla (RVLM) of anesthetized normotensive rats. To this end, Wistar rats (WT) weighing between 250-350 grams were anesthetized and instrumented for the measurement of mean arterial pressure (MAP), heart rate (HR), renal blood flow (RBF), and aortic blood flow (ABF). All experimental procedures were submitted to the Ethics Committee for Animal Use of UFG (process number: 013/21). The animals were underwent unilateral nanoinjections (50 nL) of glutamate (10 mM), saline solution (NaCl; 0.15 M, n=10), and Ang-(1-2) ( $10^{-10} \text{ mol/L}, \text{ n}=5; 10^{-8} \text{ mol/L}, \text{ n}=5; 10^{-5} \text{ mol/L}$ , n=10;  $10^{-3}$  mol/L, n=4) into the RVLM region. As anticipated, glutamate nanoinjections into the RVLM region resulted in an increase in MAP compared to saline. The saline nanoinjections did not alter the baseline values of MAP, HR, RBF, renal vascular conductance (RVC), ABF, and aortic vascular conductance (AVC). Regarding the nanoinjections of Ang-(1-2) at concentrations of 10<sup>-10</sup>, 10<sup>-10</sup> <sup>8</sup>, 10<sup>-5</sup>, and 10<sup>-3</sup> mol/L into the RVLM, no significant differences were observed in MAP, HR, RBF, RVC, ABF, and AVC compared to saline. The findings of this study allow us to assert, for the first time, that the nanoinjection of Ang- (1-2) into the RVLM region does not induce cardiovascular alterations in normotensive rats.

**Keywords:** Cardiovascular; Diesease, Renin-Angitensin System; Angiotensinergic peptides; Angiotensin-(1-2); Angiotensin-(1-7); Hypertension



Title	Analysis of the relationship between sleep quality and depression symptoms in primiparous women
Authors	Wanessa Silva de Oliveira Angelo Piva Biagini
	Vanessa Santos Pereira Baldon Eduardo Henrique Rosa Santos
Affiliations	Federal University of Uberlândia
Session	Neurobiologia

### Abstract and Keywords

Studies indicate an association between sleep quality and postpartum depression. Poor sleep during the postpartum period is hindered by childcare demands, leading to sleep interruptions, which is one of the risk factors for the development of depression. Investigating maternal depression is important due to its negative effects on both the mother and the child in the short and long term. In cases of severe depression, there is a risk of suicide, in addition to hindering infant growth. To verify the association between sleep quality and levels of depression symptoms in primiparous women. A cross-sectional study was conducted with a sample of 80 primiparous women. Data collection was carried out in a virtual environment using the Pittsburgh Sleep Quality Index (PSQI) questionnaire to assess sleep quality and the Depression Anxiety Stress Scales (DASS-21) to evaluate levels of depression, anxiety, and stress, categorized as normal, mild, moderate, severe, and extremely severe. The study was approved by the Research Ethics Committee of the Federal University of Uberlândia (Approval number: 59587922.2.0000.5152). Fisher's Exact Test was applied for data analysis. From the collected data, it was observed that 36 women (45%) of the sample exhibited normal levels of depression, of which 29 (36.25%) had poor sleep quality. Additionally, 71 women (88.75%) had poor sleep quality, with 29 (36.25%) of these women displaying normal levels of depression. Furthermore, 9 women (11.25%) demonstrated good sleep quality, with only 1 (1.25%) of these women showing mild levels of depression. No association was found between sleep quality and depression symptoms (p > 0.05). In the present study, there was no association between sleep quality and depression symptoms in primiparous women.

**Keywords:** Postpartum period, sleep, postpartum depression.



Title	Systemic inflammation and its detrimental effects on neuroplasticity during aging
Authors	Giovanna Isabelli de Freitas <sup>1</sup> Marcelo de Cássio Barreto de Oliveira <sup>1</sup> Sandra Regina Mota Ortiz <sup>2</sup> Gilberto Cândido Laurentino <sup>2</sup> Aline Gavioli <sup>2</sup>
Affiliations	<sup>1</sup> Universidade Cidade de São Paulo, São Paulo, Brazil <sup>2</sup> Universidade São Judas Tadeu, Sâo Paulo, Brazil
Session	Neurobiology

### Abstract and Keywords

The aging process is impacted by functional and cognitive changes, increased oxidative stress and chronic inflammation. Neuroinflammation is associated with cognitive decline and is the main cause of several pathologies related to the nervous system, impairing neuroplasticity, which acts as a fundamental mechanism for the formation of new neural circuits and strengthening synaptic activities, which is greatly impaired in neuroinflammation and oxidation.

Dual-task exercise (adapted for the enriched environment for rodents) combines cognitive and motor function, combining aerobic exercise with cognitive stimuli, awakening more brain areas. It helps increase synaptic plasticity, brain vascularization, synaptogenesis and improves functional performance. Objective: to evaluate the improvement in neuroplasticity associated with low systemic inflammation in animals exposed to the enriched environment. Twelve 15-month-old male Wistar rats (CEUA:091, approved in 2023) were used, divided into the Control Group (n=6) (CG) and the Enriched Environment Group (n=6) (EA). The animals were exposed 5x a week, 1 hour/day for 13 weeks in na environment consisting of: a 1.00m x 0.50m acrylic box, a PVC tunnel, cardboard, wooden blocks, a seesaw, a ladder, a suspended hemp thread , a wooden bar and sponges. Its housing boxes also contained a wooden ball, cardboard tube and wooden roll. To evaluate inflammation and oxidative stress, 3.5ml of blood from the animal was collected via venipuncture in the EDTA tube for ELISA. TNF-a, IL-6, IL-10 and reactive oxygen species will be analyzed. The material is being processed and will be statistically evaluated. Based on a subjective analysis, the EA group suggested a behavioral improvement compared to the CG, possibly caused by the improvement in neuroplasticity and eventual reduction in systemic inflammation, but serum results are needed for confirmation.

**Keywords:** Aging; Oxidative stress; Exercise; Neuroinflammation; Neuroplasticity



Title	Short-term methylglyoxal exposure decreases dentritic spine density in the hippocampus of non-diabetic Swiss male mice
Authors	Lima-Sobrinho, JAB <sup>1</sup> ; Cavalcante, KVN <sup>1</sup> ; Carvalho, GA <sup>2</sup> ; Pinto, MCX <sup>2</sup> ; Ferreira-Junior, MD <sup>1</sup> ; Gomes, RM <sup>1</sup>
Affiliations	<sup>1</sup> Laboratory of Endocrine Physiology and Metabolism, UFG, Goiânia, Goiás <sup>2</sup> Laboratory of Neurochemistry and Neuropharmacology, UFG, Goiânia, Goiás
Session	Neurobiologia

Abstract and Keywords Type 2 diabetes is caused by insulin resistance, which leads to hyperglicemia and increased circulating methylglyoxal (MG), a glycotoxin derived from glucose metabolism, and their adducts. MG are the link between diabetes and end-organ injury, mainly through increased RAGE activation and impaired redox status. DM2 are related to impaired neuron metabolism, which leads to decreased synaptic plasticity, neurofibrillary tangle formation as well as increased apoptosis and neurodegeneration. However, little is known about the CNS injury caused by systemic-derived MGO. To test whether exogenous MGO in the absence of hyperglicemia promotes memory impairment and dendritic spines, we used four experimental groups (n=10/group) of male swiss mice: a vehicle-injected group (CO), MG20 and MG60 groups which were injected once a day during 14 consecutive days with MGO (i.p.; 20 or 60 mg/kg respectively), and a STZinjected diabetic group. All protocols were approved by an ethics committee (004/24). Results were different when p-value, calculated by 2-way ANOVA followed by Tukey post-hoc test, was less than or equal to 0.05. At the end of the experimental period, we do not observed differences in the object location test. However, in the novel object recognition test, MG60 animals presented increased discrimination index compared to CO group (CO 37.4  $\pm$  4.6 vs MG60  $56.1 \pm 2.1$  %; p<0.03). In addition, we identified reduced number of golgistained dendritic spines in neurons within the CA1 region of the hippocampus in MG20 and MG60 groups (CO  $5.9 \pm 0.37$  vs MG20  $3.5 \pm 0.50$  vs MG60  $2.9 \pm 0.21$ spines/10  $\mu$ m; p<0.01) despite no differences in STZ group, compared to CO group. Although there are no behavioral effects, the results correlate exposure to MGO and early cellular changes in a neurodegenerative process in the hippocampus. These findings highlight the complexity of the interactions between diabetes, increased glycotoxins and their possible repercussions on neurocognitive functions.

**Keywords:** Type-2 Diabetes; Methylglyoxal; Glycotoxins; Hippocampus; Dendritic Spines.



Title	REAC neuromodulation therapy option for symptoms related to anxiety, depression and stress – A retrospective study
	Clarissa Aires de Oliveira <sup>1,2,3</sup> Eugenio Luigi Iorio <sup>2,3</sup>
Authors	Lara Ferreira Paraiso <sup>1</sup> Foued Salmen Espíndola <sup>1</sup>
Affiliations	<sup>1</sup> Institute of Biotechnology, Federal University of Uberlandia, Uberlandia, MG, Brazil.
	<ul> <li>International Observatory of Oxidative Stress. 84127, Salerno. Italy.</li> <li>University of Uberaba</li> </ul>
Session	Neurobiologia

# Abstract and

Psychophysiological disorders affect a significant portion of the population, underscoring the importance of simple, non-invasive, and effective therapies. Neuromodulation therapy utilizing Radio Electric Asymmetric Conveyer (REAC) technology has demonstrated promising results in treating various pathologies. Therefore, the aim of this study was to evaluate the effects of REAC therapy on symptoms associated with depression, anxiety, and stress. This retrospective study analyzed questionnaires administered to patients with symptoms of depression, anxiety, and stress who underwent REAC therapy. To assess the treatment's effects, patients completed two questionnaires—the Beck Depression Inventory (BDI) and the Depression, Anxiety, and Stress Scale (DASS-21)before and after treatment. The data were compared using the Wilcoxon matched-pairs test, with a significance value of P < 0.05. A total of 100 patients met the inclusion criteria. Analysis of the questionnaires before the neuromodulation therapy revealed that the surveyed sample did not meet the criteria for depression, anxiety, and stress disorders according to the BDI and DASS-21 instruments. However, upon comparing the results after treatment, a significant reduction (P < 0.00) in the total and partial scores of both questionnaires was observed following REAC therapy. The findings of this study align with previous clinical research, demonstrating the efficacy of neuromodulation therapy in addressing neuropsychological and behavioral symptoms. This effectiveness stems from the ability of REAC therapy to generate very low-intensity radio frequency emissions in specific brain areas, thereby optimizing ionic fluxes in areas requiring improvement and enhancing the electrogenic and electrometabolic activity of target cells.

**Keywords:** noninvasive neurostimulation, REAC, psychophysiological disorders, BDI, DASS 21.



Title	Cerebellar development of rat pups is influenced by maternal diabetes during pregnancy
Authors	Fernanda Naves Araújo do Prado Mascarenhas¹; Maria Júlia Neves Ribeiro¹; Natália Ferreira Silva¹; Rener Mateus Francisco Duarte²; Renata Rouland Teixeira²; Foued Salmen Espindola²; Daniele Lisboa Ribeiro¹; Carlos Ueira Vieira²; Renata Graciele Zanon¹.
Affiliations	<sup>1</sup> Institute of Biomedical Sciences, Federal University of Uberlandia (UFU), Uberlandia, MG, Brazil
	<sup>2</sup> Institute of Biotechnology, Federal University of Uberlandia (UFU), Uberlandia, MG, Brazil.
Session	Neurobiologia

#### Abstract and Seywords

Diabetes mellitus is characterized by a dysfunction in insulin production, secretion and/or action of insulin, causing hyperglycemia. When diagnosed during pregnancy, it is called gestational diabetes mellitus (GDM), and the hyperglycemic state affects the developing fetus. The objective was to evaluate the impact of GDM on the development of offspring, focusing on cerebellar morphofunctional aspects. For this, the adult animals obtained by REBIR-UFU, protocol CEUA 056/18, were placed for mating overnight, and the presence of spermatozoa in the females' vaginal wash, indicating the first gestational day (G1), in G5 induced diabetes, intraperitoneally, with Streptozotocin (50 mg/kg), and in G7 evaluated for blood glucose (mg/dL), diabetics when  $\geq$  200 mg/dL. And for comparison we had a control group. After birth (G21), the puppies were evaluated by body mass index (BMI) and blood glucose. After euthanasia at the ages of 7, 30 and 90 days, their encephalons were extracted and evaluated for weight (g), proceeding to the dissection of the cerebellum. This was prepared for histomorphometric, immunofluorescent and target protein expression analyses, data indicative of inflammation, gliosis, oxidative imbalance and cell death. To evaluate the functional impacts, the puppies were subjected to motor behavioral tests. The results showed that puppies from diabetic mothers compared to the control group, had lower BMIs (\*\*\*), combined with low brain weight (\*\*\*), at both ages. Cerebellar histomorphometry revealed the presence of thinner cortical layers (P30\*\*\* and P90\*\*), presence of gliosis by qualitative assessment at both ages, oxidative imbalance (P30\* ROS and FRAP), apoptosis (P30\*\*), and fewer exploratory behaviors indicating a reduced cognition (P90\*\*). Unpaired ttest (\*<0.05 \*\*<0.01 \*\*\*<0.001). We conclude that the hyperglycemic environment during pregnancy can interfere with the cerebellar development of rat pups, impacting functionally in adult life.

**Keywords:** gestational diabetes, hyperglycemia, motor nervous system, cerebellum, development.



"Fronteiras da Ciência: Explorando Novos Horizontes" - 15 a 18 de maio de 2024, Universidade Federal de Uberlândia (UFU), Campus Santa Mônica

Title	Resveratrol prevents offspring's behavioral impairment associated to immunogenic stress during pregnancy
Authors	Ribeiro-Barbosa ER <sup>1</sup> , Duarte RMF <sup>2</sup> , Ferreira FR <sup>3</sup> , Espindola FS <sup>2</sup> , Spini VBMG <sup>1</sup>
Affiliations	<sup>1</sup> UFU Uberlândia, Dpt of Physiological Sciences, Brazil; <sup>2</sup> UFU Uberlândia, Dpt of Biochemistry and Molecular Biology, Brazil; <sup>3</sup> FIOCRUZ Rio de Janeiro, Brazil.
Session	Neurobiologia

# Abstract and Keywords

Multiple pieces of evidence indicated a relation between maternal prenatal immunological stress and an increased likelihood of neurological and psychiatric developmental disorders in the offspring. Therefore, finding ways to protect the embryo from these effects becomes crucial. The neurodevelopmental process is a critical period susceptible to environmental insults, as it involves the maturation of brain circuits and architecture, making them more vulnerable to substances released from maternal inflammation. Investigating polyphenols in the treatment of neuroimmunological diseases offers advantages due to their antioxidant and anti-inflammatory properties. In this study were used male Balb/C mice (CEUA 048/16) and demonstrated a range of behavioral alterations induced by maternal immune activation (MIA) triggered by an antigenic solution derived from the H1N1 virus. These alterations include significant differences in anxious and elevated riskassessment behaviors, increased immobility in the forced swim test, impacts on memory and object recognition, and social changes like autism. RSV administered during gestation from E14 to E19, reveals beneficial effects against the harmful impacts of MIA. This was evidenced by a significant restoration of social behaviors, recognition, and memory, along with anxiolytic and antidepressant effects observed in adult offspring. These findings contribute to novel therapeutic strategies in the prevention of psychiatric disorders arising from neurodevelopmental stressors.

**Keywords:** psychiatric disorders, maternal immune activation, influenza antigens, resveratrol, neurodevelopment.



Title	Psychobiological aspects of working female university students
Authors	Silva, MJF <sup>1</sup> , Gonçalves, CASB <sup>2</sup> , Lemos, VS <sup>2</sup> , Carvalho, ANS <sup>2</sup> , Schincariol, FM <sup>3</sup> , Pena, VV <sup>1</sup> , Narciso, FV <sup>1,2</sup>
Affiliations	<sup>1</sup> Centro Universitário Mário Palmério (UNIFUCAMP), Monte Carmelo/ MG; <sup>2</sup> Centro Universitário IMEPAC, Araguari/MG; <sup>3</sup> Uberlândia Medical Center (UMC), Uberlândia/MG.
Session	Neurobiologia

# and Keywords

Introduction: The present study verifies the challenges faced by women who balance work, studies, and daily responsibilities. Understanding psychobiological aspects is crucial for enhancing the quality of life of female university students with demanding schedules. Objective: This study aims to evaluate the body composition, fatigue sensation, sleep quality, and mood state of working university students, exploring potential gender differences. Methods: The sample comprised 52 working university students, both male and female, with a mean age of  $22.1 \pm 4.1$  years, enrolled in health-related undergraduate courses. Body composition was assessed, and participants completed the Brunel Mood Scale (BRUMS-B), Pittsburgh Sleep Quality Index (PSQI), and Chalder Fatigue Scale (CFS). Results: Our results indicated poor sleep quality among both genders, particularly prominent in women (U = 187.0; p = 0.01). Furthermore, the working female students reported higher levels of fatigue sensation (Fatigue-B: U = 225.0; p = 0.04; CFS: U = 191.5; p = 0.01) compared to men. Conclusion: Consequently, there is a need to investigate the experiences of working female students and implement targeted health policies, alongside physical and psychological interventions, to mitigate fatigue and enhance sleep quality among working university students.

**Keywords:** Sleep quality, Fatigue, Working shift, Students.



Title	Evaluation of neurotrophins in the neuronal regeneration process of the enteric nervous system in Chagas disease
Authors	Michelle Aparecida Ribeiro de Freitas <sup>1</sup> , Enio C. de Oliveira <sup>2</sup> , Axel Brehmer <sup>3</sup> , Alexandre Barcelos Morais da Silveira <sup>4</sup>
Affiliations	<sup>1</sup> Departamento de Parasitologia, ICBIM, Universidade Federal de Uberlândia, Brasil
	<sup>2</sup> Departamento de Cirurgia, Faculdade de Medicina, Universidade Federal de Goiás, Brasil
	<sup>3</sup> Institute of Anatomy I, University of Erlangen–Nuremberg, Erlangen, Germany
	<sup>4</sup> Laboratório de Neurociências, ICBIM, Universidade Federal de Uberlândia, Brasil
Session	Neurobiologia

### Abstract and Keywords

Introduction: The digestive form resulting from Chagas disease is one of the main causes of morbidity and mortality in the chronic phase of the disease. Patients with the digestive form present a series of symptoms related to organ obstruction. Histological analyzes of the affected organs have demonstrated inflammatory lesions of the enteric nervous system (ENS), associated with a large reduction in the number of neurons. Studies using the GAP-43 protein, a marker of neuronal regeneration, suggest that this process of neuronal plasticity is occurring in the colon of patients with chronic infection.

Objectives: This project aims to clarify the relationship between the development of chagasic megacolon and the expression of neurotrophins, substances capable of stimulating the process of nerve regeneration.

Results: Preliminary results indicate a different expression of certain classes of neurotrophins in chagasic patients with megacolon, different from those in uninfected individuals.

Conclusion: We believe that based on the definitive results we are able to understand the development of this pathology and several others that affect the gastrointestinal tract.

**Keywords:** Neurotrophins; regeneration; Enteric nervous system; Chagas disease



Title	Effects of inulin administration on the microbiota and depressive-like behaviors in male C57BL/6J mice born to mothers treated with soluble <i>Toxoplasma gondii</i> antigen (STAg)
Authors	Victória Leandra Nunes de Assis <sup>1</sup> ; Ágatha Djiullya de Miranda Feijão <sup>1</sup> ; Ana Luiza Silva Oliveira <sup>1</sup> ; Frederico Rogério Ferreira <sup>2</sup> ; Simone Ramos Deconte <sup>1</sup> ; Angelica de Oliveira Gomes <sup>3</sup> ; Vanessa Beatriz Monteiro Galassi Spini <sup>1</sup>
Affiliations	<sup>1</sup> Federal University of Uberlandia, Dept of Physiology, Brazil. <sup>2</sup> Oswaldo Cruz Institute (FIOCRUZ), Rio de Janeiro, Brazil. <sup>3</sup> Federal University of Triangulo Mineiro, Dept of Structural Biology, Brazil.
Session	Apresentação Pôster

#### Abstract and Keywords

Studies have shown the influence of the microbiota on the development and functioning of the central nervous system, with dysbiosis considered a possible cause of mental disorders, such as depressive disorders. It is known that maternal immune activation (MIA) triggers inflammatory events associated to behavioral disorders in the offspring. Moreover, healthy microbiota can protect the offspring from such changes. Inulin is a soluble dietary fiber, considered a prebiotic because it modulates the microbiota and promote the growth of beneficial bacteria. This study aimed to investigate whether maternal immune activation using soluble Toxoplasma gondii antigens (STAg) in pregnant female C57BL/6J mice could cause neural changes in adult male offspring, resulting in depressive-like behaviors and whether inulin administered during lactation would modulate its microbiota, preventing such behavioral changes. Pregnant females were divided into two groups, one receiving STAg and the other PBS, both intraperitoneally at E16. The puppies received oral treatment, water or inulin, from the 3rd to the 21st day of life. The adult offspring were subjected to the "Splash Test" behavioral test and grooming behavior was quantified, as a measure of self-care and motivational behavior. The immunogenic challenge of the mother with STAg decreased the number of grooming in the offspring (11.33 + 0.96) in relation to the control (19.71 + 2.71; p>0.05), although it was not significant. Inulin treatment increased the number of grooming in the offspring of dams treated with STAg (31.15 + 2.89) compared to offspring that received water (11.33 + 0.96; p<0.05) and decreased the latency to initiate grooming behavior (6.60 + 1.61; p < 0.01) in relation to offspring born to unchallenged mothers (27.26 + 8.3), suggesting a possible protective effect of inulin on depressive-like behaviors. CEUA: 23117.037394/2022-63.

**Keywords:** maternal immune activation, microbiota, inulin, splash test



Title	Comparison between BACE-APP and β-Amyloid models for the study of Alzheimer's disease in Drosophila melanogaster
Authors	Joyce da Silveira Pereira Matheus Henrique Silva Serena Mares Malta Alexandre Souza Marquez Carlos Ueira-Vieira
Affiliations	Federal University of Uberlândia, Uberlândia, Brazil
Session	Neurobiology

### and Keywords

Alzheimer's disease is a neurodegenerative disease that is irreversible and associated with aging and genetics. The disease is caused by the formation of  $\beta$ amyloid plaques, which are formed when the amyloid precursor protein is cleaved by  $\beta$ -secretase. This study aims to investigate two ways to reproduce the amyloidogenic pathway that occurs in humans using transgenic Drosophila melanogaster as a study model. The flies were obtained from the Bloomington stock center and maintained in LABGEN-UFU with a standard culture medium. Crossbreeding was performed between strains BL#3605(control), BL#8765(Elav-Gal4), BL#33797(UAS-APP,UAS-BACE), and BL#64216(UAS-β-Amyloid), aiming to generate flies with the genotypes w\*;Elav-Gal4;UAS-β-Amyloid and w\*; Elav-Gal4; UAS-BACE, UAS-APP, and the control goup: w\*; Elav-Gal4;+. The RING (Rapid iterative negative geotaxis) test was conducted to assess the locomotor ability of these animals (N=30/group, 15 male and 15 female). The test evaluates the percentage of flies that climbed 5cm or more in 4s and 18s and was performed at the ages of 2,5,10,15, and 17 days posteclosion. GraphPad Prism10.2 software was used to perform statistical analysis. The affected group w\*;ELAV-Gal4;UAS-APP,UAS-BACE showed a significant decrease in climbing ability only at the age of 5 days when compared to the control in the 4s analysis p=0.03 (20.86±7.39). The group w\*;Elav-Gal4;UASβ-Amyloid when compared to the control at 4s, exhibited a significant decrease at the ages of 2-4 days p=0.0006 (52±5.8) and 15-17 days p=0.04 (6±5.8). At 18s, significance was observed only at the age of 15-17 days p<0.0001 (32.8±5.7). There was no significant difference among the control and affected groups at other ages (p>0.05). These results indicate that w\*;Elav-Gal4;UAS-β-Amyloid group had a greater reduction in locomotor activity, suggesting that this model may be more suitable to simulate Alzheimer's disease.

**Keywords:** Genetics, Neurobiology, Alzheimer, *Drosophila melanogaster*,  $\beta$ - amyloid, RING,  $\beta$ -secretase.



Title	Effects of inulin administration on the microbiota and behavioral changes related to depressive-type disorder in lactant female mice born to mothers treated with soluble <i>Toxoplasma gondii</i> antigen (STAg)
Authors	Agatha Djiullya de Miranda Feijão <sup>1</sup> ; Victória Leandra Nunes de Assis <sup>1</sup> ; Ana Luiza Silva Oliveira <sup>1</sup> ; Simone Ramos Deconte <sup>1</sup> ; Frederico Rogério Ferreira <sup>2</sup> ; Angelica de Oliveira Gomes <sup>3</sup> ; Vanessa Beatriz Monteiro Galassi Spini <sup>1</sup>
Affiliations	<sup>1</sup> Federal University of Uberlandia, Dept Physiology, Uberlândia Brazil <sup>2</sup> Oswaldo Cruz Fundation (FIOCRUZ), Rio de Janeiro, Brazil <sup>3</sup> Federal University of Triangulo Mineiro, Dept Structural Biology, Uberaba Brazil
Session	Poster Presentation

#### Abstract and Kevwords

Studies have demonstrated the influence of the intestinal microbiota on mental health through different mechanisms, such as regulation of pro-inflammatory cytokines and production of neurotransmitters with dysbiosis associated with the pathophysiology of mental disorders such as depression and anxiety. Maternal Immune Activation (MIA) is an experimental model in which a maternal immune challenge induces altered behaviors in offspring through several mechanisms, including changes in the gut microbiome. In this context, inulin a soluble fiber that acts as a prebiotic, has been considered an important modulator of the microbiota, with potential therapeutic use. This study aimed to investigate whether the immunogenic challenge of C57BL/6 female mice with soluble Toxoplasma gondii antigens (STAg) induces depressive-like behavior in adult offspring and whether inulin, administered during lactation, promotes neuroprotection by modulating the microbiota. Pregnant females were divided into two groups, one receiving intraperitoneal injection of STAg at E16 and the other receiving PBS. Female pups received inulin or water, via gavage, from the 3rd to the 21st day of life. When adults were subjected to the splash test, grooming behavior was quantified as a measure of self-care and motivational behavior. The offspring from mothers treated with STAg and who received inulin showed an increase in the number of grooming (35  $\pm$  1.2; p<0.001) in relation to the offspring that received water (28.4  $\pm$  2; p<0.005) demonstrating a possible protective effect of inulin on depressive-like behaviors. Keywords: maternal immunoactivation, Toxoplasma soluble antigens, inulin, splash test



Title	Differentially expressed brain proteins after high-refined carbohydrate diet and glibenclamide treatment
	Sttefany Nayara Sant'ana de Faria <sup>1</sup> Paula Viana Sene dos Santos <sup>1</sup>
Authors	Stêfany Bruno de Assis Cau <sup>2</sup>
	Luciana Karen Calábria <sup>1</sup>
Affiliations	<sup>1</sup> Federal University of Uberlândia, Ituiutaba-MG, Brazil.
	<sup>2</sup> Federal University of Minas Gerais, Belo Horizonte-MG, Brazil.
Session	Neurobiologia

# Abstract and Keywords

The effects of sugar-rich diet are still unknown about the expression of proteins in the brain. In order to investigate the differential expression of proteins on mice brain after treated with high-refined carbohydrate diet, the animals were fed either a standard rodent diet or a diet rich in refined carbohydrates for eight weeks, and with glibenclamide (sulphonylurea indicated in the treatment of hyperglycemia and diabetes) for four weeks (CEUA/UFMG protocol 369/2016). The brains dissected were homogenized and the supernatant was subjected to total protein assay. Aliquots of 10 µg/µL were analyzed on 12.5% SDS-PAGE and the relative molecular mass was calculated of the bands. The electrophoretic profile of the samples revealed nine proteins, but no significant difference compared fed a high-refined carbohydrate diet and control. The differential bands between control group and the treated with glibenclamide were analyzed in averages and by the teste t using the software BioEstat and values of p < 0.05were considered significant. The proteins estimated in 44.8, 42.2 and 39.8 KDa are more significant expressed in the brain of the mices treated with glibenclamide. These differential proteins are candidates for future proteomic analysis. The proteins were predicted using in silico analysys and mostly they are related to the brain's energy metabolism for carbohydrates, lipids and amino acids. The short amount of exposure time of the animals to the diet may justify the absence of protein alterations and the use of more accurate methodologies is necessary to identify the differential brain expression based on the highrefined carbohydrate diet, in order to clarify the molecular mechanisms that may be altered in the studied condition.

Keywords: Sulphonylurea. Isocaloric diet. Electrophoretic profile.



Title	Analysis of runners' performance in the 2,400m Track Test following HIIT treadmill intervention
	Robson da Silva Medeiros
	Thiago Montes Fidale
	Flander Diego de Souza
	Fanny Gonçalves de Lima
	Tathyane Fonso da Silva
Authors	Jefferson Fernandes de Souza
	Guilherme Gularte de Agostini
	Elmiro Santos Resende
Affiliations	<sup>1</sup> Federal University of Uberlândia, Uberlândia-MG, Brazil
	<sup>2</sup> Federal University of Catalão, Catalão-GO, Brazil
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Session	Physical Training Responses

#### Abstract and Keywords

**Introduction:** High-intensity interval training (HIIT) is characterized by short and repeated periods of high-intensity stimuli, with recovery intervals between them. The study aimed to analyze the effects of 4 weeks of HIIT on a treadmill on the performance of runners in the 2400m track test.

**Methods:** The study was approved by the UFU Research Ethics Committee (CAE:13624419.2.0000.5152). Eleven runners aged 18 to 39 participated. Before and after HIIT, participants performed a 2400m track test on an asphalt track, aerobic capacity (VO2 max) was measured, and pre- and post-test lactate levels were measured. A maximal incremental treadmill test was performed to identify maximum aerobic velocity (MAV), used to establish individual HIIT intensities. Training consisted of low and high-intensity stimuli in a 1:1 ratio, with the low-intensity stimulus velocity set at 50% of MAV. Two weekly sessions of HIIT were conducted, with maximal stimuli at increasing intensities in weeks 1 to 3 (90%, 100%, and 110% of MAV). Week 4 maintained 110% of MAV, with 50% of the average stimuli from week 3. Statistical analysis was performed using Student's t-test, and significance was established for p<0.05.

**Results:** Significant differences were found in running time for the 2400 meters (Min/Sec/Cent), pre (08:26.8  $\pm$  00:50.2) vs. post (08:12.8  $\pm$  00:49.3) (P=0.00); VO2 max (mL/kg/min), pre (57.38  $\pm$  5.40) vs. post (58.89  $\pm$  4.84) (P=0.00); MAV (km/h), pre (17.21  $\pm$  1.62) vs. post (17.66  $\pm$  1.45) (P=0.00); and lactate (mmol/L) pre (13.53  $\pm$  4.5) vs. post (10.40  $\pm$  3.0) (P=0.02).

**Conclusion:** We conclude that HIIT performed on a treadmill was effective in improving runners' performance in the 2400m track test, with increased VO2 max and reduced running time over the distance. There was also a lower concentration of lactate in the post-intervention test.

**Keywords:** Runners, HIIT, Performance.



Title	Heart rate variability during maximum incremental treadmill test
Authors	Felipe Farnesi Ribeiro Borges <sup>1</sup> ; Fanny Gonçalves de Lima <sup>2</sup> ; Daniel Martins Lúcio <sup>2</sup> ; Rafael Lima Santos <sup>2</sup> ; Robson da Silva Medeiros <sup>2</sup> ; Jefferson Fernandes de Sousa <sup>1</sup> ; Elmiro Santos Resende <sup>2</sup> ; Adriano Alves Pereira <sup>1</sup> ; Frederico Balbino Lizardo <sup>3</sup> ; Thiago Montes Fidale <sup>1,4</sup>
Affiliations	<sup>1</sup> Post-graduation Program in Biomedical Engineering, Federal University of Uberlândia, Uberlândia, MG, Brazil. <sup>2</sup> Laboratory of Experimental Medicine, Federal University of Uberlândia, Uberlândia, MG, Brazil. <sup>3</sup> Laboratory of Kinesiological Electromyography, Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, MG, Brazil. <sup>4</sup> Academic Unit of Biotechnology, Federal University of Catalan, Goias, Brazil.
Session	Respostas de Treinamento Físico

## Abstract and Keywords

Running requires specific training and consideration of physiological variables, especially for elite athletes. Heart rate variability (HRV) analysis is crucial for understanding autonomic responses, assessing cardiovascular health, and adjusting training loads. HRV is essential for evaluating performance and recovery, reflecting physical adaptations, and indicating fatigue and stress. The study was approved by the Ethics Committee for Human Research at UFU, opinion number: 3,397,582, and aimed to analyze the HRV of runners during a maximal incremental treadmill test. Eleven runners participated, with a mean age of  $31 \pm 5.78$  years and personal best records below 21 minutes in official 5 Km races. HRV was recorded using the POLAR® RS800cx monitor, sampling rate of 1000 Hz, and R-R intervals were analyzed by Polar Pro Trainer 5® software. The test started at a speed of 10 km/h, with load increments of 1 km/h every 2 minutes, without breaks between stages until voluntary exhaustion was reached. Analyses were performed using GraphPad Prism7 software, using the One Way ANOVA test, with Tukey and Dunn post hoc analyses, significant at p < 0.05. Results revealed statistically significant differences in HRV in the parameters: PNS, SNS, and Baevsky Stress Index at rest (5 minutes), at each stage of the test, and during recovery. A predominance of parasympathetic activity over sympathetic activity was observed, indicating stress during maximal exercise load increments. Our conclusions suggest that HRV is useful for assessing physical capacity and comparing conditioning in healthy individuals. Participants showed parasympathetic predominance at rest, possibly due to street running practice. HRV variables reflect physiological changes during exercise, showing that at high intensities parasympathetic activities decrease while sympathetic stress increases, partially remaining even after 6 minutes of recovery.

**Keywords:** HRV, Autonomic Regulation, Autonomic Nervous System, Running.



Title	Effect of high-intensity interval aquatic exercise on heart rate variability in postmenopausal hypertensive women
Authors	Tássia Magnabosco Sisconeto <sup>1</sup> ; Caroline Pereira Garcês <sup>1</sup> ; Ana Clara Ribeiro Cunha <sup>1</sup> ; Juliana Cristina Silva <sup>1</sup> ; Igor Moraes Mariano <sup>1</sup> ; Tállita Cristina Ferreira de Souza <sup>1</sup> ; Ana Carolina Kanitz <sup>2</sup> ; Guilherme Morais Puga <sup>1</sup> .
Affiliations	<sup>1</sup> Universidade Federal de Uberlândia, Uberlândia-MG, Brasil. <sup>2</sup> Universidade Federal do Rio Grande do Sul, Porto Alegre -RS, Brasil.
Session	Pôster

#### Abstract and Keywords

After menopause, there is a reduction in Heart Rate Variability (HRV), particularly notable in hypertensive women. Aerobic exercise is recognized to augment HRV, with aquatic exercise being widely favored due to the immediate physiological alterations induced by immersion in heated water (28-32°C). Consequently, it is hypothesized that intense exercise performed in heated water might amplify the effects of immersion. The objective of this study was to assess the HRV responses of hypertensive postmenopausal women following a single session of highintensity interval aquatic exercise (HIIE). To achieve this, a randomized crossover clinical trial was conducted involving 22 hypertensive postmenopausal women (mean age: 58±4.5 years; blood pressure: 116/72 mmHg; resting heart bpm) approved the Ethical Committee (CAAE: by 58033422.4.0000.5152). Two sessions were administered: HIIE and control (CON), both conducted in the same pool with an average temperature of 31°C. The HIIE session comprised a 3-minute warm-up, 2 sets of 8 minutes each (alternating 1 minute of intense activity and 1 minute of light activity) with a 2minute rest interval between sets, followed by a 3-minute cool-down. The CON session consisted of 24 minutes of static stretching exercises performed while immersed. Heart rate was assessed at rest using a Polar heart rate monitor before and after each session. HRV was calculated using both time domain (Root Mean Square of Successive Differences, RMSSD; Standard Deviation of all NN intervals, SDNN) and frequency domain (High Frequency, HF; Low Frequency, LF; LF/HF ratio) indices. Analysis revealed no significant interaction between sessions and time points for the RMSSD (p=0.26), SDNN (p=0.13), LF (p=0.82), HF (p=0.83), or LF/HF (p=0.83) indices. In conclusion, a single session of interval aquatic exercise did not induce changes in heart rate variability in hypertensive postmenopausal women.

**Keywords:** Heart Rate; Exercise; Postmenopause; Hypertension



Title	Effect of the execution order of acute aerobic and resistance exercise on heart rate variability in postmenopausal women with type 2 Diabetes
	Caroline Pereira Garcês Juliana Cristina Silva
Authors	Tássia Magnabosco Sisconeto Igor Moraes Mariano
	Ana Clara Ribeiro Cunha Nadia Carla Cheik Guilherme Morais Puga
Affiliations	Federal University of Uberlândia, Uberlândia, Brazil
Session	Oral

Abstract and Keywords	Postmenopausal women with Type 2 Diabetes (T2D) have a higher risk of cardiovascular complications due to various factors, such as decreased heart rate variability (HRV). It is known that aerobic and resistance exercises treat and prevent cardiovascular complications in individuals with T2D, however, the optimal order for a combined aerobic and resistance exercise session is unknown. Thus, this study compared the execution order of a combined aerobic and resistance exercise session on HRV in postmenopausal women with T2D. A clinical, crossover, and randomized trial included 15 postmenopausal women (60 $\pm$ 5.9 years, HbA1c 6.8 $\pm$ 0.5, 10.8 $\pm$ 5.1 years in menopause) with T2D in three experimental conditions: (I) 30 minutes of aerobic exercise (50 to 60% of reserve heart rate) + 30 minutes of resistance exercise (3 sets of 8-12 maximum repetitions) (AR); (II) Resistance exercise + aerobic exercise (RA); (III) Resting session (CON). This study was approved by the Ethics Committee (CAAE: 57236522.1.0000.5152). Heart rate was collected pre-exercise and at 10, 30, and 50 minutes post-exercise using a chest strap heart rate monitor (POLAR® H10, Kempele, Finland). HRV indices were calculated using time domain (SDNN and RMSSD) and frequency domain (LF, HF, and LF/HF) analysis. For statistical analysis, the two-way repeated measures ANOVA test (SPSS® 23.0) was used, with a significance level set at p < 0.05. After 10 and 30 minutes of exercise, the AR and RA sessions exhibited lower values in the SDNN and RMSSD indices compared to the CON session (p < 0.05). In the SDNN index, the RA session also demonstrated a statistically lower value compared to the CON session 50 minutes after exercise (p = 0.04). For the frequency domain indices HF, LF, and LF/HF, no significant difference was observed among the experimental conditions (p > 0.05). In conclusion, both exercise sessions induced acute autonomic stress, regardless of the execution order.
	<b>Keywords:</b> Exercise; Autonomic Nervous System; Postmenopause; Diabetes Mellitus



Title	Effects of online vs. face-to-face mat Pilates training in patients with long COVID
Authors	Ana Clara Ribeiro Cunha Juliana Cristina Silva
	João Luiz Rezende Nascimento
	Caroline Pereira Garcês Tássia Magnabosco Sisconeto Igor Morais Mariano
	Thulio Marquez Cunha
	Guilherme Morais Puga
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	Respostas de treinamento físico

## and Keywords

COVID-19 can lead to a variety of acute and chronic adverse health effects in patients, often known as long COVID. Engaging in physical exercise emerges as a pivotal strategy for managing COVID-19 symptoms, encompassing muscle weakness, physical constraints, exercise-induced fatique, and quarding against functional deterioration exacerbated by the syndrome's progression. The aim of the study was to compare the impacts of online versus face-to-face Mat Pilates interventions for individuals with long COVID. A randomized clinical trial was executed, involving 49 volunteers, comprising 5 men and 44 women (with a mean age of 52±5.8 years and BMI of 32±8.9 kg/m2), who were randomly allocated to the face-to-face Pilates group (n=15), online group (n=16), and control group (n=18). The project received approval from the Ethics Committee on Human Research of the Federal University of Uberlândia (CEP-UFU) - CAAE: 57164322.3.0000.5152. Both face-to-face and online group training sessions were conducted three times weekly during 12 weeks, with each session comprising a 5-minute warm-up, a 40-minute main segment, and a 5-minute cool-down. The Functional capacity were evaluated before and after the intervention: Time up and go test (TUG), upper limb endurance, flexibility, handgrip isometric strength, trunk flexion and extension isometric strength, and a 6-minute walk test. Generalized Estimated Equation GEE were used to compare groups an time effects. Upper limb endurance (16±4 to 23±4), trunk flexion isometric strength (06.6±3.0 to 12.7±6.6) and 6-minute walk test (463±73 to 514±39) improved (p<0.05) only in Face-to-face group after the intervention. These findings indicate that individuals dealing with long COVID can utilize this training approach to improve their impaired functional abilities.

**Keywords:** Post-COVID Conditions, training pilates, functional variables



Title	Physical training responses and respiratory muscle strength in elderly – a pilot study
Authors	Natália Rabelo Rocha, Ingrid Alexia Duarte Mendonça, Fernanda Borges André, Angelo Piva Biagini, Carlos Fernando Ronchi
Affiliations	Universidade Federal de Uberlândia
Session	Respostas de Treinamento Físico

## and Keywords

Aging and life expectancy tend to increase gradually over the next few years. Which causes several changes in body, mentally and functional capacities, leading to functions loss, causing fragility. One of these changes occurs in the respiratory system, affecting muscles mechanics physiological with a reduction in inspiratory and expiratory strength, reflecting changes in maximum inspiratory and expiratory pressures (PImax and PEmax). By functional decrease and inactivity, these elderlies tend to have greater dependence, which is a major risk factor. Physical activity is crucial to avoid it, preventing pathologies and promoting functional independence. Walking is an easy activity to perform and well accepted by elderlies. Therefore, the objective of this study was to evaluate whether physical activity by walking interferes in respiratory muscles strength, which could improve respiratory capacity in elderlies.

We have evaluated seven volunteers, five women and two men over 60 years. It was assessed respiratory muscle strength by maximal respiratory pressures before and after intervention, using an aerobic physical activity training. Before starting the protocol, it was carried out a 6-minute walk test, which was used as a parameter for each volunteer to progression increase. Training program lasted nine weeks, by walking progression every three weeks until the end of the intervention. Progression was carried out individually, respecting each volunteer limit and condition, and in accordance with scale effort.

There was an increase in maximal respiratory pressures, with Pimax 70,714 cmH $_2$ O and 74,286 cmH $_2$ O, as well as, in Pemax 82,857 cmH $_2$ O and 97,143 cmH $_2$ O (p>0,05), before and after respectively. However, there was no statistical difference, probably due to the number of volunteers evaluated. Therefore, it is possible to conclude that walking has beneficial effects in elderlies, however, more research with greater number of volunteers is needed to confirm it and understanding walking effects on respiratory muscle strength improvement in this population. Ethical committee: 5.568.892. **Keywords:** elderly, aging, walking, aerobic activity, respiratory muscle strength.



Title	Impact of pre-chemotherapy combined training on extracellular vesicle expression in breast cancer women
Authors	Junior GDS <sup>1</sup> , Telles GD <sup>2</sup> , Vechin FC <sup>2</sup> , Ferreira MLV <sup>1</sup> , Araujo RB <sup>1</sup> , Buzaglo GBB <sup>1</sup> , Conceicao MS <sup>1</sup>
Affiliations	<sup>1</sup> Health Sciences Program, Sao Francisco University, Braganca Paulista, Brazil <sup>2</sup> School of Physical Education and Sport, University of Sao Paulo, Sao Paulo, Brazil
Session	Physical Training Responses

#### Abstract and Kevwords

Physical exercise is increasingly recognized as a pivotal factor in mitigating tumo proliferation, particularly in breast cancer. Recent studies underscore its direct impact on reducing tumor growth and metastasis, but its molecular mechanisms remain unclear. Extracellular vesicles (EVs), crucial in intercellular communicatio can transport molecules like microRNAs (miRNAs), influencing gene expression and serving as potential intervention targets in cancer processes. Accordingly, w characterized EVs expression in 12 breast cancer patients who performed a bout of combined resistance and aerobic exercise before treatment initiation. This experimental design allows us to isolate the effects of the exercise. Blood sample were collected before and after exercise and EVs were isolated from the platelet- free plasma by size exclusion chromatography (SEC) using the the IZON AFC and the qEV10/35nm columns in accordance with the manufacturer's quidelines (IZO Science). After isolation, EV-rich fractions (1-3) were combined and concentrated to a volume of 500µl using Amicon Ultra-15 Centrifugal Filter Units. The resultin concentrated sample was then analyzed for size/diameter and quantity using the NanoSight NS300. There were  $4.26 \times 10^{12} + /-2.99 \times 10^{11}$ particles/ml, with 160.6+/

2.4 nm diameter in the EV-rich pool before exercise. Following the exercise session, the quantity increased to  $5.22 \times 10^{12} + /-3.58 \times 10^{11}$  particles/ml, with 162.8 + /-2.0 nm diameter. These findings suggest that a combined resistance an aerobic exercise bout induces the systemic release of small EVs in breast cancer patients. Investigating post-exercise EV profiles could unveil valuable biomarkers or therapeutic targets, optimizing breast cancer treatment strategies. Further exploration of the specific molecules within these exercise-induced EVs and their roles in breast cancer biology could provide valuable insights into how exercise influences the disease. CAAE no: 39031720.5.0000.5404. Ethics Committee and Research Report: 4.978.580.

**Keywords:** exercise; extracellular vesicles; breast cancer.



Title	Effects of physical training on the central expression of dopaminergic D2 receptors
Authors	Lucas Rios Drummond <sup>1,2</sup> Quezia Teixeira Rodrigues <sup>1</sup> Helton Oliveira Campos <sup>1,3</sup> Laura Hora Rios Leite <sup>4</sup> Cândido Celso Coimbra <sup>1</sup>
Affiliations	<sup>1</sup> Departamento de Fisiologia e Biofísica, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Belo Horizonte/MG, Brasil <sup>2</sup> Departamento de Educação Física, Universidade do Estado de Minas Gerais, unidade Divinópolis, Divinópolis/MG, Brasil <sup>3</sup> Departamento de Ciências Biológicas, Universidade do Estado de Minas Gerais, unidade Ubá, Ubá/MG <sup>4</sup> Departamento de Fisiologia, Instituto de Ciências Biológicas, Universidade Federal de Juiz de Fora, Juiz de Fora, MG, Brasil
Session	Respostas de Treinamento Físico  Apresentação Pôster

#### Abstract and Keywords

The present study proposed to verify the expression of dopaminergic D2 receptors in the caudate putamen (CPU), in the preoptic area (APO), and the amygdala (Amy) in response to aerobic physical training. These regions are involved in motor control, body temperature control and the reward system, which are important for performance during physical exercise. For this, male Wistar rats aged four weeks were used, divided into: untrained group (NT, n=8) and trained group (T, n=8). Physical training was performed on a treadmill for 8 weeks/5 days a week, reaching a speed of 18 m.min-1 and a duration of 60 minutes. The animals were euthanized 48 hours after the last physical exercise session (basal state), and the brain was removed to quantify the protein expression of the dopaminergic D2 receptors receptor in the CPU, APO, and Amy by western blot. Experimental procedures were approved by the CEUA-UFMG (#70/2022). The results showed that trained animals showed a 52% increase in protein expression of dopaminergic D2 receptors in Amy (NT:  $1.00 \pm 0.12$  vs. T:  $1.52 \pm 0.15$  a.u.; p<0.05). On the other hand, physical training did not change the expression of dopaminergic D2 receptors in the CPU (NT:  $1.00 \pm 0.15$  vs. T:  $1.32 \pm 0.13$  a.u.; p>0,05) and POA (NT:  $1.00 \pm 0.29$  vs. T:  $0.98 \pm 0.19$  a.u.;

p>0.05). Based on the results achieved, it can be concluded that aerobic physical training increases the expression of dopaminergic D2 receptors in the AMY but does not modify the expression of these receptors in the CPU and POA.

**Keywords:** Exercise, Dopamine, DRD2 receptor, Caudate putamen, Preoptic area, Amygdala.



Title	The effect of multicomponent training on executive functions in elderly individuals with Alzheimer's disease
Authors	Dayanne Christine Borges Mendonça Denise Rodrigues Fernandes Caroline Pereira Garcês Guilherme Morais Puga
Affiliations	UFU - Uberlândia/Brasil
Session	Oral

## and Keywords

Currently, there is a gradual increase in the number of elderly individuals diagnosed with Alzheimer's disease worldwide. This condition leads to cognitive impairments, including a decline in executive functions among the elderly. In this regard, physical exercise is seen as a promising non-pharmacological treatment for Alzheimer's disease. Thus, this study aimed to evaluate the effect of 12 weeks of multicomponent training on the executive functions of elderly individuals diagnosed with Alzheimer's disease. The study involved 8 elderly participants, including 3 men and 5 women, aged over 65 years (mean age  $82 \pm 5.9$  years), diagnosed with Alzheimer's disease in the mild to moderate stage. The participants performed multicomponent training twice a week on nonconsecutive days for 60 minutes each session. This study was approved by the Ethics Committee (CAEE:52595021.5.00005152). Multicomponent training consists of a combination of aerobic, strength, balance, agility, coordination, and flexibility exercises. The executive function of the elderly participants was assessed using the Frontal Assessment Battery, a specific battery for evaluating executive functions, both pre- and post-intervention. Paired samples t-test was used for statistical analysis in SPSS® software. The results showed that the elderly participants did not demonstrate improvement in the Frontal Assessment Battery (p = 0.51) after 12 weeks of multicomponent training. Therefore, the Multicomponent Training protocol was not effective in improving executive functions in elderly individuals with Alzheimer's disease.

**Keywords:** Alzheimer's disease; executive functions; multicomponent training.



Title	Transcriptomic analysis reveals IGFBP6 as a potential inhibitor of fish muscle growth
Authors	Rafaela Alves Ribeiro <sup>1</sup> ; Erika Stefani Perez <sup>2</sup> ; Bruna Tereza Thomazini Zanella <sup>2</sup> ; Daniel Garcia de la serrana <sup>3</sup> ; Maeli Dal-Pai-Silva <sup>2</sup> ; Bruno Oliveira da Silva Duran <sup>1</sup>
Affiliations	<sup>1</sup> Instituto de Ciências Biológicas (ICB), Universidade Federal de Goiás (UFG), Goiânia, Brasil. <sup>2</sup> Instituto de Biociências (IB), Universidade Estadual Paulista (UNESP), Botucatu, Brasil. <sup>3</sup> Faculty of Biology, University of Barcelona, Espanha.
Session	Poster presentation

#### Abstract and Keywords

Skeletal muscle is abundant in fish and has high economic value for meat production in aquaculture or in vitro systems. Different factors act on fish muscle growth, but the molecular components and pathways elicited are not yet fully elucidated. Omics approaches have been improving studies on the skeletal muscle, unraveling molecular networks and allowing the discovery of biomarkers. In addition, cell cultures systems contribute to the understanding of muscle cells, which grow in a controlled environment, without systemic variables. Our objective was to identify and evaluate the genes modulated by pro-growth signals in (Piaractus mesopotamicus) and gilthead sea bream (Sparus aurata) muscle cells. Cells were cultured in free medium (CTR), amino acid-rich medium (AA) or medium with insulin-like growth factor 1 (IGF1) (n=3 cell cultures; CEUA 026/21 and CEEA 205/19). After RNA-Seq, bioinformatic analyzes of the transcriptome were conducted and we found different classes of IGF binding proteins (IGFBPs) modulated by the treatments. We identified two main clusters of igfbps: up-regulated by AA and IGF1 (stimulatory); downregulated by AA and IGF1 (inhibitory). Interestingly, the igfbp6 showed different copies and was distinctly modulated by AA and IGF1. Igfbp6a was specifically inhibited by IGF1 in pacus, leading to myogenesis, energy production, IGF signaling, and reduced degradation/fibrosis. Igfbp6b was inhibited by both AA and IGF1 in sea bream, leading to muscle regeneration, organization of sarcomeres, prevention of atrophy, TGFβ signaling and reduced apoptosis. We also validate igfbp6b in Nile tilapia (Oreochromis niloticus) (CEUA 001/24), one of the most produced fish, and we observed an increased expression during fasting, a period of intense muscle corroborating our data. In conclusion, our work characterizes igfbp6 as a negative regulator of muscle growth, could be silenced/repressed in studies aiming to improved fish muscle mass.

Keywords: Skeletal muscle; Fish; Omics; Igfbp.



Title	Development of a sustainable, portable, fast and non- invasive biophotonic platform associated with artificial Intelligence for salivary detection of <i>Helicobacter pylori</i>
Authors	Ghabriel Honório da Silva¹; Marco Fidel Guevara-Veja¹; Nagela Bernadelli de Sousa Silva²; Marcelo Augusto Garcia-Júnior¹; Luiz Ricardo Goulart³†; Thulio Marquez Cunha⁴; Carlos Henrique Gomes Martins²; Murillo Guimarães Carneiro⁵; Robinson Sabino-Silva¹
Affiliations	¹Innovation Center in Salivary Diagnostic and Nanobiotechnology, Department of Physiology, Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlândia, Brazil.  ²Antimicrobial Testing Laboratory, Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlândia, Brazil.  ³Institute of Biotechnology, Federal University of Uberlandia, Uberlândia, Brazil. faculty of Computing, Federal University of Uberlandia, Uberlândia, Brazil.  † In Memoriam.
Session	Optical and Mechanical Technologies for Health

#### Abstract And Keywords

Helicobacter pylori (H. pylori) infection can increase the risk of peptic ulcers and gastrointestinal neoplasms. H. pylori detection in gastric epithelial tissue collected by esophagogastroduodenoscopy (EGD) is an invasive, expensive, and complex execution procedure, reducing access for isolated populations. H. pylori detection by Urea Breath Test (UBT) presents high cost with limited access in low- and middle-income countries. In this context, it is critical to develop novel alternative non-invasive platforms for the portable, fast, and reagent-free detection of H. pylori. In the work carried out in this research, we used attenuated total reflectance Fourier-transform infrared spectroscopy (ATR-FTIR) supported by Machine Learning algorithms to identify infrared vibrational modes of H. pylori diluted in human saliva. Experimental procedures approved by the Ethical Committee UFU #58115222.5.0000.5152). To perform it, saliva was diluted in 4 different H. pylori concentrations (108 CFU/mL, 107 CFU/mL, 106 CFU/mL, and 105 CFU/mL). Then, H. pylori infected saliva was applied to ATR-FTIR spectroscopy to perform a reagent-free, fast, and sustainable analysis of spectral signatures to identify unique vibrational modes. The obtained spectra were applied to Linear Discriminant Analysis (LDA) and Support Vector Machine (SVM) algorithms to perform the H. pylori detection. The best discrimination performances obtained from the concentrations ranged from 85% to 94% of accuracy, reaching 89% for 108 CFU/mL, 93% for 107 CFU/mL, 94% for 106 CFU/mL, and 85% for 105 CFU/mL. The data demonstrate that this proof-ofconcept study has significant potential for the non-invasive detection of H. pylori using a biophotonic platform supported by artificial intelligence without the use of reagents with human saliva samples obtained by self-collection.

**Keywords:** ATR-FTIR; Helicobacter pylori; Gastrointestinal infection; screening test; salivary diagnostics; saliva, non-invasive test; machine learning algorithms



Title	Development and validation of a biophotonic platform for alivary diagnosis of diabetes mellitus associated with artificial intelligence
Authors	Douglas Carvalho Caixeta <sup>1</sup> ; Ricardo Rodrigues <sup>1</sup> ; Rayany Cristina de Souza <sup>1</sup> ; Marco Fidel Guevara Vega <sup>1</sup> ; Mariana Araújo Costa <sup>1</sup> Camila Moreira de Andrade <sup>1</sup> Ricardo Rodrigues Filho <sup>1</sup> ; Murillo Guimaraes Carneiro <sup>2</sup> ; Robinson Sabino-Silva <sup>1</sup>
Affiliations	<sup>1</sup> Innovation Center in Salivary Diagnostic and Nanotheranostics, Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, Brazil <sup>2</sup> Faculty of Computing, Federal University of Uberlândia, Uberlândia, Brazil
Session	Tecnologias Ópticas e Mecânicas para a Saúde

### Abstract and Keywords

Diagnosis and monitoring of diabetes mellitus (DM) through blood is highly accurate, however, it is currently an invasive procedure, causes discomfort to patients, and has a high cost for public and private health systems. The development of sustainable platforms using non-invasive samples with high accuracy and reduced cost could be an attractive alternative to DM surveillance. In this context, the combination of infrared spectroscopy with Fourier transform coupled to attenuated total reflectance (ATR-FTIR) and artificial intelligence (AI) strategies in biological fluids can be used as an alternative diagnostic and/or monitoring tool. In this study, the aim was to apply the ATR-FTIR tool associated with AI to identify specific changes in salivary components as alternative spectral biomarkers and validate them using AI strategies for DM detection. Ethical approval CAAE: 69849223.4.0000.5152. In this context, the results indicate the distinctive vibrational patterns between the three groups: individuals without DM, with impaired glucose tolerance (IGT), and with type 2 diabetes mellitus (T2DM). The analysis of infrared spectra of saliva did not demonstrate satisfactory performance (Accuracy: 59%) in distinguishing between individuals without DM and those with IGT. On the other hand, the K-Nearest Neighbors algorithm showed adequate accuracy (71%) and high sensitivity (86%) in discriminating against patients without diabetes and with T2DM. The main vibrational modes applied with greater weight in AI algorithms were related to nucleic acids and lipids. Our findings indicate that the use of a biophotonic platform coupled with AI algorithms using saliva as a screening fluid for DM2 can be a sustainable alternative, without the use of reagents, non-invasive, large-scale, fast and highly reproducible for detection of this highly prevalent endocrine disease globally and with a major impact on public health.

**Keywords:** screening, infrared spectroscopy, machine learning.



Title	Effect of bacterial supernatants on gene expression in heterochromatin region
Authors	Alexandre Souza Marquez Serena Mares Malta Ana Carolina Costa Santos Joyce da Silveira Pereira Rafael Bernardes Ferreira Matheus Henrique Silva Carlos Ueira-Vieira
Affiliations	Instituto de Biotecnologia – UFU
Session	Terapia Gênica e Celular, Biologia Omics

## and Keywords

Drugs that modify chromatin are promising in the treatment of genetic diseases because they affect gene expression and protein production, with applications ranging from gene therapies to cancer drug development. In this context, natural products, such as those produced by bacteria, are widely used in the exploration of compounds capable of influencing gene expression. One way to investigate compounds with this potential is through the use of Drosophila melanogaster as a model organism.

We explored the ability of supernatants produced by bacteria isolated from stingless bee larval food to influence gene expression in heterochromatic regions. Bacterial supernatants (1A, 1B, 54B, S31, and 07) were administered to Drosophila melanogaster embryos from the #W1118 (negative control with white eye color) and #84099 (has a phenotypic marker gene for eye coloration in the centromeric heterochromatic region) strains, with biological and technical replicates. Individuals were analyzed in the adult stage for eye coloration, correlated with the intensity of heterochromatin decondensation and marker gene expression.

Supernatants 54B and 1B showed satisfactory results in promoting heterochromatin decompaction and changing eye color, indicating effective modulation of heterochromatin and gene expression. On the other hand, supernatants 1A, S31, and 07 caused a slight alteration in eye coloration, but not as pronounced as those treated with 54B and 1B.

All bacterial supernatants had positive results, suggesting the presence of compounds that influence gene transcription to varying degrees. Future research can delve into the molecular mechanisms involved and identify specific compounds responsible for these effects, which could be used in therapies and approaches in the treatment of genetic diseases.

**Keywords:** Chromatin modification, Gene expression., Bacterial supernatants.



Title	Exploring the capacity of Maytenus ilicifolia in inhibiting Type 1 collagen glycation, antimicrobial activity and its cellular viability
Authors	Vinícius Prado Bittar <sup>1</sup> , Ana Luiza Silva Borges <sup>1</sup> , Iasmin Aparecida Cunha Araújo <sup>2</sup> , Neide Maria Silva <sup>2</sup> , Nagela Bernadelli Sousa Silva <sup>3</sup> , Daniela Silva Gonçalves <sup>3</sup> , Carlos Henrique Gomes Martins <sup>3</sup> e Foued Salmen Espíndola <sup>1</sup>
Affiliations	¹Laboratory of Biochemistry and Molecular Biology in Institute of Biotechnology, Federal University of Uberlândia, Uberlândia - MG, Brazil. ²Laboratory of Immunopathology, Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlandia, MG, 38400-902, Brazil. ³Laboratory of Antimicrobial Testing, Institute of Biomedical Sciences, University of Uberlandia, ^ Campus Umuarama, Uberlandia, ^ MG, 38405-320, Brazil
Session	Basic a Clinical Pharmacology

#### Abstract and Keywords

The advancement of metabolomics has facilitated the identification of diverse molecules present in medicinal plants, thereby enabling the investigation of their biological potential through antiglycation activity and cellular viability. This study assessed the antiglycation potential of Maytenus ilicifolia Mart. ex Reissek (EE-Mi) ethanolic extract and its organic fractions (hexane-HX, dichloromethane-DM, ethyl acetate-EA, n-butanol-BF, hydro-methanolic-HM) in inhibiting collagen glycation induced by methylglyoxal. Additionally, antimicrobial activity Cutibacterium acnes was evaluated, and cellular viability in fibroblasts was also examined. EE-Mi and its organic fractions were exposed to a solution containing type 1 collagen (100 mM) and methylglyoxal (1 mM) for 72 hours at 37°C, followed by measurement of fluorescence intensity (340nmex/380nmem). Murine NIH/3T3 fibroblasts were employed to assess cytotoxicity concentrations using the (MTT) assay, with EE-Mi and organic fractions added at concentrations of 1000, 500, 100, and 10 µg/mL. Spectrophotometric readings were taken at 570nm emission. Samples of M. ilicifolia at a dose of 10 µg/mL induced low cytotoxicity in fibroblasts, with EAF-Mi exhibiting the lowest value. The collagen glycation model demonstrated that M. ilicifolia samples exhibited significantly higher antiglycation activity. The EC50 values for EAF-Mi (25.03  $\pm$  6.69) and BF- Mi (46.36  $\pm$  14) were lower than that of quercetin ( $86.44 \pm 21.66$ ). The formation Advanced Glycation End-products can be prevented by compounds with antioxidant and metal ion chelating properties. In the evaluation of antimicrobial activity, hexanic fraction, dichloromethane fraction, and ethanol extract fraction demonstrated substantial inhibition of bacteria through the Minimum Inhibitory Concentration experiment. These findings underscore the relevance of natural compounds, such as those found in M. ilicifolia, in the pursuit of preventive and therapeutic strategies.

Keywords: Antiglycation; Collagen; Cell Viability



Title	Ages and antibacterial effects of <i>lippia alba</i> ethanolic extract and essential oil
Authors	Annelise Arantes Rocha1 Tarcisio Paiva Mendonça1 Frank dos Santos da Silva2 Diego Godina Prado2 Nagela Bernadelli Sousa Silva3 Daniela Silva Gonçalves3 Alberto de Oliveira2 Francisco José Tôrres de Aquino2 Carlos Henrique Gomes Martins3 Renata Roland Teixeira1 Foued Salmen Espíndola1
Affiliations	1-Institute of Biotechnology, Federal University of Uberlandia, Uberlandia, MG, 38400-902, Brazil 2 - Institute of Chemistry, Federal University of Uberlandia, Uberlandia, MG, 38400-902, Brazil 3 - Institute of Biomedical Science, Federal University of Uberlandia, Uberlandia, MG, 38400-902, Brazil
Session	Basic and Clinical Pharmacology

and Keywords

Lippia alba (Mill.) N.E.Br. ex Britton & P.Wilson (Verbenaceae) is native to South America with renowned medicinal properties attributed to polyphenols. Ethanolic extract (EE) and essential oil (EO) were prepared from the dried and fresh leaves. The EE was partitioned into fractions by increasing the polarity solvent sequence using hexane (HF), dichloromethane (DM), ethyl acetate (AE), and n-butanol (BT). Antiglycation and bactericidal properties were determined. The properties to inhibit advanced glycation end products (AGEs) causing glycoxidation and aggregation of proteins were assessed with fructose and methylglyoxal (MGO) glycant agents over a 14-day period (3d, 7d and 14d) of incubation at 37C and measured by fluorescence (em/ex wavelengths) of 350/420 nm (fructose) and 340/380 nm (MGO). Aminoguanidine, catechin (positive controls), and L. alba samples were the antiglycant agents assayed. Glycoxidation results showed AE and BT over 90% inhibitory potential at 3d compared to aminoguanidine and catechin. However, a considerable reduction in inhibition percentage was observed after 7 and 14 days. Aggregation inhibition of BSA glycated by fructose revealed that the HF, DM, and AE exhibited an inhibitory percentage bigger than 46%, 49%, and 45.5%, respectively, in the formation within a period of 7d. Antimicrobial activities of L. alba fractions and OE by assessing Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) tests showed that HF displayed superior inhibitory performance with MIC below 31 and MBC at 62. EO showed MIC and MBC at 62 for Cutibacterium acnes, while other bacterial strains showed insignificant outcomes. Therefore, the results underscore the promising potential of L. alba, especially AE, BF (AGEs inhibition), HF, and OE (bactericidal activities), implying their prospective utility in forthcoming therapeutic interventions.

**Keywords**: *Lippia alba*, AGEs, protein aggregation, antibacterial effect.



Title	Wolf fruit (Solanum lycocarpum): bioactive compounds and therapeutic potential
	Laís Oliveira Souza <sup>1</sup>
A	Marcos Paulo Oliveira Almeida <sup>2</sup>
Authors	Celene Maria De Oliveira Simões Alves <sup>3</sup>
A 66:1: - 1:	<ul> <li>1 - Biomedical. Federal University of Uberlandia, Uberlandia, Brazil</li> <li>2 - Laboratory of Reproductive Immunophysiology, Institute of Biomedical</li> </ul>
Affiliations	Sciences, Federal University of Uberlandia, Uberlandia, Brazil
	3 – Department of Pharmacology, Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlandia, Brazil.
Session	Basic and Clinical Pharmacology

## and Keywords

Solanum lycocarpum, a plant species native to the Brazilian Cerrado and known as "lobeira" or "wolf fruit", is a source of plant derivatives with biological effects and potential therapeutic use. This study conducted a "state of the art" literature review on the biological effects and bioactive compounds of S. lycocarpum, using original and complete scientific articles from the "PubMed Central" and "PubMed" databases. These articles investigated the biological properties of plant derivatives of this species, published in English and Portuguese from January 2000 to June 2023. Moreover, the cytotoxic potential of Alkaloid Extract from the fruits of S. lycocarpum (EASL) was evaluated in an in vitro experimental model using RAW 264.7 macrophage-like cells, by the MTT (methylthiazole tetrazolium) assay. The results showed that the fruits are the most studied part of the plant; however, there is a lack of studies on the leaves and other parts of the plant. Phenolic acids and glycoalkaloids, solamargine and solasonine, are the main active compounds. The articles included in this study highlighted the following biological actions: antiparasitic (23.5%), cytotoxicity (20.8%), inflammatory (11.8%), antitumor (8.8%), hypoglycemic (8.8%) and analgesic (5.9%). The plant species does not cause genotoxicity *in vivo* and *in vitro* models. The alkaloid extract at concentrations of 15.62, 31.25 and 62.5 µg/mL, filtered and solubilized in DMSO, showed no cytotoxicity to RAW 264.7 cells. It is concluded that S. lycocarpum is a promising plant species for obtaining active ingredients with potential pharmacological actions, whose mechanisms of action remain a wide field for research.

**Keywords:** Solanum lycocarpum; state-of-the- art review; glycoalkaloids; cytotoxicity; RAW 264.7 cells.



Title	The antimicrobial, antioxidant, and protective activity of extracts from <i>Libidibia ferrea</i> and its potential use in animal or human
Authors	Ana Rafaela Silva de Macedo¹; Simone Sommerfeld¹; Fabiana Oliveira Notário¹; Mário Machado Martins²; Luciana Machado Bastos¹²; Vasco Azevedo³⁴; Alessandra Aparecida Medeiros-Ronchi¹; Belchiolina Beatriz Fonseca¹²
Affiliations	<sup>1</sup> Faculty of Veterinary Medicine, Federal University of Uberlândia, Uberlândia-MG, Brazil; <sup>2</sup> Biotechnology Institute of the Federal University of Uberlândia, Uberlândia- MG, Brazil; <sup>3</sup> Department of Genetics, Ecology, and Evolution, Federal University of Minas Gerais, Belo Horizonte- MG, Brazil; <sup>4</sup> Postgraduate Program in Animal Science in the Tropics at the Federal University of Bahia, Salvador – BA, Brazil
Session	Farmacologia básica e clínica

## Abstract and

The potential bioactivity of botanical extracts is increasingly acknowledged, owing to their varied characteristics. This study focused on exploring the chemical and biological properties of Libidibia ferrea, a plant native to the Caatinga region in northeastern Brazil. The aim was to uncover secondary metabolites and demonstrate the antimicrobial, antioxidant, and protective qualities of the plant extract. From the screening of several extracts, the a hydroalcoholic extract from the maceration of pods (EHMV) was chosen for the best antimicrobial activity. Metabolite and metal characterization were performed using high-performance liquid chromatography (HPLC-MS/MS) and atomic absorption spectroscopy (AAS). Antimicrobial activity against Salmonella Galinarum (SG), Salmonella Pullorum (SP), and Salmonella Heidelberg (SH) was evaluated, both independently and in combination with probiotic bacteria Bacillus velenzensis (BV), using agar diffusion and minimum bactericidal concentration (CBM) assays. The antioxidant potential was assessed through five in vitro assays and six assays involving 3T3 cells. Toxicity testing of EHMV and its efficacy against SP infection were conducted using a chicken embryo model. Results demonstrated significant antimicrobial activity in EHMV, particularly when combined with BV, resulting in enhanced antimicrobial activity and bacterial sporulation. Chemical analysis identified eight compounds, including tannins and phenolic compounds. In vitro



antioxidant assays showed increased Total Antioxidant Capacity (TAC), potent reducing power, and remarkable metal chelating effects. Analysis on 3T3 cells confirmed EHMV's protective role against oxidative stress. Toxicity assessments on chicken embryos affirmed EHMV's safety and its effectiveness in reducing SP-induced mortality. EHMV from *L. ferrea* was found to be rich in proteins and essential metabolites, contributing to its antimicrobial and antioxidant capabilities. With its non-toxic nature and protective effects, EHMV emerges as a promising supplement for animals and humans.

**Keywords**: antimicrobial, antioxidant, phytopharmaceutical, *Libidibia ferrea* 

Comitê Ético: (80/2023/CEUA/PROPP/REITO).



Title	Annona crassiflora Mart. aqueous leaf extract possesses hypotensive and cardiotoxic effects
	Ingrid Beatriz de Melo Morais¹ Denise Brentan Silva²
	Mateus Jacinto Luz <sup>1</sup> Marcos Luiz Ferreira-Neto <sup>3</sup>
Authors	Celene Maria de Oliveira Simões Alves <sup>1</sup> Carlos Alexandre Carollo <sup>2</sup>
	Patrícia Fidelis-de-Oliveira <sup>4</sup>
	Luiz Borges Bispo-da-Silva <sup>1</sup>
	1 - Department of Pharmacology, Institute of Biomedical Sciences, Federal University of Uberlândia/UFU, Campus Umuarama, Uberlândia, Minas Gerais, Brazil.
	2 - Laboratório de Produtos Naturais e Espectrometria de Massas (LAPNEM), Federal University of Mato Grosso do Sul/UFMS; Campo Grande, Mato Grosso do Sul, Brazil.
Affiliations	3 - Department of Physiology, Institute of Biomedical Sciences, Federal University of Uberlândia/UFU, Campus Umuarama, Uberlândia, Minas Gerais, Brazil.
, unitacionis	4 - Department of Physiology, Institute of Biosciences, São Paulo State
	University/UNESP, Botucatu, Brazil.
Session	Apresentação Pôster

## and

We investigated the cardiovascular effects of Annona crassiflora Mart (Myrtaceae) leaf extract (ACLE), known as "araticum-do-cerrado" in Brazil. The impact of intravenous ACLE on mean arterial pressure (MAP), heart rate (HR), and the QT interval (QTi) was assessed in an esthetized male Wistar rats (250-350 g; n = 4-5/group; CEUA/UFU 105/16). Hypotension induced by ACLE was acutely examined pre- and post-administration of methylatropine (MAT), L-N( $\omega$ )-nitro-L-arginine methyl ester (L-NAME), hexamethonium (HXT), indomethacin (IND), glibenclamide (GLB), or nifedipine (NFD). Secondary metabolites in ACLE were identified through HPLC-DAD-MS/MS analysis. ACLE administration dosedependently reduced MAP and increased QTi. The highest dose (80 mg/kg) also led to tachyarrhythmias and episodic polymorphic arrhythmias. Only NFD administration attenuated the hypotension induced by ACLE (45 mg/kg). IND, L-NAME, GLB, and MAT exacerbated ACLE-induced hypotension, while HXT had no effect. At 45 mg/kg, ACLE induced arrhythmias post-glibenclamide (GLB) administration. Flavonoids, alkaloids, and proanthocyanidins were detected in the extract, along with two oligomeric series of condensed tannins containing up to nine flavan-3-ol units. ACLE-induced hypotension appeared unrelated to muscarinic receptor stimulation, sympathetic modulation, nitric oxide, prostacyclin synthesis, or KATP channel activation. The extract seemed to trigger the release of a cyclooxygenase-dependent vasoconstrictor, whose effects were 2024 FeSBE Reg - 102



overshadowed by the vasorelaxant properties of ACLE. Secondary metabolites from ACLE potentially blocked L-type calcium channels, contributing partly to hypotension induction. High ACLE doses resulted in fatal cardiotoxicity linked to QTi prolongation.

**Keywords**: Annona crassiflora Mart, Annonaceae, Alkaloid, Arrhythmia, Proanthocyanidin.



Title	Evaluation of astrocyte neuroplastic responses mediated by cannabidiol administration
Authors	Ana Beatriz Cadeu¹  Marcelo de Cássio Barreto de Oliveira¹  Sandra Regina Mota Ortiz²  Aline Gavioli²
Affiliations	1 - Universidade Cidade de São Paulo, SP, Brazil 2 - Universidade São Judas Tadeu, SP, Brazil
Session	Neuropsychopharmacology

## and Keywords

Neuroplasticity is the brain's ability to adapt according to stimuli, and can be assessed as a synaptic configuration or a behavioral modification. The processes covered in neural plasticity are dendritic formation, synaptic remodeling, longterm potentiation, axonal development, neurite extension, synaptogenesis and neurogenesis. Astrocytes are macroglial cells responsible for maintenance, nutrition, homeostasis of the central nervous system and several other functions. Cannabidiol (CBD) has a broad neuroprotective spectrum due to its antioxidant and anti-inflammatory activity associated with the modulation of receptors and channels involved in neurological development and maintenance. As an intervention to improve neurodegenerative diseases, or prevent them, the present study proposes that the use of CBD would have effective treatment and prevention responses in the therapy of diseases that affect the nervous system, favoring brain neuroplasticity and its maintenance Objective: Analyze the astrocytic neuroplastic response during cannabidiol administration. Methodology: 12 15-month-old male Wistar rats (CEUA 070) were used, divided into 2 groups, Saline Control, administration of 20ml/km of saline (HC n=6) and CBD Control, administration of 20ml/km of CBD (HM n=6), 1x per week for 13 weeks. The animals were euthanized with transcardiac perfusion, microtomy was performed with serial frontal cuts measuring 40  $m_{\mathcal{U}}$  thick, kept in antifreeze solution. The cuts made aim to evaluate astrocytic activity in the prefrontal cortex, motor cortex, hippocampus, amygdaloid complex and basal ganglia through immunohistochemical placement. delimited. Subsequently, the data obtained will be statistically analyzed.

**Keywords**: neuroplasticity; cannabidiol; astrocytes.



Title	Analysis of neuroplastic difference mediated by cannabidiol in an enriched environment and aerobic exercise
Authors	João Guiral Nascimento  Marcelo de Cássio Barreto de oliveira Gilberto Cândido Laurentino  Sandra Regina Mota Ortiz Aline Gavioli
Affiliations	1 – UNICID-Univercidade cidade de São Paulo 2 – USJT-Univercidade São Judas Tadeu
Session	Neuropsychopharmacology

## and Keywords

Introduction: The World Health Organization recommends aerobic exercise for brain health, however environmental enrichment, where animals have access to stimulating toys and environments, also appears to be effective in enhancing neuroplasticity. Recently, cannabidiol (CBD), has emerged as a potential alternative for improving neuroplasticity.

Objective: This study aims to compare the effects of aerobic exercise and enriched environment on neuroplasticity-associated behavioral changes and running performance in adult Wistar rats. We will further investigate whether cannabidiol administration modulates these effects.

Methods: Twenty-four, 15-month-old male Wistar rats (CEUA: 090 and 091) were divided into four groups (n=6/group) for an exercise intervention. All groups received intraperitoneal injections (0.20 mL) of either CDB or saline solution. Group designations were: Aerobic Cannabidiol (A-CDB), Aerobic Saline (AS), Enriched Environment Saline (ES), and Enriched Environment Cannabidiol (E-CDB). AS and A-CDB groups underwent an incremental treadmill test before and after the experiment to assess exercise capacity. Enriched environment groups (ES and E-CDB) were allowed free play with toys for one hour per day, five days per week, for thirteen weeks. Conversely, AS and A-CDB groups ran on a treadmill at 60% of their maximal speed for the same duration and frequency.

Results: Using subjective observation of the animal behavior in the enriched environment, E-CDB expended more time and energy exploring the environment and did not sleep. Compared to ES, who slept more and explored less. Total aerobic running was similar between A-CDB and AS. Pre (p=0,599) and post (p=0,601).

Conclusion: Subjective observations appear to suggest a behavioral improvement in CBD groups. However, it needs further histological examinations to confirm neuroplasticity. Contrarily, exercise did not yield any significant enhancements.

keywords: Cannabidiol; Aerobic exercise; Neuroplasticity; Enriched Environment



Title	Exploring the effects of the chronic unpredictable stress on medial prefrontal cortex functional laterality: insights from local field potentials
Authors	Gessynger Morais-Silva <sup>123</sup> Juliana Fiúza Fernandes <sup>23</sup> Ricardo Luiz Nunes-deSouza <sup>14</sup> Nuno Souza <sup>23</sup> Hugo Leite-Almeida <sup>23</sup>
Affiliations	<ol> <li>Laboratory of Pharmacology, Department of Drugs and Medicines, School of Pharmaceutical Sciences, São Paulo State University (UNESP), Araraquara, Brazil</li> <li>Life and Health Sciences Research Institute (ICVS), School of Medicine, University of Minho, Campus de Gualtar, Braga, Portugal</li> <li>ICVS/3B's - PT Government Associate Laboratory, Braga/Guimarães, Portugal</li> </ol>
	4 - Joint Graduate Program in Physiological Sciences (PIPGCF) UFSCar-UNESP, Araraquara/São Carlos, Brazil
Session	Oral presentation

#### Abstract and keywords

Results obtained from direct manipulation of the medial prefrontal cortex (mPFC) neuronal activity suggest that an impairment of left (L)mPFC activity and an increase in right (R)mPFC activity are linked to chronic stress effects, leading to the hypothesis that long-term changes in mPFC functional lateralization are implicated in stress-related psychiatric disorders. However, it is not known yet how the dynamics of activity and communication of the different mPFC hemispheres are altered after stress exposure. In this study, we evaluated the electrical activity and communication between the (L) and (R) mPFC using the power spectrum density (PSD) and coherence of the local field potentials (LFPs), respectively, in female and male Wistar-Han rats exposed to a chronic unpredictable stress (CUS) protocol for 21 days (DGAV #023875). Animals were submitted to a stereotaxic surgery for the implantation of nickel/chromium microelectrodes in (L) and (R) mPFC. The PSD was calculated using the fast Fourier transform and, coherence using multi-taper Fourier analysis by custom scripts in Matlab®. Data was z-scored for each animal in the frequency domain (1-100Hz) and analyzed as frequency bands [delta( $\delta$ ) 1-4Hz; theta( $\theta$ ) 5-7Hz; alpha(a) 8-12Hz; beta( $\beta$ ) 13-30Hz; low gamma(Ly) 31-50Hz; high gamma(Hy) 51-100Hz]. The exposure to CUS induced an increase in  $\delta$  PSD in both mPFC hemispheres and  $\delta$  (L)-(R) mPFC coherence in both female and male rats. There was also a decrease in a PSD in both mPFC hemispheres and Ly (L)-(R) mPFC coherence due to CUS exposure, in both sexes. Intriguing, males showed higher β PSD and smaller Hy PSD compared to females in both hemispheres of the mPFC. The LFPs reflect the informational flow from neuronal networks and an integrated view of the inhibitory and excitatory synaptic events. Your results suggest that CUS exposure induced alterations in how information is transmitted between the (L)-(R) dorsal mPFC, changing from slower to higher frequency bands.

**Keywords:** medial prefrontal cortex; functional lateralization; chronic unpredictable stress; local field potentials; coherence.



Title	Evaluation of participation of medial prefrontal cortex (mPFC) and corticotropin releasing factor (CRF) type 1 receptor (CRF1) in the anxiety-like behaviors response in c57bl/6 mice exposed to a rat exposure test (RET)
Authors	Amanda Sores Lima Celina Monteiro da Cruz Lotufo Tarciso Tadeu Miguel
Affiliations	Universidade Federal de Uberlandia, Uberlandia, MG, Brasil.
Session	Neuropsicopharmacology

### and Keywords

Introduction: CRF has shown involvement in defensive and anxiety-related reactions and are highly expressed in human and rodents brains, including prefrontal cortex, important site involved in the modulation of defense reactions and anxiety.

Objectives: evaluate the effect of microinjection of the cobalt chloride, a drug that causes inactivation of neurons, into the mPFC in mice exposed to RET and assess the mPFC neuron activation and CRFR1 expression after RET exposition.

Methods: subjects: C57BL/6 male mice. CEUA-UFU protocol 111/15. Exp.1: mice underwent stereotaxic unilateral cannula implant surgery, aiming mPFC, in a group in the left side and other in the right side. After 5 days recovery, were treated intra-mPFC with vehicle or CoCl2; 1,0 nmol. Ten minutes later were exposed to RET (ten minutes). Exp.2: mice were submitted to RET in an acute (once) or a repeated (five times) way, in three groups: control (not exposed), toy rat and live rat exposed, in order to evaluate neuronal activation in mPFC with immunohistochemistry for Fos and Western blot for CRF1 receptor.

Results: Exp.1: CoCl2 treated animals in the right side remained more time in the surface, the most aversive area of the apparatus (U=6;p=0,02), and performed more the behavior of the escalation of wire close to predator (U=9;p=0,04). Same difference was not observed in the left side treatment. These results signaling an anxiolytic-like effect Exp.2: Fos protein counting in mice exposed to RET do not indicate difference in neuronal activation in the acute exposition. However, in mice repeatedly exposed, expression was decreased, when compared to the respective control and exposed to the toy, a stuffed rat (F2,11=2,89;p=0,05). There was no changes in the CRFR1 expression in the mPFC. Conclusion: it is possible that mPFC exerts mediation in the anxiety and there is a lateralization of mediation which is an interesting effect previously discussed in others anxiety models.

**Keywords**: Anxiety, Rat Exposure Test, medial Prefrontal Cortex



Title	Behavioral effects of treatment with 1,8 cineol intraperitoneally on anxiety models in mice
Authors	Milena Fontenele de Oliveira <sup>1</sup> Antonio Renan Santana <sup>1</sup> Jeremias Martins Gonçalves <sup>2</sup> Lissiana Magna Vasconcelos Aguiar <sup>1</sup> Francisco Walber Ferreira da Silva <sup>1</sup>
Affiliations	<sup>1</sup> Federal University of Ceará, Sobral-CE. <sup>2</sup> Universidade Estadual Vale do Acaraú.
Session	Neuropsychopharmacology

#### Abstract and Keywords

Anxiety disorders are a group of psychological disorders characterized by common signs and symptoms, which can compromise patient's quality of life. Although traditional psychotropic drugs plays an important role in controlling the disorder, they show adverse effects that can lead patients to discontinue the treatment. In this context, 1,8-cineole (CIN) is a plant-derived monoterpene with anti-inflammatory, antioxidant activities, a possible neuroprotective effect and has inhibitory activity on the central nervous system, suggesting a potential anxiolytic. Thus, this study investigated the treatment with CIN on animal model of anxiety. This work was submitted to Federal University of Ceará Ethical Committee and received approval No. 04/23. Male Swiss mice (n=48) weighing 20-25g were used for CIN treatment and behavioral tests. 1,8-cineole was administered via i.p. for 14 consecutive days, once a day at doses of 5 and 10 mg/kg. Saline was administered to the negative control group and diazepam (DZP,1 mg/kg) was used as a positive control. The behavioral test used were Open Field Test (OFT), Elevated Cross Maze (ERM), Perforated Plate Test (PPT) and Light/Dark Test Box (LDTC) at days 0 and 14. In the PPT there was an average of 23.83 entries and 25 s of permanence in CIN 5 mg/kg and 15.33 entries and 13.33 s of permanence for 10 mg/kg. In the OFT, there was an average of 14.42 and 8.50 rearings for CIN 5 and 10 mg/kg, respectively. In these two tests, CIN 5 mg/kg showed best results compared to DZP. In the CTCE, CIN 5 and 10 had an average rearing rate of 15.83 and 18,25, respectively, which was higher than the DZP. In the TLCE, CIN 5 had an average of 45,23 entries and 75,22 s in the open arms, a similar value of DZP group. The results showed that the CIN was capable to prevent anxiety in animal model and the dosage of 5 mg/kg presented more satisfactory results, being superior than DZP, the gold standard, in some parameters.

**Keywords**: Anxiety; 1,8-Cineol; Behavior



Title	Effects of cyanobacteria Arthrospira (Spirulina platensis) spp. (AP) on liver tumor evolution on diethylnitrosamine (DEN) induced-mice model
Authors	Luis Manuel Sarmiento-Machado¹ Guilherme Ribeiro Romualdo¹² Luis Fernando Barbisan¹
Affiliations	<sup>1</sup> Department of Structural and Functional Biology, Biosciences Institute, São Paulo State University (UNESP), Botucatu, Brazil. <sup>2</sup> Department of Pathology, Botucatu Medical School, São Paulo State University (UNESP), Botucatu, Brazil.
Session	Toxicologia

#### Abstract and Keywords

Liver cancer is the second leading cause of death among cancers worldwide, and hepatocellular carcinoma (HCC) the most common subtype (75-90%). AP may reduce HCC development given its anti- inflammatory and antioxidant properties observed in previous preclinical studies. Thus, we assessed whether dietary 5% SP attenuates liver tumor development on DEN-induced hepatocarcinogenesis model. Two-week-old female mice C3H/He received four intraperitoneal administrations of DEN (25 mg/kg) once a week or 0.9% NaCl vehicle (control), and were allocated into four groups: (G1) DEN (n=28), (G2) DEN+5%AP (n = 28), (G3) control (n=10) and (G4) control+5%AP (n = 10). During DEN administration, all groups received standard chow. After carcinogen regimen, G2 and G4 received standard chow containing 5%AP (w/w) for 43 weeks. After this period, all animals were euthanized and liver tumors were collected for histopathological analysis. Statistical data were compared among groups using One-Way ANOVA or Kruskall-Wallis. Two-Way ANOVA for food consumption analysis. Chi-square and Mann-Whitney U for tumor incidence and multiplicity, respectively. Food consumption evolution (p>0.05) and final body weight were similar between DEN (G1) and DEN+5%AP (G2) groups. Absolute and relative liver weights were higher in DEN (G1) in comparison to control (G3) (p<0.0001) and control+5%AP (G4) (p=0.0094;p<0.0001,respectively). In contrast, DEN+5%AP (G2) attenuate the alteration induced by DEN on relative (p<0.0237) and absolute liver weights (p=0.0231), as well as in absolute left (p=0.0075) and right kidney weights (p=0.0002) (G1). Incidence and multiplicity of hepatocellular adenomas and HCC did not differ between DEN (G1) and DEN+5%SP (G2), as well as cell proliferation Ki-67 were similar between these two groups. Hence, AP may not exert a protective effect against liver tumor development on histopathological assays.

**Keywords:** hepatocarcinogenesis; chemoprevention; histopathology.



Title	Anti-svPLA2 IgY antibodies: Development of an immunobiological against antigenic targets of Bothropic venom and the soluble extract of Trypanosoma cruzi
Authors	Leonardo Alves Garcia <sup>1</sup> , Isabela de Oliveira Cavalcante Pimentel <sup>1</sup> , Tássia Rafaela Costa <sup>1</sup> , Lorena Pinheiro Morais <sup>1</sup> , Vinícius Queiroz Oliveira <sup>1</sup> , Samuel Cota Teixeira <sup>2</sup> , Álvaro Ferreira Júnior <sup>3</sup> , Veridiana de Melo Rodrigues Ávila <sup>1</sup> .
Affiliations	1- Laboratory of Biochemistry and Animal Toxins, Institute of Biotechnology – Federal University of Uberlândia, Uberlândia, MG, Brazil. 2 – Affiliation Two, City, Country
	2- Laboratory of Reproductive Immunophysiology, Institute of Biomedical Sciences – Federal University of Uberlândia, Uberlândia, MG, Brazil
	3- Departamento de Medicina Veterinária, Universidade Federal de Goiás
Session	[Parasitologia]

# Abstract and Keywords

Phospholipases A2 (PLA2) are vital enzymes studied extensively for their role in phospholipid metabolism across various species, including snake venoms and parasites. In parasites, PLA2 plays a crucial role in the infection process, influencing cell proliferation and modulating the host's immune response. This study evaluated the recognition profile of the polyclonal IgY antibody anti-BnSP-7, a Lys49 svPLA2 isolated from Bothrops pauloensis snake venom, on various Bothropic venoms and isolated svPLA2s from these venoms. Additionally, the study explored its recognition on protein extract of Trypanosoma cruzi and its cytotoxicity against the parasite. Initially, the antibody was purified from the egg yolk protein extract of chickens immunized with the BnSP-7 PLA2 using affinity chromatography on a HiTrap IgY HP column, showing homogeneity on SDS-PAGE analysis. ELISA assays demonstrated that the polyclonal IgY antibody anti-BnSP-7 recognized its antigen (IE = 9.4) and specifically recognized isolated PLA2 isoforms (IE= 10.5 for BthTX-I and IE= 8.7 for BthTX-II) as well as PLA2s present in the crude venoms of B. jararacussu and B. pauloensis (IE= 8.4 and IE= 7.7, respectively). Furthermore, using ELISA, it was observed that IgY anti-BnSP-7 antibodies also recognized targets in the protein extract of T. cruzi, regardless of the parasite extract concentration but dependent on the antibody concentration. Finally, assessing the cytotoxicity of IqY anti-PLA2 on Trypanosoma cruzi in its infectious form (trypomastigotes) revealed a reduction in cellular viability ranging from 20 to 35%. In conclusion, the findings confirm the recognition of phospholipases A2 by the IqY anti-BnSP-7 antibody, highlighting its capability to identify protein targets in parasites. This underscores the biomolecule's potential for future studies and the creation of a novel biotechnological tool for diagnostic applications.

**Keywords**: Antibody IgY, Phospholipase A2, Snake Venom, Trypanosoma cruzi.



Title	Effects of phthalates on the 3D spheroid cell culture as a model for the human ovarian extracellular matrix
Authors	Eloá Alves Bento <sup>1</sup> ; Saba Nikanfar <sup>2</sup> ; Ellen Cristina Rivas Leonel <sup>1</sup> ; Pauliina Damdimopoulou <sup>3</sup> ; Jodi Flaws <sup>4</sup> ; Christiani A. Amorim <sup>2</sup>
Affiliations	<ul> <li>1 - Department of Histology, Embryology and Cell Biology, Institute of Biological Sciences, Federal University of Goiás, Brazil</li> <li>2 - Pôle de Recherche en Physiopathologie de la Reproduction, Institut de Recherche Expérimentale et Clinique, Université Catholique de Louvain, Brussels, Belgium</li> <li>3 - Division of Obstetrics and Gynecology, Department of Clinical Science, Intervention and Technology, Karolinska Institutet, Stockholm 14186, Sweden</li> <li>4 - Department of Comparative Bioscience, University of Illinois at Urbana-Champagne, Urbana, IL 61802.</li> </ul>
Session	Poster Presentation

#### Abstract and Keywords

The widespread production and pervasive use of phthalates as plasticizers in everyday products significantly increases human exposure to these substances. Their documented toxic effects on reproductive and endocrine functions raise substantial public health concerns. Yet, a full understanding of how phthalates affect ovarian functions and especially the extracellular matrix (ECM) structure in females remains incomplete. In the study, stromal cells were isolated from the ovarian cortex of post-menopausal organ donors. Cells were seeded onto agarose micro-well molds to form the spheroids. After 6 days, spheroids were treated with phthalates mix (0 and 100  $\mu$ g/ml) for 4 days. After seeding of cells, their diameter was measured on days 6 and 10 using an inverted microscope to assess their development. The study measured the protein levels of collagen VI, Emilin, and Fibrilin using immunohistochemistry and Ki67 using immunofluorescence in ovarian spheroids. The picrosirius red staining (PSR) was used to measure the collagen content. The evaluation of spheroid diameter indicated comparable sizes on day 6 for both groups. By day 10, there were no notable differences in spheroid size for the group exposed to PM in comparison to the control group. The assessment of proliferation revealed no significant difference in the expression of ki67-positive cells between the groups. On the other hand, immunostaining results displayed a decrease in EMILIN-1 and fibrillin levels post-PM treatment. Conversely, collagen VI levels were significantly higher in spheroids treated with PM compared to the control group. Although PSR revealed an increase following treatment this change did not reach statistical significance. Findings suggest ovarian stiffness by decreased elastogenesis-associated



proteins and increased collagen VI post-phthalate exposure. This study prompts further exploration of phthalate-induced ECM changes and their potential influence on steroidogenesis and folliculogenesis.

**Keywords**: Extracellular matrix, Ovarian toxicity, Phthalates, Spheroids.



Title	Assessment of ethidium bromide toxicity in <i>Drosophila</i> melanogaster
Authors	Maria Eduarda Souza Alves Alexandre Azenha Alves de Rezende
Affiliations	Universidade Federal de Uberlândia, Ituiutaba, Minas Gerais
Session	Toxicology

### Abstract and Kevwords

Ethidium bromide (EB) is a non-radioactive intercalating dye, with moderate toxicity and mutagenic properties, causing fluorescence at UV light and induces loss of supercoiling in double-stranded DNA. It disrupts mitochondrial function and membrane polarization, inhibiting DNA and RNA replication, transcription, and RNA polymerase activity and and is still widely used in molecular genetics laboratories. Aiming to obtaining new compounds as positive controls in mutagenicity tests the present study investigated the toxic activity of EB in somatic cells of *Drosophila melanogaster* using the wing Somatic Mutation and Recombination Test (SMART). Among the tests performed with *D. melanogaster* as a model organism, the SMART stands out, considered one of the "gold standard" tests for the evaluation of mutagenicity and toxicity. Two crosses were used: standard cross (ST), with basal levels of cytochromes P450 (CYP450) enzymes, and high bioactivation cross (HB), with high levels of CYP450. To determine the toxicity of EB, we transferred 20 third-stage larvae into vials containing alternative culture medium (mashed potatoes) hydrated with 5mL of different concentrations of EB (0.2, 1, 5 or 30mg/mL). Survival rates were compared to the negative control using the X2 test. The mortality rate was observed (p>0.05) at the highest concentrations (1, 5 and 30mg/mL). The results have shown that at these concentrations, the EB is extremely genotoxic to both crosses. Comparing the survival rate of ST cross with the HB cross at 0.2 or 1.0mg/mL treatments, is possible to detect a small difference between them. The survival rate of HB cross is slightly higher than ST cross. The EB can induce the expression of enzymes of CYP450. Thus, these concentrations can induce the expression of these enzymes and this induction was greater in the HB cross, as this cross naturally has more CYP450 enzymes. More studies must be performed to elucidate the mode of action of EB through CYP450 metabolization.

Keywords: Genotoxicty; intercalating agent; CYP450



Title	Synthesis conditions of nanoparticles of titanium dioxide doped with Europium influence its properties and cell viability
	Foued Salmen Espíndola <sup>1</sup> ; Vinícius Prado Bittar <sup>2</sup> ; Ana Luiza Silva Borges <sup>1</sup> ;
Authors	Renata Roland Teixeira <sup>1</sup> ; Iasmin Aparecida Cunha Araújo <sup>2</sup> ; Neide Maria Silva <sup>2</sup> ;
	Anielle Christine <sup>3</sup>
	1 - Laboratory of Biochemistry and Molecular Biology in Institute of
	Biotechnology, Federal University of Uberlândia, Uberlândia - MG, Brazil.
Affiliations	2 - Laboratory of Immunopathology, Institute of Biomedical Sciences, Federal
	University of Uberlandia, Uberlandia, MG, 38400-902, Brazil.
	3 – Laboratory of Strategic Materials, Institute of Physics, Federal University of
	Alagoas, Brazil.
Session	Toxicology

# and Keywords

Nanoparticles (NPs) of titanium dioxide (TiO2) are promising candidates for nanomedicine applications. This study investigated the physicochemical and biological properties of an innovated synthesis of NP-TiO2 doped with europium III (Eu), a metal from the lanthanides group with fluorescence attributed. We proceed with this synthesis at different temperatures (500, 600, and 800 °C). TiO2:Eu NPs characterization was performed, including scanning electron microscopy (SEM) analysis to assess morphology and composition, as well as Raman spectroscopy to confirm the absorption peaks of europium at the three temperatures of TiO2:Eu synthesized. Concern biological activities this sturdy evaluated the cellular viability in Fibroblast and Macrophage cell lines at varying concentrations of each NP of TiO2:Eu (CEUA/UFU nº 028/21). Scanning electron microscopy (SEM) revealed the morphology of the nanoparticles, confirming their primarily titanium and oxygen composition although europium was not initially detected. However, Raman spectroscopy successfully identified europium peaks. The cell viability assay showed a concentration and temperature of synthesisdependent in cellular viability, with NPs synthesized at 500°C exhibiting higher reactivity and cytotoxicity compared to those synthesized at 800°C. Therefore, higher synthesis temperatures led to more organized TiO2:Eu NPs, impacting their biochemical properties (not showed) and biological interactions. This study highlights the pivotal role of synthesis conditions in shaping the properties and biological responses of titanium dioxide (TiO2) nanoparticles conjugated with europium, emphasizing their potential applications across diverse biomedical fields.

Keywords: Raman Spectroscopy; Nanoparticles; Cell Viability



Title	Antioxidant and antimicrobial properties of simonkolleite nanocrystals modified with quercetin
Authors	Allisson Benatti Justino¹ Phelipe Augusto Borba Martins Peres¹ Ivis de Melo Agra² Vinícius Prado Bittar¹ Iasmin Aparecida Cunha Araújo¹ Natieli Saito¹ Lucas Correia Peres¹ Arthur Franco Demétrio¹ Neide Maria da Silva¹ Anielle Christine Almeida Silva² Foued Salmen Espindola¹
Affiliations	1 - Federal University of Uberlandia, Uberlândia, Brazil 2 - Federal University of Alagoas, Maceio, Brazil
Session	Toxicologia

#### Abstract and Keywords

Nanomaterials engineered from Simonkolleite (SK) present promising prospects for theranostic applications. Enhancing nanomaterials with polyphenols offers a potential avenue for easier manipulation and improved stability, efficacy and accessibility. We synthesized nanohybrids of SK combined with gold (Au) and quercetin. The synthesis was confirmed using ultraviolet-visible, Fouriertransform infrared, and Raman spectroscopy, and scanning electron microscopy. Our study aimed to understand how these ENMs influence antioxidant and oxidative parameters (official approval CEUA-UFU 07/2022) and evaluate their effects on cytotoxicity and antimicrobial properties. The results were expressed as mean ± standard deviation (n = 3). Nanohybrids incorporating quercetin exhibited antioxidant properties, as confirmed by ORAC (663  $\pm$  16  $\mu$ mol trolox eq/g) and DPPH (9.2  $\pm$  4.9% of activity) evaluations, indicating the presence of quercetin within the inorganic matrix. SK triggered ROS production (an increase of 26  $\pm$  7%) and lipid peroxidation (an increase of 10  $\pm$  5%), while Au and quercetin functionalization normalized these parameters. However, SK combined with quercetin led to protein oxidation (an increase of 8±4%). Both SK and its nanocomposites with Au and quercetin reduced fibroblast viability from 62.5  $\mu$ g/mL (52.9  $\pm$  8.2, 74.4  $\pm$  1.5 and 57.5  $\pm$  2.8% of viable cells, respectively), with SKAu and its quercetin hybrid howing cytotoxicity to macrophages only at higher concentrations (starting from 250 µg/mL). Moreover, the nanohybrids of SK with Au and quercetin displayed antibacterial activity against specific bacterial strains (Salmonella Gallinarum), resulting in a reduction of bacterial viability by 37±2% at 125µg/mL. This investigation underscores how ENMs based on ZnO and modified with quercetin can impact antioxidant and oxidative processes and bacterial viability, alongside highlighting the need to examine the in vivo safety of phytoantioxidantenhanced nanoparticles.

**Keywords:** antioxidant capacity; bacteriostasis; flavonoid; oxidative stress; zinc oxide nanoparticles.



Title	Beyond drug delivery: Preliminary investigation the hemostatic interactions of metallic nanoparticles
Authors	Analydia Ferreira Netto da Paixão <sup>1</sup> ; Lucas Ian Veloso Correia <sup>1</sup> ; Augusto Silva Moreira <sup>1</sup> ; Pedro Henrique Amiante Bugatti <sup>1</sup> ; Noélio Oliveira Dantas <sup>2</sup> ; Anielle Christine Almeida Silva <sup>2</sup> ; Suely Vilela Sampaio <sup>3</sup> ; Veridiana de Melo Rodrigues <sup>1</sup> ; Luiz Ricardo Goulart Filho <sup>1</sup> ; Robson Oliveira Junior <sup>1</sup> ; Luciana Machado Bastos <sup>1</sup>
Affiliations	1 – Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, MG, Brazil. – Institute of Physics, Federal University of Alagoas, Maceió, AL, Brazil.
	2 - Department of Clinical Analysis, Toxicology and Food Science, School of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, Ribeirão Preto, SP, Brazil.
Session	Toxicologia

# Abstract and Keywords

Nanoparticles have already revolutionized the treatment of various challenging conditions, including cancer and other diseases, and still hold the potential to enhance drug delivery to elusive target sites. However, the interactions between nanoparticles and blood cells remain poorly understood, and the significance of evaluating nanoparticle-hemostasis interactions is vastly underestimated. To overcome this limitation, we conducted various assays to study the hemocompatibility and coagulant effects of different metallic nanoparticles on hemostasis. For this purpose, blood was obtained from 20 healthy volunteers (protocol number 4.532.791) and centrifuged at 500 x g for 5 minutes to separate erythrocytes and plasma. The blood cells were washed and diluted at a 1:50 ratio in phosphate saline buffer. Subsequently, 90 µL of red blood cells were plated per well, and 10 µL of different nanoparticles were added in a 2-fold dilution series and incubated for 1 hour at 37 °C. Following the incubation, the plates were centrifuged at 500 x g, and 50  $\mu$ L was transferred to another plate and read at 400 nm. For the coagulation assay, 150 µL of plasma was incubated with 50 μL of different nanoparticles in 0.06 M Tris-HCl buffer, pH 7.2, at 37 °C. The time required to initiate the formation of the fibrin network was recorded using a photometric system in the coagulometer Quick Timer II (Draker-BR). A clotting time duration of more than 240 s was considered indicative of a noncoagulant sample. As a control for the reaction, we used 50 µg of Crotalus durissus collilineatus venom. The metallic nanoparticles did not exhibit hemolytic



activity at the concentrations tested, they did not demonstrate the capability to induce coagulation in citrated plasma. The results highlight the metallic nanoparticles as safe for initial hemostasis viability studies, providing valuable insights into their potential for biotechnological and pharmacological applications.

**Keywords**: Metallic nanoparticles; hemostasis; blood, hemolysis; coagulation.



Title	Quantum dots in hemostasis: Preliminary insights into hemostasis viability
Authors	Ruan Pires da Silva <sup>1</sup> Lucas Ian Veloso Correia <sup>1</sup> Augusto Silva Moreira <sup>1</sup> Pedro Henrique Amiante Bugatti <sup>1</sup> Analydia Ferreira Netto da Paixão <sup>1</sup> Sabrina Cristina Guedes <sup>1</sup> Noélio Oliveira Dantas <sup>2</sup> Anielle Christine Almeida <sup>2</sup> Suely Vilela Sampaio <sup>3</sup> Veridiana de Melo Rodrigues <sup>1</sup> Luiz Ricardo Goulart Filho <sup>1</sup> Robson Oliveira Junior <sup>1</sup> Luciana Machado Bastos <sup>1</sup>
Affiliations	<ol> <li>I - Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, MG, Brazil.</li> <li>Institute of Physics, Federal University of Alagoas, Maceió, AL, Brazil.</li> <li>Department of Clinical Analysis, Toxicology and Food Science, School of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, Ribeirão Preto, SP, Brazil.</li> </ol>
Session	Toxicologia

# and Keywords

In the realm of advanced therapeutics, quantum dots have emerged as transformative agents, reshaping the landscape of treatment of various diseases. Their unique properties hold promise for elevating drug delivery precision to previously elusive target sites. Yet, the intricate interplay between quantum dots and blood cells remain an enigma, underscoring the underestimated significance of probing quantum dot-hemostasis interactions. To bridge this knowledge gap, we explored the hemocompatibility and coagulant effects of diverse quantum dots. Drawing blood from 20 healthy volunteers (protocol number 4.532.791), we subjected the samples to centrifugation at 500 x g for 5 minutes to isolate erythrocytes and plasma. After meticulous washing and dilution at a 1:50 ratio in phosphate saline buffer, 190  $\mu$ L of red blood cells were plated per well. 10  $\mu$ L of 2-fold dilution series of various quantum dots was introduced, followed by a 1-hour incubation at 37 °C. Post-incubation, the plates underwent centrifugation at 500 x g, and 100  $\mu$ L of the resultant solution was transferred to another plate, where readings were taken at 400 nm. For the coagulation assay, 150  $\mu L$  of plasma was incubated with 50 µL of different quantum dots in 0.06 M Tris-HCl buffer, pH 7.2, at 37 °C. The initiation time for the fibrin network formation was measured using the photometric system in the coagulometer Quick Timer II (Draker-BR). Samples with a clotting time exceeding 240 s were classified as non-coagulant. As a reaction control, 50 µg of Crotalus durissus collilineatus 2024 FeSBE Reg - 118



venom was employed. Quantum dots exhibited no hemolytic activity at the tested concentrations and did not induce coagulation in citrated plasma. This robust evaluation underscores the safety of quantum dots for preliminary hemostasis viability studies, offering novel perspectives for their potential applications in biotechnology and pharmacology.

**Keywords**: Quantum dots; hemostasis; blood, hemolysis; coagulation.



Title	Quantification of post-mortem concentration of heavy metals in the vitreous and aqueous humor of human eyeballs from the Triângulo Mineiro region
Authors	João Lucas Oliveira de Souza¹ Eduardo Ferreira Arbache² Raquel Belluco Ribeiro² Fábio Tonissi Moroni² Sueli Moura Bertolino³ Gabriel Menezes Martins²
Affiliations	<ol> <li>Graduando de Engenharia Ambiental e Sanitária, Universidade Federal de Uberlândia, Brasil.</li> <li>Graduando de Medicina, Universidade Federal de Uberlândia, Brasil</li> <li>Instituto de Ciências Agrárias, Universidade Federal de Uberlândia, Brasil</li> </ol>
Session	Toxicology.

### Abstract and Keywords

The eyeballs can be used as biomarkers in postmortem toxicological analyses, however, there are few studies on the quantification of trace elements in scientific databases. Iron, zinc, copper, and manganese are essential elements for ocular physiology, nevertheless, an imbalance in the behavior of these elements may indicate toxicity due to external factors. Atomic absorption spectrometry (AAS) was used to quantify the total amount of trace elements in the vitreous and aqueous humor of human eyeballs, unsuitable for corneal transplantation and donated by the Eye Tissue Bank (BTOC) of the Clinical Hospital of the Federal University of Uberlândia (CAAE 59267822.8.0000.5152). Thus, this project could help increase knowledge of the epidemiological profile and ecotoxicological factors contributing to the increased incidence and prevalence of heavy metal poisoning in the Triângulo Mineiro macro-region, in the State of Minas Gerais. Copper, zinc, manganese, and iron were quantified in the eyeballs of twelve individuals (n=12). The most abundant metal in the eyeball was iron, followed by zinc, copper, and manganese. Iron levels had a mean of  $1.31 \pm 0.99$  mg.g-1, followed by zinc ( $1.31 \pm 0.15$  mg.g-1), copper  $(0.109 \pm 0.06 \text{ mg.g-1})$ , and manganese  $(0.026 \pm 0.022 \text{ mg.g-1})$ . Clustering statistical analyses or data clustering analyses will be performed to find similarities between the levels of trace elements in individuals and their social, regional/environmental characteristics.

**Keywords**: trace elements, eyeball, humor vítreo, toxicology.



Title	Assesing the antimicrobial effects and oxidative stress of ectatomma opaciventre ant venom: an in vitro and in vivo approach
Authors	Augusto Silva Moreira <sup>1</sup> Lucas Ian Veloso Correia <sup>1</sup> Fernanda Gobbi Amorim <sup>2</sup> Renata Aires <sup>3</sup> Larissa Zambom Côco <sup>2</sup> Bianca Brandi Campagnaro <sup>2</sup> Silvana dos Santos Meyrelles <sup>3</sup> Veridiana Melo Rodrigues <sup>1</sup> Luiz Ricardo Goulart <sup>1</sup> Robson Oliveira Junior <sup>1</sup> Luciana Machado Bastos <sup>1</sup> Renata Santos Rodrigues <sup>1</sup>
	1 – Institute of Biotechnology, Federal University of Uberlândia, Uberlândia,
Affiliations	Minas Gerais, Brasil.
Allillations	2 – University of Vila Velha, Vila Velha, Espírito Santo, Brasil
	3 – Federal University of Espírito Santo, Vitória, Espírito Santo, Brasil.
Session	Toxicologia

### Abstract and Keywords

Animal venoms evolved as mechanisms for defense and prey capture. In addition, ant venoms are also important for social communication and defense against pathogens. This study aimed to investigate the antimicrobial potential of the Ectatomma opaciventre venom, in vitro, against Gram-positive, negative and multi-resistant bacterial strains and the in vivo oxidative envenomation effects. The crude venom was extracted and measured by Bradford assay. The strains were reactivated in a brain-heart infusion medium overnight at 37°C and plated on tryptic soy agar medium for 24 hours. In due course, the colony strains were diluted for an antibiogram test against 10 µg of crude venom and gentamicin for positive control. Furthermore, the test was evaluated in a Mueller-Hinton broth, with the strains treated with serial dilutions of the samples, starting in 100 μg/mL. Antimicrobial activity was analyzed after 24 hours by adding 10 μL MTT (5mg/mL) for 2 hours. The in vivo assays (CEUA 044/19) utilized Balb C57BL/6 mice (n=5) which were inoculated with 10, 100 and 300 μg of crude venom for 6 hours. Subsequently, they were euthanized and their blood collected. Peripheral blood mononuclear cells were isolated and analyzed by flow cytometer to determine viability by propidium iodide and production of reactive oxygen species by DCFH probe; for DNA damage, it was conducted comet and micronucleus assay. The blood's plasma was utilized for oxidative damage verification to lipids (TBARS) and proteins (AOPP). The study exhibited a broadspectrum antimicrobial activity, with the exception of Klebsiella pneumoniae and its resistant form, along with the validation of no toxicological effects on animals and, surprisingly, demonstrated an antioxidant effect in the raw blood, after envenomation. It is the first evaluation of the toxic systemic effects by ant poisoning which revealed the E. opaciventre venom as potential source of novel biopharmacological agents.

**Keywords**: Ant venom, *Ectatomma opaciventre*, Antimicrobial effects, Oxidative stress.



Title	Therapeutic effect of an IL-10 mimetic in experimental model of fibromyalgia
	Tamara Marques da Silva
Authors	Ana Cláudia Gontijo Couto
Authors	Luiz Ricardo Goulart Filho (in memorian)
	Cássia Regina da Silva
Affiliations	Post Graduate Program in Genetics and Biochemistry, Institute of Biotechnology,
	Federal University of Uberlândia/Uberlândia (MG), Brazil
Session	Pain and Inflammation

## Abstract and Keywords

Fibromyalqia is a burdensome chronic condition, reaching a worldwide prevalence of 2-3 %. It is characterized by chronic widespread pain, highly associated with other comorbidities such as anxiety and depression. Despite progress, the actual available treatment does not resolve the disease, and also have several undesirable adverse effects. In this scenario, it is well known that fibromyalgic patients have an imbalance in the production of pro- and antiinflammatory cytokines, such as IL-10, which acts reducing pro-inflammatory cytokines production. Thus, we aimed to investigate the possible protective effects of IL-10 in pain, anxious and depression in an animal model of fibromyalgia. Male C57BL6 mice were used in this study (CEUA 030/19). The fibromyalgia-like pain model was induced by a subcutaneous administration of reserpine (0,25mg/Kg) or saline (10 mL/Kg) for three consecutive days, once a day. After the last administration of reserpine, the animals were treated intranasally, with 40 µL of vehicle or doses of 10  $\mu g/\mu L$ , 30  $\mu g/\mu L$  and 100  $\mu g/\mu L$  of the IL-10 mimetic. On the last day after the administration of reserpine and after the treatment with the IL-10 mimetic, parameters of pain. The reserpine caused mechanical hypersensitivity in response to Von Frey filaments on nine days after reserpine injection. Treatment with the IL-10 mimetic was able to reduce mechanical allodynia in animals from the 1st to the 9th day (p<0.001). Furthermore, spontaneous behavior increased by 22% (p<0.01). However, more studies are necessary to understand the mechanisms involved in these effects.

**Keywords**: pain; fibromyalgia; IL-10; mimetic; therapeutic targets



Title	Dendropsophin 1, an antimicrobial peptide from skin secretion of dendropsophus columbianus, inhibits inflammation and stimulates angiogenesis and collagen deposition in a murine model of chronic inflammation
Authors	Isabella Silva Cassimiro¹ Bruno Antonio Ferreira¹ Simone Ramos Deconte¹ Taís de Campos Lima¹ Francyelle Borges Rosa de Moura² Mariana de Souza Castro³ Fernanda de Assis Araújo¹
Affiliations	<ol> <li>Department of Physiological Sciences; Federal University of Uberlandia, Uberlandia, Brazil;</li> <li>Department of Biological Sciences, Federal University of Catalao, Catalao, Brazil;</li> <li>Laboratory of Toxinology, Department of Physiological Sciences, Institute of Biological Sciences, University of Brasilia, Brazil;</li> </ol>
Session	Poster

# and Keywords

Various species of anurans act as reservoirs for antimicrobial peptides (AMPs), which, in addition to exerting antibacterial activity, may also have immunomodulatory effects. In this study, we evaluated the effects of dendropsophin 1 (Dc1) during chronic inflammation induced by polyesterpolyurethane sponge implants in mice. Dc1 is a peptide isolated from the cutaneous secretion of the hylid frog Dendropsophus columbianus and was used as a daily treatment at three different concentrations (32, 64 and 128 µM). The implants allow the evaluation of parameters related not only to inflammation, but also to the development of new blood vessels and the organization of the extracellular matrix. All animal procedures were conducted under conditions approved by the Ethics Commission on Animal Use (CEUA) of the Federal University of Uberlandia (protocol no. 096/19). Dc1 decreased the levels of proinflammatory cytokines CCL2 and CXCL-1. The levels of CCL2 was reduced at all tested concentrations (34.2%, 52.3% and 71.5 % reduction, respectively) and CXCL-1 levels decreased in the treatment with Dc1 at 64 and 128  $\mu M$  $(1.17\pm0.14; 1.0\pm0.13 \text{ pg/wet weight})$  versus the control group  $(2.07\pm0.21)$ . The activation of neutrophils and macrophages into the implant (assessed by the activities of the enzymes MPO and NAG) was reduced too. MPO activity was attenuated in implants treated with Dc1 at 64 and 128  $\mu M$  and the treated



implants showed reduction in NAG activity at all tested concentrations. Additionally, an increase of more than 140% in the number of blood vessels and in the levels of the pro-angiogenic cytokine FGF was observed in the groups treated with Dc1 at 64  $\mu$ M. Finally, the administration of Dc1 augmented the levels of the cytokine TGF- $\beta$  and collagen deposition, showing a pro-fibrogenic effect. Together, these results present Dc1 as a potential anti-inflammatory agent that promotes both angiogenesis and fibrogenic events, which are crucial for tissue repair.

**Keywords**: anti-inflammatory; fibrogenic; dendropsophin 1



Title	Evaluation of the role of nitric oxide in glutamatergic communication between nociceptors and satellite cells of rat dorsal root ganglion
Authors	Santos AOB <sup>1</sup> , Lotufo CMC <sup>1</sup> .
Affiliations	UFU Uberlândia, PPG Applied Cell and Structural Biology, Brazil
Session	20- Pain and Inflammation

# and Keywords

Interactions between satellite glial cells (SGCs) and nociceptive neurons in dorsal root ganglia (DRG) affect neuronal excitability during pain signal transmission. Previous studies have shown that neurons release glutamate, which activates CGSs. These, in turn, interfere with neural activation by promoting membrane depolarization. In this current work, we evaluate the role of nitric oxide (NO) in the depolarization of primary afferent neurons resulting from the activation of NMDA receptors in glial satellite cells. Wistar rats (n=6) were euthanized by decapitation under inhalation anesthesia (isoflurane 5%). Primary DRGs cultures were obtained and the cells were dissociated and plated in 35 mm glass bottom culture dishes coated with Matrigel (CEUA 23117.086099/2022-31). The effect of a nitric oxide donor, sodium nitroprusside (NPS 10  $\mu\text{M},$  100  $\mu\text{M},$  and 1mM) was evaluated, as well as the effect of L-NAME (100 µM), a non-specific inhibitor of nitric oxide synthase enzyme (NOS). Changes in membrane resting potential were assessed by confocal microscopy using the fluorescent indicator DiBAC4(3), whose movement on the membrane due to depolarization results in increased fluorescence. The administration of the NO donor at a concentration of 1 mM induced a significant neuronal depolarization (0.18  $\pm$  0.02) when compared to the vehicle ( $-0.03 \pm 0.005$ ), while there was no effect with the lower concentrations. The administration of the NOS inhibitor inhibited NMDA-induced neuronal depolarization ( $-0.07 \pm 0.03$ ) when compared to the NMDA group (0.15 ± 0.04). Administration of the TRPV1 agonist capsaicin resulted in increased depolarization in the presence of L-NAME ( $0.60 \pm 0.06$ ) compared to the vehicle  $(-0.02 \pm 0.03)$ . Results indicate that NO can depolarize nociceptive neurons and NOS inhibition interfere with NMDA induced depolarization, which corroborates the hypothesis that this mediator might participate in the communication between neurons and SGCs.

Keywords: Pain, Glia, Nitric Oxide, Glutamate.



Title	Protective potential of Xeomin in anxious and depressive-like behavior in chronic orofacial pain in mice
Authors	Ana Claudia Gontijo Couto <sup>1</sup> ; Thays Crosara Cunha <sup>2</sup> ; Tamara Marques da Silva <sup>1</sup> ; Naiara Morais <sup>1</sup> ; Graziella Silva <sup>2</sup> ; Eduardo Januzzi <sup>2</sup> ; Cássia Regina da Silva <sup>1</sup>
Affiliations	<ol> <li>Universidade Federal de Uberlândia. Uberlândia/Uberlândia (MG), Brasil.</li> <li>Programa de Pós-graduação em Dor Orofacial/CIODONTO. Instituto de Biotecnologia, Belo Horizonte (MG), Brasil.</li> </ol>
Session	20 – Dor e Inflamação

# and Kevwords

Trigeminal neuralgia (TN) is a severe chronic pain condition, characterized by recurrent, unilateral, paroxysmal electric shock-like pain. Depression and anxiety are often observed in patients with chronic pain. The management of the TN, and its associated comorbidities, is a big challenge and involves different medications, which increases the occurrence of adverse effects. Therefore, TN patients are in urgent need of more treatment options without side effects. Here we evaluated the protective effect of Xeomin, a botulinum neurotoxin brand, in the development of anxiety and depression-like behavior associated with trigeminal pain in mice. To induce the model of TN, animals were anesthetized with ketamine and xylazine, and two ligatures were made on the infraorbital root of the trigeminal nerve of adult male mice C57BL/6J (20-25g) (CEUA 23117.011633/2022-55). On the 7th day after the surgical procedure, the animals were treated with Xeomin 0.06U s.c on the vibrissial area. Mechanical allodynia was accessed 3, 7, 10, 12, 15, and 18 days after the model induction, using the "Up and Down" method with Von Frey filaments. The spontaneous pain behavior was evaluated observing the face rubbing for 30 minutes. The anxious like behavior were accessed on the 14th, by the elevated plus maze (EPM). The treatment with Xeomin reduced the mechanical allodynia on 12th and 15th (p<0,05) days and the spontaneous pain behavior on  $10^{th}$  and  $12^{th}$  (p<0,001)days, reinforcing its analgesic effect. Furthermore, Xeomin increased in 31% The time spent in the open arms of EPM. Our results show that the treatment with Xeomin was able to reduce the pain, and the development of anxiety like behaviors. Moreover, the data suggests the use of a therapy that can treat the different dysfunctions associated with chronic orofacial pain. Despite those effects, more studies are needed to understand the mechanisms involved in these results.

**Keywords**: Trigeminal neuralgia, botulinum neurotoxin, pain, anxiety, depression



Title	Evaluation of the analgesic potential of Botox in a model of temporomandibular disorder induced by CFA in mice
Authors	Héllen de Paula NogueiraSoares <sup>1</sup> ; Ana Claudia Gontijo Couto <sup>2</sup> ; Thays Crosara Cunha <sup>3</sup> ; Tamara Marques da Silva <sup>2</sup> ; Naiara Morais <sup>1</sup> ; Graziella Silva <sup>3</sup> ; EduardoJanuzzi <sup>3</sup> ; Cássia Regina da Silva <sup>12</sup>
Affiliations	1-Grupo de Pesquisa em Analgesia e Inflamação, Laboratório de Bioquímica e Toxinas Animais, Instituto de Biotecnologia, Universidade Federal de Uberlândia/Uberlândia (MG), Brasil; 2-Grupo de Pesquisa em Analgesia e Inflamação, Laboratório de Bioquímica e Toxinas Animais, Programa de Pós Graduação em Genética e Bioquímica, Instituto de Biotecnologia, Universidade Federal de Uberlândia/Uberlândia (MG), Brasil; 3-Programa de Pós-graduação em Dor Orofacial/ CIODONTO/ Belo Horizonte (MG), Brasil
Session	20 – Dor e Inflamação

### Abstract and Keywords

The temporomandibular disorder (TMD) is described as a multifactorial pain disorder caracterized by damage in the temporomandibular joint and its function. The conventional treatment for TMD is the use of anti-inflammatory, analgesics and muscle relaxants. However, some patients are resistant or refrectory to those treatment, motivating the studies of more treatment options, as the use os botulinum neurotoxin-A (BONT-A). Here we evaluated the analgesic effect of Botox in a model of temporomandibular disorder in mice. To induce the model of DTM, animals were anesthetized with ketamine and xylazine, and an i.a injection of Complete Freund Adjuvant was made in the temporomandibular joint of adult male mice C57BL/6J (20-25 g) (CEUA 23117.011633/2022-55). On the  $14^{th}$ day after the injection, the animals were treated with Botox 0.06U s.c on the vibrissial facial area. The nociceptive parameters were accessed 7, 14, 21, and 28 days after the model induction. Mechanical allodynia was accessed using the "Up and Down" method with Von Frey filaments. The spontaneous behavior was evaluated by observing the rubbing behavior for 30 minutes. The cold allodynia was evaluated in the acetone test. The treatment using Botox reduced the mechanical allodynia in 16% in the 21<sup>St</sup> day and 30% in the 28<sup>th</sup> day. Furthermore, the animals treated with Botox have diminishing 30% of the time in rubbing in the 21<sup>st</sup> day and 45% in the 28<sup>th</sup> day. Otherwise, the treatment did not reduce the cold allodynia. Our results suggest that the treatment using Botox has antinociceptive effects in animals with TMD induced by CFA. Moreover, more studies about the effects of this medication are necessary. Keywords: temporomandibular disorder, botox, pain



Title	Evaluation of the antinociceptive and/or anti-inflammatory activities of the ethyl acetate extract of <i>Celtis iguanaea</i> leaves in a chronic model of rheumatoid arthritis.
Authors	Dionys de Souza Almeida <sup>1</sup> Lorrane kelle da Silva Moreira <sup>1</sup> Elson Alves Costa <sup>1</sup> James Oluwagbamigbe Fajemiroye <sup>1</sup> Yohanny Souza Silva <sup>2</sup> Fernanda Cristina Alcantara dos Santos <sup>2</sup> Renê Oliveira do Couto <sup>3</sup> Larissa Córdova Turones <sup>4</sup>
Affiliations	1 - Laboratory of Pharmacology of Natural and Synthetic Products, Department of Pharmacology, Federal University of Goiás/Institute of Biological Sciences, Campus Samambaia – Goiânia, GO, Brazil. 2 - Laboratory of Microscopy Applied to Reproduction Department of Histology, Embryology and Cell Biology/Institute of Biological Sciences/Federal University of Goiás. 3 - Pharmacotechnical Development Laboratory /Federal University of São João Del-Rei, Dona Lindu Midwest Campus, Divinópolis, Minas Gerais, Brazil. 4 - Laboratório de Patología Celular do Instituto de Anatomía, Histología y Patología da Facultad de Medicina, Universidad Austral de Chile, Valdivia.
Session	Poster presentation

## Abstract and Keywords

Rheumatoid arthritis (RA) is a chronic systemic inflammatory disease that affects 5 out of every 1,000 people worldwide and can be associated with high morbidity and mortality rates. Research has shown that medicinal plants may contain some pharmacologically active compounds capable of contributing to future advances in the pharmacotherapy of various pathologies. Celtis iguanaea (Jacq.), known as spurge, is a plant used by the population to treat inflammatory diseases and as an analgesic. This study aimed to evaluate the possible antinociceptive and/or anti-inflammatory activities of the ethyl acetate extract of the leaves of esporãode-galo (EAeEG) in the model of chronic inflammation, RA induced by Complete Freund's Adjuvant (CFA). The project was approved by the Ethics Committee for the Use of Animals at UFG under protocol number 073/21. Wistar rats weighing between 180 and 220 g were used. EAeEG was obtained by exhaustive extraction of the leaf powder in a Soxhlet apparatus for 72 hours and then concentrated under reduced pressure, with a yield of 6.75%. Treatment with EAeEG (300 mg/kg x 7 days) reduced joint edema, paw elevation time, and poly- and mononuclear infiltrates (by 66.28% and 34.1%, respectively). In addition, myeloperoxidase (MPO) activity in the synovial fluid was reduced by 44.3% and serological levels of TNF-a and IL-1 $\beta$  were reduced by 14.8% and 25%. Treatment with repeated doses also managed to reverse the impairment of liver



and kidney function caused by CFA by 57% and 30.1%, as well as reversing the reduction in glutathione levels in the liver by 57%. We can conclude that EAeEG at the dose used shows antinociceptive and anti-inflammatory effects in the chronic model of inflammation used, suggesting that the mechanisms of action involve: the reduction of pro-inflammatory cytokines, and MPO activity at the inflamed site, which may be due to a reduction in cell migration.

**Keywords**: Rheumatoid arthritis; Pain; Inflammation; esporão-de-galo.



Title	Evaluation of the protective potential of Botox in anxiety- depressive behaviors in I different chronic orofacial pain models in mice
Authors	Naiara Morais Oliveira <sup>1</sup> Héllen de Paula Nogueira Soares <sup>1</sup> Ana Claudia Gontijo Couto1, <sup>2</sup> Thays Crosara Cunha <sup>3</sup> Tamara Marques da Silva1, <sup>2</sup> Graziella Silva <sup>3</sup> Eduardo Januzzi <sup>3</sup> Cássia Regina da Silva1, <sup>2</sup>
Affiliations	<ol> <li>Grupo de Pesquisa em Analgesia e Inflamação, Laboratório de Bioquímica e Toxinas Animais, Instituto de Biotecnologia, Universidade Federal de Uberlândia/Uberlândia (MG), Brasil;</li> <li>Programa de Pós Graduação em Genética e Bioquímica, Instituto de Biotecnologia, Universidade Federal de Uberlândia/Uberlândia (MG), Brasil;Programa de Pós-graduação em Dor Orofacial/ CIODONTO/ Belo Horizonte (MG), Brasil</li> <li>Programa de Pós-graduação em Dor Orofacial/ CIODONTO/ Belo Horizonte (MG), Brasil</li> </ol>
Session	20 – Dor e Inflamação

#### Abstract and Keywords

Orofacial pain affects 25% of the world's population, located in the face, head and neck region. Temporomandibular Disorder (TMD) and trigeminal neuralgia (TN) are the main causes of chronic orofacial pain, impacting patients' lives, causing anxiety and depression. The treatment of these conditions and their comorbidities is challenging in health services, requiring more therapeutic options, such as botulinum neurotoxin-A. We investigated the protective effect of Botox on depressive-anxiety behavior associated with TMD and in the infraorbital nerve constriction (CION) model in mice. In the TMD model, animals were anesthetized with ketamine and xylazine and an injection of 20  $\mu L$  of Complete Freund's Adjuvant (CFA) (1 µg/ml) was made into the temporomandibular joint of adult male C57BL/6J mice (20-25 g). (CEUA 23117.011633/2022-55). In the NT model, animals were anesthetized and two ligatures were placed on the infraorbital root of the trigeminal nerve. On the 14th day after injection, the animals were treated with Botox 0.06U s.c in the vibrissial region. Anxious and depressive behaviors were assessed on the 19th and 20th days, with the elevated plus maze test (LCE) and the forced swimming test (FST). Botox treatment in CION reduced anxious behavior, increasing 45% of the time spent in the open arms of the EPM, while in TMD it increased the same behavior by 22%. Furthermore, in CION, depressive behavior decreased, reducing 51.5% of immobility time in FSTt, while in DTM it increased 13% of this time. Our results



show that Botox prevented the development of anxiety and depressive behaviors in the CION model, but did not prevent it in the TMD model. These opposing effects on CION and TMD reinforce the urgency of research evaluating the possible protective effects of neurotoxins in different conditions, because it is suggestive that toxins prevent anxiety- and depressive-like effects in some conditions, but not all.

**Keywords**: Pain, neurotoxin, anxiety, depression.



Title	Therapeutic potential of gold in the treatment of acute gout attack inflammation and pain in mice model
Authors	Thiago Neves Vieira <sup>1</sup> , Sofia Lafetá <sup>1</sup> , Foued Spindola <sup>2</sup> , Cássia Silva <sup>1</sup>
Affiliations	1-Grupo de Pesquisa em Analgesia e Inflamação GPANI, Laboratório de Bioquímica e Toxinas Animais LABITOX, Graduate Program in Genetics and Biochemistry, Institute of Biotechnology, Federal University of Uberlandia, 38408-100 Uberlândia (MG), Brazil;
	2-Laboratório de Bioquímica e Biologia Molecular, Graduate Program in Genetics and Biochemistry, Institute of Biotechnology, Federal University of Uberlandia, 38408-100 Uberlândia (MG), Brazil;
Session	Dor e Inflamação

# Abstract and Keywords

Gout is a prevalent form of arthritis caused by the deposition of monosodium urate crystals in the joints, leading to inflammation and pain. Its occurrence has been increasing due to the longer lifespan of the population, poor dietary habits, and an increase in a sedentary lifestyle. Gold complexes exhibit antiinflammatory properties and are used as a disease-modifying treatment for rheumatic diseases and when used as a treatment, has shown efficacy in other arthritic conditions such as osteoarthritis in phase II clinical trials. Therefore, our objective is to validate the treatment with the potassium dicyanoaurate KAu(CN)<sup>2</sup> in gout, a therapy proposed in collaboration with Gold Flex Life Science. Procedures were approved by CEUA (046/19). Male Wistar rats (200g) were used and housed in the animal facility (REBIR/UFU). They were divided into a model group, receiving a first challenge with intra-articular (ia) injection of MSU crystals (1.25 mg) in the ankle joint; the treated group received both MSU and coadministration of KAu(CN)<sup>2</sup> (SIGMA/298115) (0.8 µg); the vehicle group received an ia injection of PBS. All groups were analyzed for mechanical allodynia and joint edema up to 72h. After no pain was shown, the model and treated groups received a second MSU crystal challenge without the treatment, and all parameters were observed up to 72h. Synovial fluid was collected 4h postinjection for myeloperoxydase activity molecular analyses. Statistical analyses were performed using ANOVAs (P<0.05) in the GraphPad Prism 8 statistical program, along with GPower 3.1 to assess the sample size power. The treatment reduced mechanical allodynia from 2 to 48h, with 37% reduction 4h after administrations, articular edema 2 to 4h, and 24 to 96h with 41% of reduction 24h after administrations. Molecular analyses indicated a reduction of 57.1% for myeloperoxydase activity. These data suggest that the suggested gold treatment has promising therapeutic potential against gout attacks.

**Keywords**: Gout, Potassium dicyanoaurate, Gold treatment, Inflammation.



Title	Sesquiterpene 9-deoxymuzigadial modulates inflammation and fibrosis induced by subcutaneous sponge implants in mice
Authors	Bruno Antonio Ferreira <sup>1,2</sup> Francyelle Borges Rosa de Moura <sup>3</sup> Isabella Silva Cassimiro <sup>2</sup> Taís de Campos Lima <sup>2</sup> João Henrique Ghillardi Lago <sup>1</sup> Fernanda de Assis Araújo <sup>2</sup>
Affiliations	1- Center for Natural and Human Sciences, Federal University of ABC, São Paulo 09210-170, Brazil 2- Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, 38408-100, Brazil 3- Federal University of Catalão, Catalão, 75704-020, Brazil
Session	20 - Dor e Inflamação

### Abstract and Keywords

The phytochemical composition from the leaves and barks of Drimys brasiliensis, a native species of Brazilian flora, reveals a high content of compounds from the sesquiterpene class, many of which show anti- inflammatory action. The aims of this study were to investigate the in vitro cytotoxicity potential of the sesquiterpene 9-deoxymuzigadial and the effects of its daily administration (0.1, 1 or 10 µg of 9- deoxymuzigadial diluted in 0.5% DMSO) during a chronic inflammatory process induced by sponge implants in C57BL/6 male mice (n = 64animals, CEUA 049/21). The implanted material served as a scaffold for the development of a fibrovascular tissue, allowing the concomitant analysis of inflammatory and fibrogenic processes. One sponge disc per animal was aseptically implanted into the dorsal region and nine days post-implantation collected and processed to biochemical and histological analyzes. The compound showed no cytotoxic activity in RAW 264.7 macrophages, as assessed by the MTT method. In implants treated with 0.1  $\mu g$  of 9-deoxymuzigadial, we observed a reduction in NAG activity (2.01  $\pm$  0.12 versus 3.13  $\pm$  0.38) and in the mean number of mast cells (0.28  $\pm$  0.08 versus 1.15  $\pm$  0.15), observed in toluidine blue stained section, when compared to control. On the other hand, at the highest dose, we observed an increase in NAG and MPO activities, suggesting a greater inflammatory infiltrate. In the groups treated with 0.1 and 10 µg we observed a pro-fibrogenic effect, with an increase in collagen synthesis (74% and 64%, respectively) and deposition (39% and 43%, respectively) when compared to control. In addition, in the implants treated with 0.1  $\mu$ g, we observed a higher proportion of thinner collagen fibers, usually identified as type III collagen. Our results show that, at least at the lowest dose evaluated, the sesquiterpene 9- deoxymuzigadial was able to attenuate some of the inflammatory parameters and incite the fibrogenesis.

**Keywords:** sesquiterpenes; natural products; inflammation; fibrogenesis; plant metabolites



Title	Effect of acute aquatic exercise on humoral immune response against <i>Dermatophagoides farinae</i> in postmenopausal women
Authors	Thiago Alves de Jesus <sup>1</sup> Tássia Magnabosco Sisconeto <sup>2</sup> Alessandro Sousa Correa <sup>1</sup> Laura Alves Ribeiro de Oliveira <sup>1</sup> Rafael de Oliveira Resende <sup>1,3</sup> Guilherme Morais Puga <sup>2,4</sup> Ernesto Akio Taketomi <sup>1</sup>
Affiliations	<ul> <li>1 - Institute of Biomedical Sciences, Federal University of Uberlândia,</li> <li>Uberlândia, Brazil</li> <li>2 - Faculty of Medicine, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>3 - Oswaldo Cruz Institute, Fiocruz, Rio de Janeiro, Brazil</li> <li>4 - Faculty of Physical Education and Physiotherapy, Federal University of Uberlândia, Uberlândia, Brazil</li> </ul>
Session	Immunology

# and Keywords

Exercise is a recommended non-pharmacological strategy for preventing and treating diseases. However, intense or prolonged exercise can play a suppressive role in the immune system. Salivary IgA (sIgA) concentration is a marker for immune impairment post-exercise since decreased levels of this antibody correlate to a higher incidence of infections after exercise. However, many respiratory symptoms after exercise can stem from asthma and rhinitis. Variation of sIgA antibodies specific to allergens triggered by physical exercise has not been investigated so far. This study aimed to evaluate the effect of acute water exercises on the variation of salivary-specific IgA antibodies to house dust mites in postmenopausal women. This study was approved by the Federal University of Uberlândia's Ethics Committee (5.449.371). A total of 22 postmenopausal women had anthropometric data collected and were submitted to an acute water exercise (AWE) session, with intensity varying between moderate and vigorous, and a relaxation session (control). Saliva samples were collected before and after the sessions, and anti-Dermatophagoides farinae sIgA was analyzed. Later, participants answered a clinical questionnaire for rhinitis and had blood samples collected for detection of D. farinae specific-IgA and -IgE antibodies. The results indicated that AWE did not cause significant variations in the levels and secretion of anti-D. farinae sIgA. Also, these antibodies had high daily variability as well as among individuals. Specific sIgA did not correlate with rhinitis symptoms, specific serum IgE and IgA, or any other analyzed demographic, metabolic, or body composition data. However, specific serum IgA directly correlated with allergic rhinitis symptoms, although inversely related to the body mass index and the volunteers' body fat percentage. Therefore, allergen-specific sIgA levels may not be affected by AWE, suggesting no heightened susceptibility to allergic responses after sessions.

**Keywords:** Aquatic Exercise; IgA; House Dust Mites; Postmenopausal Women



Title	Biological characterization of the bristle extract from the fire caterpillar ( <i>Dirphia moderata</i> ) and the green stinging caterpillar ( <i>Automeris illustris</i> )
Authors	Álefe Cardoso Cruz <sup>1</sup> , Carolina de Souza Gardenghi <sup>1</sup> , Marielle Máximo Barbosa <sup>2</sup> , Licia Ludendorff Queiroz <sup>3</sup> , Fernanda Maria Santiago <sup>1</sup>
Affiliations	<ul> <li>Laboratory of Immunoparasitology "Dr. Mário Endsfeldz Camargo", Institute of Biomedical Sciences, Federal University of Uberlândia, Minas Gerais, Brazil.</li> <li>Laboratory of Allergy and Clinical Immunology, Institute of Biomedical Sciences, Federal University of Uberlândia, Minas Gerais, Brazil.</li> <li>Laboratory of Molecular Microbiology, Institute of Biomedical Sciences, Federal University of Uberlândia, Minas Gerais, Brazil.</li> </ul>
Session	Immunology

### Abstract and Keywords

The investigation of natural products is crucial for the identification of novel therapeutic strategies against diverse pathologies, particularly cancer and infectious diseases. Bioactive compounds sourced from plants, animals, and microbes exhibit potential for innovative treatments owing to their varied biological activities. Although limited studies on D. moderata exist, research on related species underscores the potential of these natural compounds in developing effective treatments, particularly in highlighting antivenom effects and enzymatic activities. The caterpillars (D. moderata and A. illustris) were collected in the city of Uberlândia, Minas Gerais. The bristles were extracted and solubilized with physiological saline solution (0.9% NaCl). Venom protein concentrations were determined using the method with bovine serum albumin as a standard. To ascertain the protein profile, the extracts underwent SDS-PAGE polyacrylamide gel electrophoresis. Biological activities of the extracts were assessed through hemolysis assays to determine hemolytic activity, MTT assays for cell viability (9,37  $\mu$ g/ $\mu$ l, 18,75  $\mu$ g/ $\mu$ l, 37,5  $\mu$ g/ $\mu$ l, 75  $\mu$ g/ $\mu$ l, 150  $\mu$ g/ $\mu$ l, 300  $\mu g/\mu I$ ), disk diffusion assays for antimicrobial activity against bacteria (50  $\mu g/\mu I$ e 100 μg/μl), and MTT assays for antitumor activity against lung (A549) and prostate cancer (PC3) cells. The extracts exhibited no significant hemolytic activity at any of the tested concentrations. Similarly, in the cell viability assay, none of the concentrations displayed a viability below 70%, indicating that the extracts do not possess a toxic profile towards the tested cells. The extracts did not exhibit significant hemolytic activity at any of the tested concentrations, and in the cell viability assay, none of the concentrations yielded a viability index below 70%. This observation demonstrates that the extracts do not possess a toxic profile towards the tested cells. When exposed to PC3 and A549 tumor cells, the D. moderata extract displayed significant antitumor activity, particularly at the concentration of 300 µg/µl (78% and 88%) for A549 and PC3, respectively.



Title	In vitro evaluation of the antitumour, antimicrobial and antiparasitic effects of protein fractions from <i>Cycas revoluta</i> seeds
Authors	Carolina de Souza Gardenghi¹, Jhoan David Aguillón Torres¹, Lícia Ludendorff Queiroz², Fernanda Maria Santiago¹
Affiliations	1 – Department of Institute of Biomedical Sciences, Laboratory of Immunoparasitology "PHD Mário Endsfelfz Camargo", Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil.
	2 - Department of Institute of Biomedical Sciences, clinical bacteriology laboratory, Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil.
Session	Immunology (poster presentation)

### Abstract and Keywords

Cycas revoluta, known as "garden sago" (Cycadaceae) is a gymnosperm plant cultivated mainly for its ornamental appeal. Recent research has highlighted its remarkable immunomodulatory potential, evidenced by an isolated component extracted from its leaves and stems. Although the protein fraction of the seeds has not yet been addressed in the literature, this study aims to investigate its potential against lung and prostate cancer, as well as the protozoan Toxoplasma gondii, and the bacteria Escherichia coli and Staphylococcus aureus. To this end, protein fractions were isolated from the crude extract using chromatographic techniques, resulting in the identification of four types of protein compounds that will be characterised using spectroscopic techniques. These compounds, named Peak 1, Peak 2, Peak 3 and Associated peak. In vitro studies on the antitumour cytotoxic effects were conducted using two assays: one directly on cancer cells and the other allowing cell development with the drug. To assess the toxoplasmocidal potential of these compounds, it was investigated which protein fraction has the ability to inhibit the invasion and proliferation of the parasite inside the cell, while antibiogram tests were carried out to see if they inhibit the growth of bacteria. As a result, each fraction showed biological activity against some pathogen, the crude extract and the associated peak showing the best results against tumour strains. The Peak 2 fraction stood out in the results against anti-Toxoplasma gondii activity, while Peak 1 showed efficacy against antibacterial activity. Considering the promising results of the *C. revoluta* protein fractions against the pathogens studied, we also investigated how these samples act against cancer and the parasite, in order to deepen our understanding of the immunology related to these diseases.

**Keywords**: *Cycas revoluta*, Lung cancer, Prostate cancer, *Toxoplasma gondii*, *Escherichia coli*, *Staphylococcus aureus*.



Title	Selection of peptides by phage display technology for Salmonella Typhimurium detection
Authors	Gabriela Zangari Cardoso <sup>1</sup> Fabiana De Almeida Araújo Santos <sup>2</sup> Pedro Lucas Figueiredo Nunes <sup>2</sup> Bruna Patrícia Do Couto <sup>3</sup> Maria Vitória Mendes Felix Costa <sup>1</sup> Belchiolina Beatriz Fonseca <sup>1, 2</sup>
Affiliations	1- Graduate Program in Veterinary Science, Faculty of Veterinary Medicine, Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil 2- Laboratory of Nanobiotechnology prof. doctor Luiz Ricardo Goulart, Institute of Biotechnology, Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil 3- Graduate Program in Applied Immunology and Parasitology, Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil
Session	21 Imunologia

# Abstract and Keywords

Salmonella Typhimurium (ST) is one of the major foodborne pathogens globally, causing gastroenteritis in both animals and humans. Current tests to identify this serotype are expensive and laborious. The development of rapid, safe, and specific identification methods is important for controlling the disease caused by ST. Its impact demands rigorous surveillance, research, and education to ensure public safety. Having said that, the aim of this project was to discover whether the bacteriophages selected by phage display technology could recognize ST in chicken carcasses. We performed biopanning using the phage display library to bind to ST. Consequently, we isolated 21 phages and through ELISA testing and sequencing, we selected two highly specific phages, B-4 and E-6 for colonies of ST. These phages were amplified, and we conducted phage-ELISA on chicken carcasses. To perform the test, we took three chicken carcasses after dripping and inoculated them with different strains of Salmonella (15 strains of ST and 15 strains of other serotypes), in addition to the controls. Carcass preparation was carried out according to ISO 6579:2002 standards using buffered peptone water. For the phage ELISA, we sensitised a 96-well microplate with B-4 or E-6 and tested the buffered peptone water from the carcasses infected or not with ST. Phage B-4 presented an area under curve (AUC) of 90.48%, and E-6, 87.45%. Phage B-4 exhibited a sensitivity of 86% and specificity of 82.6%. It is known from the literature that phage ELISA can yield non-specific reactions and lower AUC than the selected phage-binding peptide. Therefore, based on these results, we are confident in selecting ST-binding peptides B-4 to be synthesized for future tests. This test demonstrated that we have a peptide with significant potential to be highly specific as an ST ligand and could serve as an alternative for identifying this pathogen in food and clinical samples.

Keywords: Bacteriophage, Recognize, Poultry, ELISA



Title	Exploring novel microRNAs as regulators of innate immunity during intracellular pathogen infection
Authors	Marya Fernanda Santos Lopes João Vitor Eleuterio da Silva Jhoan David Aguillón Torres Tiago Wilson Patriarca Mineo Caroline Martins Mota
Affiliations	Universidade Federal de Uberlândia, Uberlandia, Brazil
Session	21 - Imunologia

#### Abstract and Seywords

MicroRNAs (miRNAs) are small endogenous non-coding RNAs that post-transcriptionally regulate gene expression and are ubiquitous in eukaryotic cells. Recent research has delved into their regulatory roles across various biological processes, with promising implications as disease biomarkers and therapeutic targets. Our study aims to elucidate the involvement of novel miRNAs in regulating the host's innate immune response during infection by intracellular pathogens. Transcriptome analysis of murine RNAseq data revealed elevated expression levels of miRNA6236 in cells infected with *Toxoplasma gondii* and *Neospora caninum* compared to uninfected cells. Using the Crispr/Cas9 system to deplete miRNA6236 in L929 cells (miRNA6236-/-), we observed impaired cell cytokinesis, leading to defective replication and inviability in homozygous clones. Our findings underscore the pivotal role of miRNA6236 in cellular development and underscore its potential as a therapeutic target. This study suggests the potential of miRNAs in modulating innate immunity and unveils new avenues for exploring their interactions in host-pathogen responses.

**Keywords**: microRNA, CRISPR-CAS9, Transcriptome



Title	The induction of heme oxygenase-1 is able to improve adverse pregnancy outcome in congenital toxoplasmosis in mice
Authors	Iasmin Aparecida Cunha Araújo <sup>1</sup> Marcos Paulo Oliveira Almeida <sup>1</sup> Anna Laura Gomes de Jesus <sup>1</sup> Maria Eduarda Barbieri Machado Soares <sup>1</sup> Neide Maria da Silva <sup>1</sup>
Affiliations	Laboratory of Immunopathology, Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlandia, MG, 38400-902, Brazil
Session	21 - Imunologia

# Abstract and Keywords

Congenital infection with *Toxoplasma gondii* leads to abnormalities in mouse gestation, which is related to a pro-inflammatory immune response at the maternal-fetal interface. Thus, to prevent abortion or embryo resorption, a regulatory immune response is necessary. Heme oxygenase-1 (HO-1) is an enzyme that catalyzes heme oxidation to produce metabolites with antiinflammatory, cytoprotective, and anti-apoptotic effects which are involved in the success of gestation under infection with Listeria monocytogenes. Therefore, the aim of this study was to investigate the effect of HO-1 induction (CoPPIX treatment) or inhibition (ZnPPIX treatment) on the gestational outcome in congenital toxoplasmosis. BALB/c and C57BL/6 mice are known to be susceptible to congenital toxoplasmosis, being that C57BL/6 present higher resorption rate in relation to BALB/c mice. For this purpose, females of both mouse lineages were infected on the day of vaginal plug detection or on day 8 of gestation and, C57BL/6 received an inductor and BALB/c an inhibitor of HO-1 on days 5, 6 and 7 or 11, 12, and 13 of gestation, respectively. On day 8 of gestation and infection BALB/c mice that received ZnPPIX-treatment presented higher resorption rate in comparison with infected untreated mice of the same lineage and; C57BL/6 that received CoPPIX showed lower resorption rate in comparison with infected untreated mice of the same mouse lineage. On day 14 of gestation and 6 of infection, BALB/c mice that received ZnPPIX showed a lower mean fetal weight compared to untreated or non-infected mice. In conclusion, Heme oxygenase-1 induction are involved in the improvement of gestational outcome in congenital toxoplasmosis.

**Keywords**: *Toxoplasma gondii*; Heme oxigenase -1; congenital toxoplasmosis; CoPPIX; ZnPPIX.



Title	The intestinal immune response in acute Toxoplasma gondii infection in the absence of CCR5
	Maria Eduarda Barbiéri Machado Soares Ester Cristina Borges Araujo
	Júlia Maria Fernandes Pituba Rafael Queiroz Freitas Pereira
Authors	Lucas Maurício Lopes de Navasquez Anna Laura de Jesus Gomes
	Iasmin Aparecida Cunha Araújo Neide Maria da Silva
Affiliations	Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlandia
Session	21 - Imunologia

## Abstract and Keywords

Approximately one-third of the population is infected with *Toxoplasma gondii*, an obligate intracellular parasite of warm-blooded animals that invades various types of cells. CCR5, is a chemokine receptor important in T. gondii-induced IL-12 production by Dendritic Cells through the cyclophilin-18, an unique chemokine mimic. The main chemokine ligands of CCR5 are CCL3 (MIP-1a), CCL4 (MIP-1b) and CCL5 (RANTES), which play an important role in cell recruitment to the local of inflammation. CCR5 deficient mice (CCR5 $^{-/-}$ ) are highly susceptible to oral T. gondii infection, developing severe intestinal lesions. To elucidate the mechanisms related to susceptibility in the acute phase of infection, the intestines of CCR5<sup>-/-</sup> animals were morphologically evaluated. CCR5<sup>-/-</sup> mice in the C57BL/6 Background and C57BL/6 mice were infected with 5 ME-49 T. gondii cysts by oral route and the survival and morbidity score accompanied over the time. The intestines were collected on day 7 of infection to histological evaluation. As previously observed, the CCR5<sup>-/-</sup> mice were more susceptible than C57BL/6 mice under infection. The small intestine of infected animals showed inflammatory cell infiltrations leading to disturbed mucosal architecture, however, knockout mice showed more severe intestinal lesions with crypt abscess and superficial necrosis of the intestinal villi. Additionally, CCR5<sup>-/-</sup> mice showed lower numbers of goblet cells and intraepithelial lymphocytes than uninfected CCR5<sup>-/-</sup> mice and lower numbers of goblet cells in comparison with C57BL/6 mice on day 7 of infection. In conclusion, the CCR5 chemokine receptor is essential to control, at least partially the intestinal lesions in oral T. gondii infection.

**Keywords**: Toxoplasma gondii, intestine, acute infection, CCR5.



Title	Dense granule proteins of <i>Neospora caninum</i> are capable of modulating the host immune response through the production of IL12 and are necessary for the parasite virulence
Authors	Jhoan David Aguillón Torres <sup>1</sup> , Ruth Opeyemi Awoyinka <sup>1</sup> , Ana Paula Navarro Gonçalves <sup>1</sup> , Caroline Martins Mota <sup>1</sup> , Peter John Bradley <sup>2</sup> , Tiago Wilson Patriarca Mineo <sup>1</sup> .
Affiliations	<ul> <li>1 - Laboratory of Immunoparasitology "Dr. Mário Endsfeldz Camargo", Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil.</li> <li>2- Department of Immunology, Microbiology and Molecular Genetics, University of California, Los Angeles, USA.</li> </ul>
Session	Immunology

# Abstract and Keywords

Neospora caninum is an apicomplexan parasite that induces reproductive disorders in cattle and neuromuscular disease in dogs. This parasite requires a synchronized protein machinery for cellular invasion, intracellular replication, and survival. This machinery includes the secretion of specialized proteins such as rhoptries (ROPs), micronemes (MICs), and dense granule proteins (GRAs). GRAs are conserved proteins across the Apicomplexa phylum and are essential for the establishment of the parasitophorous vacuole (PV) and for protein traffic to the host cell. In this study, we aimed to describe four new GRAs based on Toxoplasma gondii orthologs and one exclusive protein of N. caninum, in order to elucidate their function using CRISPR/Cas9 technology. The five selected proteins were endogenously tagged with hemagglutinin epitopes and, subsequently, parasite lines with genetic ablations of the selected genes were generated. For functional analyses, we performed in vitro experiments using bone marrow-derived macrophages (BMDM) to determine mRNA and cytokine levels and In vivo experiments using C57BL/6 mice to elucidate the role of these proteins in virulence and cytokine release. The localization of the five selected proteins into the PV was confirmed. In BMDM, we observed significant changes in mRNA levels of Nos2 and Nox2 for some GRAs. In addition, the IL12p40 cytokine release was significantly reduced in BMDMs infected with all knockout parasites and this was partially confirmed in vivo during acute infections. Finally, some of the knockout parasites induced reduced parasite burden during the acute phase of the infection, while all the selected parasites presented reduced chronic phase brain parasitism. These results indicate the potential role of these proteins in the host immune response modulation and parasite virulence, and further studies are underway to unravel the mechanisms by which these proteins are so relevant to the N. caninum life cycle.

**Keywords**: Apicomplexa, CRISPR/Cas9, virulence, cytokine release, parasite burden.



Title	Assessment of oxidative stress generated by L-amino acid oxidase from Bothrops moojeni (Pollonein-LAAO) in tumor progression
Authors	Morais LP <sup>1</sup> , <sup>2</sup> , Polloni L <sup>2</sup> , Costa TR <sup>3</sup> , Oliveira VQ <sup>1</sup> , <sup>3</sup> , Ferreira EL <sup>1</sup> , <sup>3</sup> , Ávila VMR <sup>1</sup> , <sup>3</sup>
Affiliations	1 - Laboratory of Biochemistry and Animal Toxins, Institute of Biotechnology - Federal University of Uberlândia, Uberlândia, Brazil. 2 - Institute of Biomedical Sciences, Federal University of Uberlândia – UFU, Uberlândia, MG, Brazil 3 - Institute of Biotechnology, Federal University of Uberlândia – UFU, Uberlândia, MG, Brazil
Session	22 Sinalização e Terapêutica do Câncer

# Abstract and

Neoplastic cells elevate the generation of reactive oxygen species (ROS), thereby inducing signaling pathways that favor tumorigenic promotion or result in oxidative stress. Due to the vulnerability of these cells to intracellular ROS increases, exploring exogenous inducers of ROS, such as the enzyme L-amino acid oxidase, may represent significant biotechnological tools and therefore deserve better investigation. In this study, we purified Pollonein-LAAO from the venom of the Bothrops moojeni snake, verified its homogeneity, quantified posttreatment ROS increase, and analyzed oxidative damage in prostate cancer cell lines (PC3) and normal cells (PNT2), as well as the recovery capacity of these cells after oxidative stress. Isolation was carried out through chromatography, homogeneity verification by electrophoresis, cell viability assays using MTT to evaluate cellular effects and recovery capacity, and ROS quantification by labeling with H2DCFDA followed by cytometry analysis. The toxin was obtained with a yield of 1.6% and a molecular weight of 44 kDa. The treatment was able to increase ROS production at all tested concentrations, decreased viability and recovery capacity of PC3 cells more intensely when compared to PNT2. These results emphasize the relevance of oxidative stress as a potential therapeutic target in cancer treatment and underscore the need for further investigation of Pollonein-LAAO in this context.

**Keyword**: Snake-venom, L-aminoácido oxidase, ERO's.



Title	In vitro antitumor and anti-metastatic properties of BthMP, a P-I metalloproteinase isolated from <i>Bothrops moojeni</i> snake venom against lung cancer cells
Authors	Emanuelle Lorrayne Ferreira <sup>1</sup> , Vinícius Queiroz Oliveira <sup>1</sup> , Luísa Carregosa Santos <sup>2</sup> , Thiago Macedo Lopes Correia <sup>2</sup> , Leonardo Oliveira Silva Bastos Andrade <sup>2</sup> , Samuel Cota Teixeira <sup>3</sup> , Sarah Natalie Cirilo Gimenes <sup>4</sup> , Mônica Colombine <sup>4</sup> , Lucas Miranda Marques <sup>2</sup> , Patrícia Bianca Clissa <sup>4</sup> , Lorena Pinheiro Morais <sup>1</sup> , Veridiana de Melo Rodrigues Ávila <sup>1</sup> , Cristiani Baldo <sup>5</sup> , Daiana Silva Lopes <sup>2</sup>
Affiliations	¹Laboratory of Biochemistry and Animal Toxins, Institute of Genetics and Biochemistry, Federal University of Uberlandia (UFU), Uberlandia-MG, Brazil;  ²Institute Multidisciplinary in Health, Federal University of Bahia (UFBA), Vitoria da Conquista, BA, Brazil;  ³Department of Immunology, Institute of Biomedical Sciences, Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil;  ⁴Laboratory of Immunopathology, Institute of Butantan, São Paulo, SP, Brazil;  ⁵Department of Biochemistry and Biotechnology, State University of Londrina, Paraná, PR, Brazil.
Session	22- Sinalização e Terapêutica do Câncer

### Abstract and Keywords

Lung cancer is the most common cancer and the leading cause of mortality worldwide. New treatment approaches that are less aggressive and more efficient are needed. Thus, snake venom presents a promising approach for cancer treatment, due to its variety of biomolecules with potential therapeutic applications. An interesting example of this are metalloproteinases, which are highlighted for their antitumor potential. In this study, we aimed to characterize the antimetastatic effect of BthMP, a P-I metalloproteinase isolated from Bothrops moojeni venom, on A549 lung adenocarcinoma cell line and BEAS-2B human bronchial epithelial cell line. Cellular viability assays were performed using MTT, LDH, and colony formation assays. Additionally, migration, invasion, adhesion assays, as well as measurements of reactive oxygen species (ROS) and nitric oxide (NO) were conducted. The results revealed that BthMP decreased colony formation by clonogenic assays and elevated LDH levels in A549 cells, showing a cytotoxic effect at concentrations of 40 and 5  $\mu$ g/mL. BthMP specifically hindered invasion, migration, and adhesion while augmenting levels of reactive oxygen and nitric oxide species in tumor cells. Additionally, BthMP pre-treated with EDTA failed to diminish A549 cell migration, indicating that BthMP requires the catalytic domain to execute its cytotoxic effect. Taken together, the results presented here demonstrate the ability of P-I SVMP BthMP in inhibiting the proliferation, invasion, adhesion and migration of lung cancer cells A549, an important process involved in tumor growth and metastasis. These findings demonstrate that BthMP possesses a significant antitumor effect on lung cancer, presenting a valuable biotechnological asset for innovative therapeutic approaches in lung cancer treatment.

**Keywords**: adhesion, antitumoral, cancer, metalloproteinase, BthMP, snake venom.

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Title	Aptamers A6 and A11 with potential theranostic application in prostate cancer
Authors	Maria Paula Silva Simião Sabrina Lorenti Vivian Alonso Goulart
Affiliations	Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart Filho, Institute of Biotechnology (IBTEC), Federal University of Uberlândia, Uberlândia, Brazil
Session	Signaling and Therapy of Cancer

## Abstract and Keywords

Prostate cancer (PCa) is the second most common type of cancer in men. Metastatic PCa is considered a more advanced type of tumor with a worse prognosis, therefore, it is important to diagnose the disease at an early stage, as well as choose the best treatment option for the patient. Aptamers are oligonucleotides that allow the search for biomarkers in target cells and can also provide information related to diagnosis, therapy, and disease mechanisms. The study's objective was to evaluate, using the resazurin method, whether there was a change in cell viability and proliferation of normal and tumoral prostate lines when incubated with aptamers A6 and A11 in three different concentrations (1,5; 2,5 e 5  $\mu$ M) for 24h and 48h. The highest concentration of aptamer A6 used (5 µM) showed cytotoxicity in the tumor lineage after 48h and was not cytotoxic to the normal prostate lineage. The A11 aptamer did not influence cell viability and proliferation characteristics in both prostate lineages. These results suggest that the A6 aptamer can be used as therapy in patients with PC, and the results shown by the A11 aptamer may suggest the use of this aptamer as a possible diagnostic tool.

**Keywords**: Prostate cancer; Aptamers; Theranostics.



Title	Use of serum from obese individuals in the in vitro study of the cell nucleus in a human glioblastoma line
Authors	Phelipe Elias da Silva <sup>1</sup> Maria Eduarda Marques <sup>2</sup> Natália Ferreira Silva <sup>3</sup> Fernanda Mascarenhas <sup>4</sup> Tiago Kerr <sup>5</sup> Natália Cunha <sup>5</sup> Tatiana Tomiosso <sup>1</sup> Robson José Júnior <sup>4</sup> Ricardo Rodrigues <sup>6</sup> Renata Zanon <sup>1</sup>
Affiliations	<ul> <li>1 - Postgraduate Program in Applied Cellular and Structural Biology, Institute of Biomedical Sciences, Federal University of Uberlândia; Uberlândia, MG; Brazil</li> <li>2 - Institute of Biotechnology, Federal University of Uberlândia; Uberlândia, MG; Brazil</li> <li>3 - Postgraduate Program in Applied Cellular and Structural Biology, Institute of Biology, UNICAMP; Campinas, SP; Brazil</li> <li>4 - Postgraduate Program in Genetics and Biochemistry, Institute of Biotechnology, Federal University of Uberlândia; Uberlândia, MG; Brazil</li> <li>5 - Postgraduate Program in Statistics, Institute of Mathematical and Computer Sciences, University of São Paulo; São Carlos, SP; Brazil</li> <li>6 - Faculty of Medicine, Department of Endocrinology and Bariatric Surgery at Hospital de Clinicas, Federal University of Uberlândia; Uberlândia, MG; Brazil</li> </ul>
Session	Pôster

## and Keywords

The development and support of the tumour environment is a complex and multifaceted process involving cellular and molecular pathways. Nuclear alterations, such as genetic instability and changes in chromatin organization, play a significant role in this process. Studies have reported that nuclear alterations associated with obesity can promote DNA damage, increase resistance to apoptosis and contribute to angiogenesis, thus favouring the progression of tumour cells. In addition, the reduction in heterochromatin can lead to a loss of nuclear rigidity and induce changes in gene expression, facilitating future chromosomal translocations and promoting genetic instability. Gliomas are the most common primary brain tumours. Among them, glioblastoma is one of the most aggressive malignant neoplasms, with an average survival of 12 and 15 months after diagnosis. Although there is evidence linking obesity to various types of cancer, the relationship between obesity and gliomas remains unclear. This study analyzed nuclear changes in human glioblastoma cell cultures when exposed to serum from individuals with obesity. In this context, serum from 12 volunteers, 6 individuals with obesity and 6 with normal weight, was collected



and used to evaluate genetic instability using the micronucleus assay, a marker of genotoxic stress, and to evaluate chromatin organization using fluorochrome DAPI (Ethical Committee: 45449821.2.0000.5152). Cells treated with serum from the obese group showed a higher frequency of micronuclei, a larger nuclear area and a lower presence of heterochromatin. Further studies are needed to identify the specific blood components that can lead to these changes, but these results indicate the obesogenic environment favors genetic instability and induces changes in chromatin organization, which can influence a cascade of events that lead to the progression of glioblastoma, highlighting an association between obesity and gliomas.

**Keywords**: Brain cancer, Obesity, Cell nucleus



Title	Analysis of the influence of serum from obese individuals on the in vitro study of cell viability in human glioblastoma
Authors	Laura Rangel Oliveira Moraes¹ Phelipe Elias da Silva² Natália Ferreira Silva³ Maria Eduarda Marques⁴ Fernanda Mascarenhas⁵ Tiago Kerr⁶ Natália Cunha⁶ Tatiana Tomiosso² Robson José Junior⁵ Ricardo Rodrigues³ Renata Zanon²
Affiliations	1 – Institute of Biology, Federal University of Uberlândia; Uberlândia, MG; Brazil 2 – Postgraduate Program in Applied Cellular and Structural Biology, Institute of Biomedical Sciences, Federal University of Uberlândia; Uberlândia, MG; Brazil 3 – Postgraduate Program in Applied Cellular and Structural Biology, Institute of Biology, UNICAMP; Campinas, SP; Brazil 4 – Institute of Biotechnology, Federal University of Uberlândia; Uberlândia, MG; Brazil 5 – Postgraduate Program in Genetics and Biochemistry, Institute of Biotechnology, Federal University of Uberlândia; Uberlândia, MG; Brazil 6 – Postgraduate Program in Statistics, Institute of Mathematical and Computer Sciences, University of São Paulo; São Carlos, SP; Brazil 7 - Faculty of Medicine, Department of Endocrinology and Bariatric Surgery, Clinical Hospital, Federal University of Uberlândia; Uberlândia, MG; Brazil
Session	Pôster

### and Keywords

Malignant neoplasms arising from central nervous system (CNS) cells are associated with increased mortality and morbidity, accounting for around 25% of total cancer deaths. Currently, obesity has been associated with an increased incidence of all CNS tumors. Data indicates that this disorder may be responsible for approximately 20% of all cancer cases. Therefore, the present study evaluated, in vitro, the viability of the human glioblastoma cell line, U-87MG, after conditioning with serum from patients with grade I and II obesity. 31 obese patients and 31 eutrophic patients were recruited for blood collection. Cells were cultured with DMEM supplemented with 10% fetal bovine serum and 1% antibiotic at a concentration of  $2 \times 10^4$  cells per well. After reaching 80% confluence, they were treated with 10% of the patient's serum and kept in an incubator for 24 hours. The cell viability assay was carried out, in triplicate, by Alamar Blue, with the absorption reading taken at wavelengths of 570nm and 600nm. Significance differences were determined by the student's t-test using



the GraphPad Prism 6.0 program, occupying a 95% confidence interval (p<0.05). The results showed an increased viability of tumor cells when treated with serum from patients with obesity (p=0.0314). Although additional studies must be carried out to better understand the relationship between obesity and cancer, the present work showed that serum from obese individuals may contribute to glioma development.

Keywords: Brain cancer, Obesity, Glioblastoma



Title	Analysis of hematological parameters and their relationship with histopathological alterations in a murine model of lingual carcinogenesis induced by 4-nitroquinoline-1-oxide and ethanol
Authors	Isabella Moura Pereira <sup>1</sup> ; Anaíra Ribeiro Guedes Fonseca Costa <sup>2</sup> Sérgio Vitorino Cardoso <sup>2</sup> ; Adriano Mota Loyol <sup>2</sup> ; Debora de Oliveira Santos <sup>2</sup> Paulo Rogério de Faria <sup>2</sup>
Affiliations	<ol> <li>I - Institute of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>Department of Oral and Maxillofacial Pathology, Federal University of Uberlândia, Uberlândia, Brazil</li> </ol>
Session	Session III

### Abstract and Keywords

Peripheral blood biomarkers are significant prognostic predictors for several human tumors, including oral squamous cell carcinoma (OSCC), for they reflect the dynamics between anti-tumor and tumor promoting effects of the inflammatory response. Among the parameters employed in this assessment, the neutrophil-to-lymphocyte ratio (NLR) stands out, in which increased levels are associated with unfavorable outcomes. To investigate such relationship in oral carcinogenesis, 120 C57BI/6J mice were treated with 4-nitroquinoline-N-oxide (4NQO) at 100ug/mL for 10 weeks followed by 8% ethanol (EtOH) for 15 weeks. Males (n=60) and females (n=60) were subdivided into four groups with 15 animals each: treated with propylene glycol (PPG) and water (H2O); treated with PPG and EtOH; treated with 4NQO and H2O; and treated with 4NQO and EtOH (CEUA-UFU, #100/18). At the end of the 25-week treatment period, tongues and blood samples were collected. Tongues were submitted to histopathological analyses and blood samples were processed for hematological profiling and calculation of NLR values. Approximately 92.9% of the male mice from 4NQO/H2O and 4NQO/EtOH groups developed OSCC; the remaining had epithelial dysplasia (ED). In females, the occurrence of OSCC was observed in 66.7% of the 4NQO/H2O group and in 60% of the 4NQO/EtOH group. Differences in OSCC incidence between males and females of the 4NQO/EtOH group were significant ( $X^2$  test, p=0.04). The median NLR significantly increased from control animals to animals with OSCC in males and females of the 4NQO/H2O and 4NQO/EtOH groups (Kruskal-Wallis test, p<0.05). Females of the 4NQO/H2O group with ED also had higher NRL values (0.35) when compared to controls (0.17), a significant difference (Kruskal-Wallis test, p < 0.05). Considering that the NLR values altered during experimental carcinogenesis in the murine model, it could be further explored as a biomarker for oral cancer development and progression.

**Keywords**: mice, 4-nitroquinoline-N-oxide, inflammation, carcinogenesis, oral cancer



Title	Immunohistochemical expression of H3K9ac and H3K27ac in premalignant and malignant tongue lesions of wild-type and Nos2-knockout mice treated with 4NQO
Authors	Wender Rodrigues Nazário <sup>1</sup> Marcondes Pedro Souza Novais <sup>1</sup> Débora de Oliveira Santos <sup>1</sup> Anaíra Ribeiro Guedes Fonseca Costa <sup>1</sup> Pedro Victor Silva Duarte <sup>2</sup> Tássio Edno Atanásio Pitorro <sup>3</sup> Lúbia Cristina Fonseca <sup>1</sup> Sérgio Vitorino Cardoso <sup>1</sup> Adriano Mota Loyola <sup>1</sup> Paulo Rogério de Faria <sup>1</sup>
Affiliations	<ol> <li>Department of Oral and Maxillofacial Pathology, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>Department of Diagnosis and Surgery, Júlio de Mesquita Filho São Paulo State University, Araçatuba, Brazil</li> <li>Department of Periodontology, Federal University of Uberlândia, Uberlândia, Brazil</li> </ol>
Session	Session II

#### Abstract

and

Nitric oxide is an important regulator of the epigenetic landscape of cellular homeostatic and pathological states by promoting post-translational modifications of the epigenome-modulating enzymes. Studies have shown that histone acetylation are highly expressed in oral cancer samples. Another study has shown that nitric oxide signaling induces autoacetylation of p300 and then the histone H3 hyperacetylation in downstream proliferative genes in oral squamous carcinoma cells. However, there is no in vivo report investigating the role of nitric oxide synthase in the acetylation of the histones H3K9 and H3K27 and its association with oral cancer development. The purpose of this study was to analyze H3K9ac and H3K27ac expression at different stages of oral carcinogenesis induced by 4-nitroquinoline-N-oxide (4NQO) in Nos2+/+ (wildtype) and Nos2-/- (knockout) mice. C57BL/6J and B6.129P2-Nos2tm1Lau/J mice were treated with 4NQO in the drinking water at 50µg/mL for 16 weeks and observed for 8 weeks (Protocol was approved by the institutional Ethics Commission on Animal Use (100/18)). Tongues were submitted to histopathological analysis and immunohistochemistry for H3K9ac and H3K27ac expression. The antigen-antibody reaction was analyzed with Quickscore (QS). Both histone acetylation marks were expressed in the normal epithelium. QS values were higher in moderate dysplasia of Nos2-/- mice (p=0.025) when



compared to Nos2+/+, and mild dysplasia had lower values for H3K9ac when compared to moderate and severe dysplasia in Nos2-/- group (p=0.015). H3K27ac significantly increased from normal mucosa to mild dysplasia in Nos2+/+ mice (p=0.007). Additionally, Nos2+/+ mice had a higher number of H3K27ac-positive mild dysplasias when compared to Nos2-/- (p=0.023). We concluded that the pattern of histone acetylation changes in murine oral carcinogenesis, mainly when the epithelial lining of the tongue becomes dysplastic, and that such epigenetic modifications might be iNOS-mediated.

**Keywords:** precancerous conditions, carcinogenesis, epigenetic process, histones, immunohistochemistry.



Title	Relationship between systemic inflammatory markers and depth of invasion in oral squamous cell carcinoma in Nos2- knockout mice following 4NQO treatment
Authors	Marcondes Pedro Souza Novais¹ Wender Rodrigues Nazário¹ Débora de Oliveira Santos¹ Anaíra Ribeiro Guedes Fonseca Costa¹ Pedro Victor Silva Duarte² Tássio Edno Atanásio Pitorro³ Lúbia Cristina Fonseca¹ Sérgio Vitorino Cardoso¹ Adriano Mota Loyola¹ Paulo Rogério de Faria¹
Affiliations	<ul> <li>1 - Department of Oral and Maxillofacial Pathology, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>2 - Department of Diagnosis and Surgery, Júlio de Mesquita Filho São Paulo State University, Araçatuba, Brazil</li> <li>3 - Department of Periodontology, Federal University of Uberlândia, Uberlândia, Brazil</li> </ul>
Session	Panel Session I

#### Abstract and Keywords

Peripheral blood analysis is a non-invasive and low-cost technique of prognostic value for several diseases, including oral cancer. Considering the role of inducible nitric oxide synthase (iNOS) in tumor-associated inflammation, this study purposed to evaluate the influence of this enzyme on peripheral blood parameters and systemic inflammatory biomarkers during murine oral carcinogenesis. A 50ug/mL solution of 4-nitroquinoleine-N-oxide (4NQO) was provided to 15 C57BL/6J (Nos2+/+) and 16 B6.129P2-Nos2tm1Lau/J (Nos2-/-) for 16 weeks. Animals were followed for 8 weeks after treatment (The protocol was approved by the institutional Ethics Commission on Animal Use (100/18)). Blood samples and tongues were collected for hematological and histopathological analyses. Red blood cells, white blood cells, and platelet cell parameters were analyzed. The neutrophil-to-lymphocyte (NLR), platelet-to-lymphocyte ratio (PLR), and the immune-inflammation index (SII) were also calculated. The depth of invasion (DOI) of all carcinomas was measured. Nine out 13 (69.2%) and 13 out of 16 (81.2%) Nos2+/+ and Nos2-/- mice developed OSCC (oral squamous cell carcinoma), respectively, but no difference was achieved. Differences were found in several blood parameters. The DOI in Nos2-/- was lower than in Nos2+/+ (p=0.009), and strong correlations were found between DOI and neutrophil count (p=-0.68, p=0.017), lymphocyte count (p=0.72, p=0.011), NLR (p=-0.65, p=0.017)p=0.025), PLR ( $\rho$ =-0.73, p=0.013), and SII ( $\rho$ =-0.67, p=0.037) in Nos2-/mice. iNOS seems to have an important role in OSCC invasion and progression, which might be associated to alterations in immune-inflammatory cell dynamics evidenced by peripheral blood and systemic inflammatory biomarkers.

**Keywords:** carcinogenesis, nitric oxide, oral cancer, tumor biomarkers, hematological



Title	Analysis of tumor cfDNA in normal prostate cells in 3D culture
Authors	Lorrayne Cristina Moura Garcia <sup>1</sup> ; Aline Gomes de Souza <sup>2</sup> ; Vivian Alonso Goulart <sup>1</sup>
Affiliations	<ol> <li>Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart Filho, Institute of Biotechnology (IBTEC), Federal University of Uberlândia, Uberlândia, Brazil.</li> <li>Translational Oncology Laboratory (FMRP-USP), Institute of Medicine, University of São Paulo, São Paulo, Brazil.</li> </ol>
Session	Cancer Signaling and Therapeutics

#### Abstract and Keywords

Prostate cancer (CaP) is one of the main malignancies that affect men after the age of 50. Because of this, researchers have sought to understand the different mechanisms of tumor cells so that it is possible to identify and treat PCa early. One of the targets of investigation is circulating cell free DNA (cfDNA), which refers to circulating DNA derived from apoptotic cells and cellular activity and is generally found in the bloodstream and can be captured through liquid biopsy. Current work shows that patients affected by cancer have a higher level of cell free DNA in biological fluids compared to healthy individuals, in this way, cell free DNA coming from tumor cells is a promising molecule for tumor control. Therefore, this work aimed to quantify cell free DNA from prostate cell lines in 3D culture (PC-3 and PNT-2), as well as evaluate cell morphology, migration and viability, after normal prostate cell line (PNT -2) being treated with cell free DNA isolated from the tumor line (PC-3) grown in a three-dimensional (3D) system. We used wound healing assay for migration analysis and in vitro Neutral Red Based (Sigma) for viability analysis. Statistical differences were calculated using Kruskal-Wallis test and t-Student. All statistical analysis was analyzed using Graph-Pad Prism Software. p-values < 0.05 were considered statistically significant. The data showed that cell free DNA released by the PC-3 lineage is capable of altering the morphology, migratory capacity of normal prostate cells and their viability, suggesting an important role of this molecule in the tumor process.

**Keywords:** Prostate cancer; Cell free DNA; Liquid biopsy.



Title	Preliminary assessment of redox state in plasma of diverse cancer patients
Authors	Ana Luiza Silva Borges¹ Vinícius Prado Bittar¹ Tarcisio Paiva Mendonça¹ Renata Roland Teixeira¹ Allisson Benatti Justino¹ Lara Peixoto² Foued Salmen Espíndola¹ Clarissa Aires de Oliveira² Eugenio Luigi Iorio²
Affiliations	<ul> <li>1 - Laboratório de Bioquímica e Biologia Molecular, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>2 - Clínica Conceito Saúde, Uberlândia, Brazil</li> </ul>
Session	22 Sinalização e Terapêutica do Câncer

#### Abstract and Keywords

Oxidative stress (OE) results from an imbalance between the production of reactive oxygen species and the body's antioxidant capacity to neutralize them. Cancer is closely linked to oxidative stress and its markers play crucial roles in understanding cancer development and progression. The aim of this study is to quantify OE biomarkers in the plasma of patients with various types of neoplasms. Volunteers blood samples (n=19, 11 women and 9 man) with different neoplasms (31.5%= breast cancer, 15%= prostate cancer and 47%= others) and healthy donors (n=16, 9 women and 7 men) were collected (CEP: 5,671,038) in heparin vacutainer tubes and centrifuged at 2000 xg for 15 minutes at 4°C to obtain plasma, which were stored in a ultrafreezer at -80°C. The total antioxidant capacity was measured using FRAP assay at 593nm. The glutathione (GSH) levels were obtained using a fluorimetric assay at ex=350 nm and em=420 nm. The GSH redutase, GSH peroxidase and superoxide dismutase (SOD) activity were accessed by kinetic reaction assays. To evaluate advanced oxidation products in proteins, chloramine T standard curve was used. The results were expressed as mean  $\pm$  standard deviation. Normality and lognormality tests were evaluated and analysis of variance between groups were performed by t test and values of p < 0.05 were considered significant. FRAP indicated no statistical difference between groups, showing that the total antioxidant response remains unaltered. Sulfhydryl content and AOPP indicate that proteins are not damaged in cancer group. The GSH levels are lower in cancer group when compared to individuals with no cancer. It explains the higher GR and GPx activity in cancer group, showing an overcharge in the glutathione system. SOD activity was not altered in cancer patients. This preliminary investigation underscores the enhanced antioxidant response within the glutathione system in cancer patients when compared to the non-cancer control group.

**Keywords:** cancer, oxidative stress, D-ROM, glutathione



Title	Investigation of the cytotoxic activity of Jatropha multifida extract in murine melanoma cells
Authors	Eduardo Batista de Sousa¹ Clara Maria Faria Silva¹ Ricardo Campos Lino¹ Mariana Cristina Teixeira de Moura¹ Robson José de Oliveira Júnior¹
Affiliations	1 – CITOGEN, University of Uberlandia, Uberlandia, Brazil.
Session	Cancer Signaling and Therapeutics

#### Abstract and Keywords

Biodiversity is one of the main characteristics of the biosphere, encompassing plants, animals, and microorganisms. Brazil is the region of the planet with the greatest biodiversity, boasting a vast array of species in the Amazon Rainforest, as well as unique biomes such as the Caatinga. Given this scenario, the Brazilian territory becomes an ally for research and bioprospecting of new molecules with economic and therapeutic interest. In line with the search for new compounds, popular knowledge leads us to the use of the Merthiolate plant (Jatropha multifida), which is used to treat lesions by applying the latex extracted from its leaves locally. Thus, the following work conducted cytotoxicity assays in murine melanoma cells (B16F10) with the purpose of clarifying at what concentration the aqueous portion of the extract becomes harmful to these cells. Therefore, from plants located in the Cytogenetics Laboratory (Citogen) of the Federal University of Uberlândia - UFU, an aqueous stock solution of latex at 1% was prepared. Subsequently, using the serial dilution technique, the solution was fractionated into concentrations of 1%; 0.5%; 0.25%; 0.13% and 0.063%, and applied in the colorimetric assay of resazurin reduction. As a result of the assay, the qualitative results were processed in a spectrophotometer at 570 and 600 nm. From the obtained values, the IC50 was determined to establish the level of cytotoxicity in the cells. In summary, with the acquired data and the calculations performed, it is possible to infer that the latex presents cytotoxicity at around 0.67%. Affirming the need for further study of the material's properties and its activity since cytotoxicity and antitumor activity are described in plants of the Jatropha genus in the literature. Bringing the possibility of new formulations of chemotherapeutics.

**Keywords:** latex, cytotoxicity, bioprospecting, Merthiolate.



Title	Assessing the comparative effects of two PLA2-Lys 49 in prostate cancer cells
Authors	Pedro Henrique Amiante Bugatti; Lucas Ian Veloso Correia; Fernanda Van Petten de Vasconcelos; Mariana Alves Pereira Zóia; Lara Vecchi; Luiz Ricardo Goulart; Veridiana de Melo Rodrigues.
Affiliations	Biotechnology Institute, Federal University of Uberlândia, Uberlândia, MG, Brazil
Session	Sinalização e Terapêutica do Câncer

#### Abstract and Kevwords

Phospholipase A2 enzymes, gained attention for their diverse biological activities, who are potential candidates for cancer research. In this study, we evaluated these activities of two PLA2 Lys-49 enzymes: BnSP-6 from Bothrops pauloensis and BthTx-I from Bothrops jararacussu venom. Our investigations were conducted using metastatic prostate cancer cells (PC3 and LNCAP) and a nontumorigenic cell line (RWPE). The enzymes were purified according to established protocols. To assess cell viability, we employed the MTT assay. Cells (2x104) were plated and treated with varying concentrations of the toxins for 24 hours. Following, 20 µL (5mg/mL) of MTT was added to each well for 4 hours. Autophagy induced by PLA2s was examined using the MDC probe in cells treated for 24 hours and visualized under a fluorescence microscope. For antiproliferative activity, cells were treated for 72 hours with the toxins and CFSE. Cell fluorescence was monitored every 24 hours using a spectrophotofluorometer. Calcium flow analysis in cells treated with PLA2 was investigated using the Fluor-4/AM probe, with data acquired via spectrophotofluorometer readings. Cell cycle analysis was performed through flow cytometry using propidium iodide in cells treated for 24 hours. Production of reactive oxygen species was assessed with the DCFH probe using flow cytometry. Both toxins exhibited cytotoxicity in tumor cell lines while displaying low toxicity in the non-tumorigenic cell line. They induced autophagy predominantly in tumor cells, with BnSP-6 exhibiting higher cytotoxicity. Both toxins inhibited proliferation after 72 hours and concentration-dependent cell cycle arrest. PLA2 Lys-49 impacted calcium flow primarily in the PC3 cell line, evoked by BnSP-6. ROS production was induced by BnSP-6 in both tumorigenic cell lines. Findings suggest that PLA2 Lys-49 induces damage in cancer cells, rendering them a promising model for the development of innovative therapeutic agents against prostate cancer.

**Keywords:** Phospholipase A2, Prostatic Cancer, BnSP-6, BthTx-I



Title	Discovery of new antineoplasics agents: exploring the in silico and in vitro interaction of the Rubpy metallocomplex with DNA
Authors	Letícia Alves de Oliveira <sup>1</sup> ; Marcella Alves Nogueira <sup>2</sup> ; Ricardo Campos Lino <sup>2</sup> ; Tayana Mazin Tsubone <sup>3</sup> ; Antônio Otávio de Toledo Patrocínio <sup>3</sup> ; Robson José de Oliveira Júnior <sup>2</sup>
Affiliations	<sup>1</sup> Faculty of Medicine of the Federal University of Uberlândia, Uberlândia Brazil. <sup>2</sup> Biotechnology Institute of Federal University of Uberlândia, Uberlândia, Brazil. <sup>3</sup> Institute of Chemistry of the Federal University of Uberlândia, Uberlândia, Brazil.
Session	Sinalização e Terapêutica do Câncer

#### Abstract and Keywords

Many drugs used in cancer treatment have low selectivity for neoplastic cells, which can develop cause chemoresistance, turning the search for new effective and less harmful antitumor drugs essential. Recent studies have shown that compounds based on the transition metal ruthenium have promising chemotherapeutic activity, with lower toxicity and greater specificity. Thus, this work aimed to evaluate the interaction between the photoactivated metallocomplex Rubpy, whose metallic center is a ruthenium atom, with DNA by molecular docking, and the in vitro cytogenotoxic potential. Rubpy molecule was treated, using the ChemDraw and ChemBio3D software, to obtain greater stability. The Autogrid4 and Autodock4 tools were used to calculate the docking between the organometallic and a DNA molecule obtained from the Protein Data Bank (PDB). Furthermore, two murine cell lines were cultivated: (i) B16F10 melanoma and (ii) NIH/3T3 fibroblasts, to carry out cytotoxicity tests using resazurin colorimetric assay with and without photoactivation. A plasmid DNA (pDNA) interaction and cleavage assay was performed. The results obtained by molecular docking demonstrated that the metallocomplex interacts noncovalently with the major groove of DNA, in the least energetic conformation (binding energy = -6.78 Kcal/mol), through a hydrogen bond with nitrogen 1 of DNA adenine 8 and other intermolecular interactions. However, Rubpy did not show cytotoxicity to the tested cell lines at the used concentrations, even when photoactivated. In the pDNA interaction assay, the compound was unable to cleave pDNA at dosages of 50 and 100  $\mu$ M, indicating that there was no genotoxicity. Therefore, although Rubpy interacted with DNA in in silico analyses, it did not exhibit in vitro cytogenotoxic activity. Despite this, its affinity for DNA serves as a guide for the design of new metallodrugs with similar ligands, capable of acting as more effective and tolerable antineoplastics.

**Keywords:** Chemotherapy; Metallocomplex; Ruthenium.



Title	From phages to peptides: Developing novel strategies against triple- negative breast cancer with BnSP-6 mimetics
Authors	Lucas Ian Veloso Correia; Fernanda Van Petten Azevedo; Emília Rezende Vaz; Fabiana de Almdeida Araújo Santos; Mariana Alves Pereira Zóia; Sarah Natalie Cirilo Gimenes; Luciana Machado Bastos; Luiz Ricardo Goulart; Veridiana de Melo Rodrigues.
Affiliations	Biotechnology Institute, Federal University of Uberlândia, Uberlândia, MG,
Session	Sinalização e Terapêutica do Câncer

### Abstract and Keywords

Breast cancer (BC), renowned for its heterogeneity and diverse subtypes, presents a significant clinical challenge. Among these, the triple-negative BC stands out as notably resistant to conventional treatments. Within the realm of venom biomolecules, phospholipases A2 (PLA2) emerge as alternative for therapeutic exploration. However, the druggability of phospholipases A₂ poses a significant hurdle. Miniaturization of these molecules has emerged as a strategic approach to enhance their pharmacological potential. In this context, we developed peptides mimicking BnSP-6, PLA<sub>2</sub> isolated from *B. pauloensis*, and explored their antitumor potential through in vitro and in vivo approaches. To select mimetic peptides for BnSP-6, a library of random phage-fused peptides underwent three cycles of selection, incorporating the IgY antibody for competition with BnSP-6. The Phage-ELISA technique identified the most promising phages, whose DNA was sequenced. The top phages were tested for cytotoxicity using the MTT method in BC cell culture, with a wild-type phage as a control. 2 best sequences were synthesized and evaluated in vitro for cytotoxicity, breast cancer cell proliferation inhibition, and apoptosis/necrosis induction. In vivo approach in D. melanogaster were conducted by toxicity tests over 21 days, measuring budding, hatching, and peptide exposure through food. Furthermore, the modulating potential of peptides on carcinogenicity was evaluated using the Epithelial Tumor Test assay. The phage strategy yielded 13 reactive phages, with 9 sequenced and 2 peptides synthesized. Synthetic peptides 8B and 1C exhibited cytotoxic effects on BC cells, inhibiting cell proliferation and inducing apoptosis/necrosis pathways. Moreover, the peptides modulated the carcinogenicity of in a D. melanogaster model without inducing toxic effects. This pioneering work identifies and evaluates potential peptides mimicking PLA<sub>2</sub> structures, showcasing their antitumor potential against breast cancer.

**Keywords:** Triple Negative Breast Cancer; Venom; Phospholipase  $A_2$ ; Phage Display.



Title	4-Nerolidylcatechol's promise in prostate cancer therapy
	Sabrina Cristina Guedes <sup>1</sup> Lucas Ian Veloso Correia <sup>1</sup> Raoni Pais Siqueira <sup>1</sup> Ademar Alves da Silva <sup>2</sup>
Authors	Lígia Nunes de Morais Ribeiro <sup>1</sup> Thaise Gonçalves de Araújo <sup>1</sup>
	<sup>1</sup> Biotechnology Institute, Federal University of Uberlândia, Uberlândia, MG,
Affiliations	<sup>2</sup> Universidade Federal de Juiz de Fora (UFJF)- Juiz de Fora, Brasil
Session	Sinalização e Terapêutica do Câncer

#### Abstract and Keywords

Metastatic prostate cancer represents a formidable challenge in the realm of oncology, necessitating innovative approaches to treatment. Despite advances in therapeutic strategies, metastatic prostate cancer often displays resistance to conventional treatments, demanding a constant exploration of novel and effective therapeutic avenues. In this pursuit, bioprospecting for plant biomolecules emerges as a promising frontier. In this context, we study the effects of 4-nerolidylcatechol (4NC) against prostate cancer cells (PC3) in vitro and the inhibition of tumor growth and inflammation in chicken embryo model, an alternative in vivo model. The IC<sub>50</sub> of cytotoxicity of PC3 was measured by resazurin assay. Following, the membrane coryoalontoid chicken assay was assessed using eggs from the W-36 strain of Gallus gallus. For the tumor inhibition and tumor regression tests, eggs with 7 days of development were inoculated with PC3 and the extracellular matrix where the cells were pretreated or treated after inoculation with different concentrations of 4NC. The eggs were incubated and, on the 14th day, they were opened to assess the size of the tumor mass. The tumors were then collected, weighed and stored in formalin for histopathological evaluations. 4NC shows a cytotoxicity agains PC3 cells. Through the in vivo model, 4NC demonstrated partial tumor growth inhibition and regression. Histopathological evaluation revealed mild inflammatory process and the presence of necrotic areas. The findings presented in this study are extremely important, as they highlight the therapeutic potential of the 4NC compound.

**Keywords:** Prostate cancer; plant metabolite; 4-nerolidylcatechol; PC3.



Title	Escape responses and looming visual habituation in adult zebrafish
Authors	Joaquim Carlos Rossini
Affiliations	1 – Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	session 23 – Visão e oftalmologia

## and Keywords

Escaping a predator is an essential behavior for survival and all animals with a visual system were selected by evolution to process certain characteristics of motion that can prioritize the motor response to avoid a potential threat.

Specifically, the zebrafish (Danio rerio) presents a robust escape response elicited by an object fast approaching (looming). Here, we investigated the habituated escape behaviors with repeated looming and receding stimulation. We asked whether the escaping response of the looming stimulus is less susceptible to habituation than the escaping response elicited by the receding stimulus. To that end, we investigated the effect of repeated exposure to looming or receding stimuli (15 repetitions). The speed of escapes was recorded and analyzed using Toxtrac software for behavior analysis. Thirty-four zebrafish (Danio rerio) of the wild strain (50-50% male-female) were used in the experiments. Each experimental fish was tested once and individually in an experimental arena (28 cm  $\times$  3 cm  $\times$  21.5 cm, width  $\times$  depth  $\times$  height). To mimic the forward or backward motion of the rapid looming or receding of an object, a dot was shown above the experimental arena using a computer monitor. The dot increased or decreased its size with a linear acceleration from 0.5 cm to 13 cm in diameter (50 cm/s). The dot presentation periods were interspersed with 60 s interstimulus intervals during which the dot was not shown. The results showed a main effect of stimulus type (looming or receding) with high speeds on the looming condition (106 mm/s) compared to the receding condition (76 mm/s), t (33) = 2.75, p < .01, d = 0.95. Both, looming and receding stimuli, elicited habituation along the 15-repetition, but the responses were faster always in looming condition. The results suggest that zebrafish show habituation for looming and receding stimuli, but the looming stimulus drives fast escapes and could characterize a special neural circuit to urgent responses.

**Keywords:** visual looming; looming-evoked escape; habituation; zebrafish model



Title	Do biostimulators calcium hydroxyapatite and poly-D-lactic acid combined or not with ozone change the cell viability, proliferation and migration of HFF-1 fibroblasts?
Authors	Lucas Correia Peres <sup>1</sup> ; Ludmilla Sousa Quirino <sup>1</sup> ; Vivian Alonso-Goulart <sup>1</sup> ; Letícia de Souza Castro Filice <sup>2</sup>
Affiliations	<ul> <li>1 - Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart Filho, Institute of Biotechnology (IBTEC), Federal University of Uberlândia, Uberlândia, Brazil</li> <li>2 - Faculty of Medicine, Federal University of Uberlândia, Uberlândia, Brazil</li> </ul>
Session	Regenerative Medicine and Development Biology

#### Abstract and Keywords

The search for new procedures to reduce signs of aging in which the patient does not need to undergo intense pain and long periods of rest, combined with longer and more satisfactory results, has been increasing. The collagen biostimulators calcium hydroxyapatite (CaHA) and polylactic acid (PDLA) stand out on the market and ozone appears as a possible enhancer of the effects, however there are limited studies defining safe and effective working concentrations on these compounds in vitro. Therefore, the objective of the study was to investigate whether the viability and migration of fibroblasts (HFF-1) are affected when cultivated with the compounds at different concentrations. The methodology used for cell viability was the reduction of resazurin, which is a blue dye that upon entering viable cells is converted to a high fluorescent pink compound known as resofurin. For the analysis of cell proliferation and migration, Scratch Assay was conducted and ImageJ analyses were performed on images captured at 0, 8, 24, and 48 hours after the start of the treatment. The results of biostimulator applied alone by the resazurin viability assay indicate that three concentrations (300, 100, and 30 µg/mL) tested for CaHA showed cytotoxicity, whereas the same concentrations of PDLA or ozone (15, 10, and 5 µg/mL) remained viable after 24 hours of treatment. Although combining biostimulators with ozone (CaHA and ozone or PDLA and ozone) led to an apparent improvement in viability by the resazurin test when compared to the results of the biostimulators used alone. Concentrations of 100 µg/mL of PDLA and 10 µg/mL of ozone either alone or in combination with ozone, did not negatively interfere with cell proliferation and migration. In conclusion, these findings may demonstrate that the combination of the biostimulators with ozone may increase cell viability, while cell proliferation and migration remain unaffected by the administration of PDLA and ozone at the concentrations tested.

Keywords: Biostimulation; Fibroblast; Cell Viability; Cell Migration; Ozone.



Title	Parasitology promotion via Instagram: The Experience of @parasito.logico
Authors	Vanessa da Silva Ribeiro Juliana Silva Miranda
Affiliations	Parasitology Department, Biomedical Sciences Institute – Federal University of Uberlândia
Session	25 - Educação, História e Filosofia da Ciência, Comunicação Científica

#### Abstract and Keywords

Introduction: Currently, access to information has become easy and daily with the use of electronic devices. Applications and plataforms like Instagram have become part of the new routine of information. In this sense, scientific dissemination is an important way to disseminate topics in public health such as Parasitology. Objectives: To expand the reach Parasitology topics, through the Instagram page @parasito.logico. Methods: The page was created in September 2019, and since then an average of three weekly posts have been made, in different formats such as: stories, reels, reposts of parasite videos, detailed article topics in a clear and uncomplicated manner, memes, photos, news about disease outbreaks, a series with researchers addressing their lines of activity, commemorative dates, a series with parasites in artworks called parasitic invasion, routine in the laboratory, glossary, study/teaching tool tips, among others. In order to engage the audience and provide knowledge about Parasitology. Results: In the analyzed period (09/04/2019 to 01/31/2024) we reached 3,522 followers and 16,7 thousand people who are not followers but follow the page. Among the followers, the majority are female (71.4%), with the predominant age group being 25 to 34 years old (44.1%). Among the locations of followers, the majority are from Brazil (71.7%). There were 677 publications, of which 233 were videos (video reels and reposts), 38 posts with articles, 73 news, 11 videos from the series with researchers, 73 glossary terms, 25 study tool tips, among others. In addition, in the bio, there are links to free materials such as e-books published by the founders of the page. Conclusion: Through social networks it was possible for us to carry out different posts that have reached diverse audiences. Actions in this sense promote the scientific dissemination of Parasitology, achieving the purpose of popularizing useful content in public health, as well as providing an environment for the exchange of knowledge and experiences.

**Keywords**: Parasitology, Education, Science popularization, public health.



Title	The drawing as a learning tool: Parasitology in the daily lives of elementary school students in public schools
Authors	Juliana Silva Miranda Vanessa da Silva Ribeiro Alessandro Sousa Correa Daliane Faria Grama Daniela Beraldo Barbosa
Affiliations	Instituto de Ciências Biomédicas – Universidade Federal de Uberlândia
Session	25 - Educação, História e Filosofia da Ciência, Comunicação Científica

#### Abstract and Keywords

**Introduction:** Drawing is a playful tool that aids in a child's socialization, allowing them to attribute meaning to their reality. In daily life, alongside experiences with adults and peers, children recognize drawing as a language to be used as a form of communication, employing creativity to assist in the symbolic structuring of the content that surrounds them. **Objective:** To assess, through drawings, the perception of elementary school students (6th to 9th grade) on the theme: Parasites transmitted via food, water, or soil and its relation to their daily lives. Methods: Different teachers were contacted and asked to discuss drawing activity during class, prompting students to create illustrations involving parasites transmitted via food, water, or soil and their daily lives. This activity was proposed to elementary school students (6th to 9th grade), regardless of whether they had previous exposure to parasite-related content. After completion, the drawings were handed over to the evaluation team for analysis. Results: In total, 158 students engaged in the activity and produced illustrations: 6th grade (n=7); 7th grade (n=83), 8th grade (n=23), and 9th grade (n=45). Analysis of these illustrations revealed that students from 6th grade (85.7%), 7th grade (24.1%), 8th grade (65.5%), and 9th grade (77.7%) were able to creatively depict various content related to the theme in their drawings, such as parasite shapes, cycles, soil, water, and food contamination, which were consistent with important concepts in Parasitology. Conclusion: Through analysis of the drawings, it was possible to observe that students utilized their imagination in constructing representations within the context of Parasitology, aligned with content related to various parasites causing diseases in humans. This reinforces the use of drawing as a tool to assess students' perceptions of diverse topics covered in the classroom, enabling the development of learning strategies.

**Keywords:** parasitology; education; school students; drawing.



Title	Essential Scent
Authors	Alisson Júnio Parreira Peixoto Danusa Radi Gomes Santiago Nathalia Vieira Kamimura
Affiliations	Sistema Gabarito
Session	Education

#### Abstract and Keywords

The "Essential Scent" project applied principles of educational neuroscience by integrating knowledge from various areas in middle and high school education, promoting a transdisciplinary approach and environmental awareness. The production of herbal soaps involves activities that stimulate different areas of the brain, such as mathematics, natural sciences, humanities, languages, and technologies, favouring a more meaningful learning experience. The methodology included the development of a website and an application, encouraging the use of varied cognitive skills. The collaboration between high school students in producing the glycerin base and those from middle school in making the soaps is noteworthy, reflecting integration across different levels of education and promoting a broader understanding of content. The results highlighted student engagement and the importance of interaction between school segments in shaping a sustainable society, fostering ethical, social, and environmental reflections. Furthermore, it became evident that interdisciplinary and transdisciplinary approaches in education are aligned with the principles of educational neuroscience. Both approaches promote teaching that considers the functioning of the brain and students' cognitive processes, aiming to achieve more effective, meaningful, and enduring learning.

**Keywords:** transdisciplinary education, environmental awareness, phytotherapeutic soaps, interdisciplinary collaboration, student engagement, educational neuroscience.



Title	Philosophical Magazine
Authors	Alisson Júnio Parreira Peixoto Danusa Radi Gomes Santiago Maria Aparecida Martins Cordeiro Nathalia Vieira Kamimura
Affiliations	Sistema Gabarito
Session	Education

### Abstract and Keywords

The "Philosophical Magazine" project applied principles of neurolearning by integrating diverse knowledge in high school, promoting an interdisciplinary approach and philosophical reflection. The production of the magazine involved activities that stimulate different areas of the brain, such as research, writing, design, and social interaction, favouring a more significant learning. The methodology included the creation of philosophical content by students, as well as optional elements like advertising, cooking, and interdisciplinary dialogues, encouraging the use of various cognitive skills. The collaboration among students in the magazine's development is highlighted, reflecting an integration of different skills and promoting a broader understanding of philosophical content. The results evidenced the students' engagement and the importance of philosophical reflection in shaping a critical and responsible society, stimulating ethical, social, and cultural reflections. Furthermore, it became evident that interdisciplinary education aligns with the principles of neurolearning, promoting teaching that considers students' cognitive processes, aiming to achieve more effective and lasting learning.

**Keywords:** "Philosophical Magazine" Project, Neurolearning, High School, Interdisciplinary Approach, Philosophical Reflection, Cognitive Stimulation, Varied Activities, Student Collaboration, Student Engagement, Educational Interdisciplinarity.



Title	Use of three-dimensional (3D) tardigrade models for scientific dissemination
Authors	Alexsandre Domingues Junior Ricardo Corbetta
Affiliations	University of Vale do Itajaí, Itajaí, Brazil
Session	25 - Education, History and Philosophy of Science, Scientific Communication

### Abstract and Keywords

Use of 3D teaching models are very important in the absence of preserved biological specimens, especially due to the difficulty of legal access to content of this type outside Universities. They facilitate teaching in schools and museums, also helping people with visual impairments learn, for example. However, they become even more relevant when it comes to meiofauna, as the specimens are microscopic in size, and yet they are too complex for just one image. Considering pop culture's interest in tardigrades, which are mentioned in the re-recording of the series Cosmos, currently narrated by Neil deGrasse, the scarcity of teaching material with tardigrade species is notable. To this end, a model measuring approximately 11.5cm in epoxy mass was produced to represent a specimen of Echiniscus granulatus. The piece was exhibited at the UNIVALI Environmental Sciences Laboratory during the 2023 OPA (Professional Option by Area), along with images and data on the genus Echiniscus, as well as slides with fixed specimens. The model facilitated the pointing out of structures and better assimilation by visiting students.

Unlike an image, a three-dimensional model can be rotated, in addition to becoming more familiar to those who see it, considering its similarity to Arthropods. To facilitate its reproduction, a two-part silicone mold was made, initially tested with stone plaster. One difficulty in such models is representing more delicate structures. To overcome this problem, disposable Pasteur pipettes were used, as they have a malleable and resistant plastic, in addition to allowing them to be heated for stretching filaments, for cirrus and spines. As for the claws, cut acetate showed good results. Although they make the replication process difficult, as these structures need to be made separately, they allow for greater fidelity, without compromising the durability of the piece.

Keywords: 3D Modeling, Exhibition, Meiofauna, Tardigrada



Title	Teaching project "Human Physiology: Let's learn how to learn?"
Authors	Fabiane Fernandes da Silva Simone Acrani
Affiliations	Department of Biochemistry, Pharmacology and Physiology, Federal University of Triangulo Mineiro, Uberaba, Brazil
Session	Oral Presentation

#### Abstract and Keywords

The learning process continually seeks to develop individuals' knowledge, skills and competencies. Active methodologies (AMs) have gained prominence as an innovative way of approaching teaching, by connecting the student's reality with the learning strategy and, by placing the student at the center of the educational process, shifting the focus from the teacher (teaching) to the student (learning), making them the protagonist of their own education. Gamification, one of the MAs strategies, encompasses these principles by focusing on student motivation and interest. Explore the potential of MAs, in the context of Human Physiology, in order to simplify the contents and transform them into topics of interest and appreciation for their study, improving understanding and consolidating learning. This project was carried out with students from the 3rd period of the Occupational Therapy course at the Federal University of Triângulo Mineiro, through the application of didactic games that addressed the different contents of the Physiology discipline, such as: Cell Bingo, Cell Signaling Board, ANS Word Search and Crossword, Somatosensory System Quiz, Naval Battle of the Heart and Who Am I of the Respiratory System. A form was distributed to assess students' impressions of the dynamics. Analysis of the responses revealed an average engagement of 62% of students in the activities, with unanimous approval regarding their contribution to learning. Around 32% had no prior experience with AMs, but expressed confidence in applying these dynamics to other content. The results showed the importance of adapting educational strategies to the needs of students, promoting engaging and meaningful learning and enabling the consolidation of technical knowledge and the making of connections between the different subjects covered.

**Keywords:** active methodologies, gamification, teaching-learning, human physiology, education.



Title	Health education: Information to prevent accidents with scorpions	
Authors	Letícia Duarte Silva Rachel campos Ornelas Filipe Ferreira Santos Ana Carolina Vieira Maísa Silva Ione Maria de Matos	
Affiliations	Universidade Federal de Juiz de Fora – <i>Campus</i> Governador Valadares, Governador Valadares, Minas Gerais, Brasil.	
Session	25- Educação, História e Filosofia da Ciência, Comunicação Científica	

#### Abstract and Keywords

We aimed to describe an experience of developing and applying a Didactic Sequence (DS), including an educational game to study scorpion sting prevention. We developed a didactic game entitled "Journey against Scorpionism". The SD took place in three stages: in the first stage we taught a dialogued expository class to address the topic of scorpionism. The second stage, we showed specimens of scorpions to the students. The third stage was the application of the didactic game. The project was carried out in 5 public schools in the city of Governador Valadares, Minas Gerais, Brazil, with primary and secondary school students. Around 150 students participated in the activities, with an average of 30 students per class. We observed that the students were interested in the topic, everyone interacted, telling stories they knew about scorpion stings and/or asking for clarification on the topic. We realized that it was a moment of great interaction. When we showed the scorpions, there was a lot of interest in learning about the animal's biology. The students played the didactic game, and we realized that it was possible to consolidate the knowledge acquired. We believe that these educational actions have the potential to contribute to the prevention of scorpionism and consequently reduce mortality caused by such accidents.

**Keywords:** scorpionism. didatic sequence. didatic game.



Title	Building and manipulating proteins: a different teaching practice
Authors	Mariana Alves Messias Souza Bomfim
	Francislene Glória de Freitas Reis
Affiliations	Federal University of Triângulo Mineiro, Uberaba, Brazil
Session	poster

### and Keywords

Understanding Biochemistry becomes a challenge when considering that students must deal with the abstract and submicroscopic world of molecules, ions, and chemical reactions that occur in living organisms during the classes of this discipline that make up the core of health-related courses. Therefore, this work aims to improve students' understanding of a group of biomolecules, proteins, by making them more tangible and enabling their construction and manipulation without the need for modern technologies. To achieve this, a practical class script was developed and implemented, including a theoretical framework for reading, an atlas with images from the Protein Data Bank (PDB), a list of craft materials to be used to build insulin, hemoglobin, and myoglobin, the procedures to be performed for the construction of these molecules, and questions for online research about them: the number of amino acids present in the structure, the organizational level found in the protein in its native condition, and the classification of proteins regarding composition, function, and location. During the practical activity and after its completion, it was possible to gather feedback from students who expressed satisfaction in "seeing" the proteins, gaining a better understanding of their composition, organizational structures, native conformation, and denaturation.

**Keywords:** Biochemistry; Teaching; Proteins; Teaching resource; Education; Learning; Learning difficulties.



Title	Understanding cellular respiration among health sciences undergraduates
Authors	Mariana Alves Messias Souza Bomfim Francislene Glória de Freitas Reis
Affiliations	Universidade Federal do Triângulo Mineiro, Uberaba, Brasil
Session	poster

# and Keywords

Biochemistry is a mandatory discipline in health sciences courses, covering complex and challenging content for students due to the presentation of processes that occur in the cellular environment, being abstract and submicroscopic, unlike the Anatomy discipline. One of the major difficulties involves the metabolic process known as cellular respiration, which, due to its crucial role in ATP production, must be comprehended by healthcare professionals. This study aims to analyze the development of understanding among undergraduate students in health sciences courses regarding cellular respiration. In a Biochemistry course at a higher education institution offered to two health sciences courses (Course I: n=24; Course II: n=17), a questionnaire containing two questions about cellular respiration was administered at two moments (beginning and end of the course). The tool used was a questionnaire composed of two equally valued questions (value: one point). Exclusion criteria included not considering responses from students who had previously taken the course and those who participated in only one moment. The percentage development in the response of Course I students was 26.9±11.5%, and for Course II, it was 47.4±18.6%. It is believed that the percentage for Course I might be higher since the questions were answered evaluatively as part of the course grades, unlike Course II. In general, everyone understands the need for oxygen uptake and carbon dioxide elimination by the lungs and the presence of these gases in the bloodstream. However, understanding the role of oxygen within cells and where carbon dioxide is produced is a source of mystery and distress for students initially. It is crucial that the teaching methodology employed by the instructor helps them "see" what is actually happening in their cells.

**Keywords:** Cellular respiration, teaching, learning difficulties, Biochemistry.



Title	Role of macrophage migration inhibitory factor (MIF) in the development of the ventral prostate of mice
Authors	Júlia Eduarda Mesquita Matos Marina das Graças Carneiro e Silva Laura Eduarda Dinatto Sudário Luiz Felipe Fernandes Peixoto Daniele Lisboa Ribeiro
Affiliations	Department of Cell Biology, Histology and Embryology at the Institute of Biomedical Sciences of Federal University of Uberlândia, Brazil.
Session	Poster

#### Abstract and Kevwords

The macrophage migration inhibitory factor (MIF) is a multifunctional pleiotropic protein that can activate anti-apoptotic, pro-proliferative and pro-inflammatory mechanisms. Although it is expressed in the prostate, its importance in this gland is not fully understood. Thus, the contribution of MIF for the development of ventral prostate was evaluated in pre-pubertal and pubertal mice. In this study, histological sections of the prostate from C57BL/6 WT (wild type) and MIF-/-(knockout) mice, aged 30 and 60 days (CEUA-UFU 30/21), were stained for stereological analysis of the epithelium, lumen and stroma, and for the quantification of collagen. Immunohistochemical reactions were also performed to analyze smooth muscle, fibroblasts and cell proliferation. It was found that, although there was no difference in body and prostate weight between the groups, weight gain throughout development was much lower in the MIF group. MIF<sup>-/-</sup> animals at 30 days showed a 20% reduction in epithelium, an almost 50% reduction in smooth muscle stroma, a 34% reduction in collagen and a 30% reduction in smooth muscle cells. As regards cell proliferation, MIF-/- mice exhibited a reduction of 28% and 78% of proliferating cells in the epithelium of 30 and 60 days group, but in stroma, this reduction was found only in 60 days group. During the course of prostate development from 30 to 60 days, although the prostate gains volume, a reduction in the epithelium and stromal area is expected, but with an increase in the luminal area. In MIF group this reduction is already very evident at 30 days and therefore there is no big difference between the pre-pubertal and pubertal phases as seen in the control group. These results show an early development of the prostate in the absence of MIF, anticipating the changes that are expected for the transition of prepubertal to pubertal life. More importantly, the absence of MIF impacted the signaling pathways that control cell proliferation, reducing this activity that is so important for the postnatal development of the prostate. Thus, we conclude that MIF has an important role in stimulating proliferation and perhaps in the temporal control of changes that occur in the course of postnatal prostate development.

**Keywords:** Prostate, MIF, Knockout, Development, Morphology.



Title	Influence of heterogeneity in sperm chromatin compaction on rooster (Gallus gallus) spermatozoa head size
Authors	Luciana Beatriz Tiago Oliveira, Helen de Oliveira Duarte Ferreira, Bruno Augusto Nassif Travençolo, Marcelo Emílio Beletti
Affiliations	Federal University of Uberlândia, Uberlândia, Brazil
Session	Development Biology and Regenerative Medicine

### and Keywords

Assessing morphological changes in sperm is one of the main techniques used to identify fertility problems in roosters. However, roosters with normal sperm morphology often have fertility problems. This is probably because this type of assessment fails to identify some sperm defects, such as changes in the chromatin of the sperm. These alterations can decrease the sperm's fertilizing capacity and, when a sperm with this type of alteration manages to fertilize an oocyte, embryonic development is usually made impossible. There are several types of chromatin alteration, one of which is heterogeneous chromatin compaction in the different regions of the sperm head. This study aimed to determine whether heterogeneous sperm chromatin compaction influences the size of the sperm head in cockerels (Gallus gallus). We used 1500 images of rooster sperm obtained from smears stained with 0.025% toluidine blue, pH 4.0, under a light field microscope. The images were automatically analyzed by computer, using an algorithm developed in Matlab, and the heterogeneity of chromatin compaction, length, and area of each sperm head were evaluated. Pearson's correlation test was then carried out between these variables. Heterogeneity showed a low positive correlation (R = 0.16) with the variables related to head size. This shows that sperm with heterogeneous chromatin compaction does not always have an increased head size. Therefore, this is a possible explanation for why the evaluation of sperm head morphology often fails to identify fertility problems (ethics committee opinion 71/2022/CEUA/PROPP/REITO).

**Keywords:** heterogeneity, rooster, sperm chromatin.



Title	Changes in sperm chromatin identified by integrated optical density interfere with the head size of rooster spermatozoa ( <i>Gallus gallus</i> )
Authors	Helen de Oliveira Duarte Ferreira, Luciana Beatriz Tiago Oliveira, Bruno Augusto Nassif Travençolo, Marcelo Emílio Beletti
Affiliations	Federal University of Uberlândia, Uberlândia, Brazil
Session	Development Biology and Regenerative Medicine

#### Abstract and Keywords

In roosters, the shape of the sperm head is similar to a slightly spiral stick and may show morphological changes. Visual assessment of rooster sperm head morphology using light microscopy is subjective, but it is possible to use computer image analysis of semen smears stained with toluidine blue (TB) to objectively assess sperm head size and sperm chromatin. Integrated optical density (IOD) is a variable that can be assessed by image analysis and reflects the amount of TB bound to the sperm DNA. It is directly related to chromatin compaction, since the greater the chromatin compaction, the less access the TB has to the DNA. Decreased sperm chromatin compaction leads to DNA fragmentation, which will make embryonic development unfeasible if a sperm with this alteration fertilizes an oocyte. This study aimed to evaluate the influence of chromatin alterations identified by IOD on the head size of rooster sperm (Gallus gallus). We used 1500 images of rooster sperm obtained from smears stained with 0.025% toluidine blue, pH 4.0, under a light field microscope. The images were automatically analyzed by computer using an algorithm developed in Matlab. The IOD, length, and area of each head were evaluated. Pearson's correlation test was then carried out between these variables. IOD showed a positive and significant correlation with the variables, which means that alterations in the sperm chromatin of roosters increase the size of the sperm head. This can interfere with the sperm's fertilizing capacity and reduce the possibility of spermatozoa with alterations fertilizing (ethics committee sperm an oocyte opinion 71/2022/CEUA/PROPP/REITO). **Keywords:** integrated optical density, sperm chromatin, rooster.



Title	Staging and viability of <i>Gallus gallus</i> domesticus embryos treated with Hidantal <sup>®</sup> phenytoin
Authors	Roberta Vaz Assunção <sup>1</sup> , Vinícius Borges de Faria <sup>2</sup> , Augusto César da Fonseca Neto <sup>1</sup> , Francyelle Borges Rosa de Moura <sup>1</sup>
Affiliations	1 - Universidade Federal de Catalão, Catalão, Brasil 2 - INATA Produtos Biológicos, Uberlândia, Brasil
Session	Regenerative Medicine and Developmental Biology

#### Abstract and Keywords

Phenytoin, a medication used to treat epilepsy, when used by pregnant women, tends to promote malformations in the nervous system, lip and palate cleft, hypoplasia of the nails and distal phalanges and craniofacial abnormalities during embryonic development. Although many of its side effects are known, due to seizures and their consequences, it is not always possible to stop treatment during pregnancy. The objective of this study was to stage embryos treated and not treated with phenytoin using the Hamburger and Hamilton methodology. The project has a scientific certificate from CEUA UFCAT - 002-2023. Thirty eggs were donated by the INATA and distributed into two study groups, control (CO) treated with saline and phenytoin (FE) treated with 6 mg/kg. The embryos were incubated with temperature and humidity control, 24 hours after the start of incubation, the embryos were treated with 20µL of the solution, and the embryos were collected after 3 days. Staging was analysed for Hamburger and Hamilton methodology. Statistical evaluation was performed using the student's t-test, average, standard error and p≤0.05. The CO group presented a stage with an average of ~ 50 hours ± 6 hours of development, while the FE group showed delayed development, with a stage of  $\sim$  30 hours  $\pm$  4 hours. In the CO group 22.2%; 44.5% and 33.3% were at 24h, 48h, and 72h of development, respectively. In FE, 71.4% of the embryos were 24 hours old and 28.6% were 72 hours old. The formation of a vascular network and heart rate were only identified in embryos that had more than 72 hours of development. The embryos that had a formed vascular network were 33.3% in CO and 28.6% in FE. Regarding vitality, the results were 60% of live embryos in the CO group and 46.6% in embryos of the FE group. With the results, we observed that phenytoin causes delayed embryonic development in chicken embryos.

**Keywords:** Embryo chicken; Teratogenic; Epilepsy; Embryotoxicology; Embryonic development.



Title	Evaluation of sperm chromatin in stallions using the Nicolleti method
Authors	André Luiz Oliveira de Souza Pedro Sanches Oquendo Elisa Santanna Monteiro da Silva Marcelo Emilio Beletti
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	26 - Biologia do Desenvolvimento e Medicina Regenerativa

### and Kevwords

Alterations in sperm chromatin generally led DNA fragmentation, which can inhibit embryonic development when an oocyte is fertilized by a sperm with these alterations. Sperm Chromatin Structure Assay (SCSA) is used for evaluating sperm chromatin (gold method). Methods that identify apoptosis in somatic cells, such as Nicoletti, have been suggested as an alternative to SCSA. Nicoletti method uses propidium iodide (PI) staining after acid hydrolysis and evaluation in flow cytometry, since apoptotic cells with fragmented DNA would lose DNA and them show less intense fluorescence than normal cells. The aim of this study was to test the Nicoletti method to identify sperm chromatin alterations in stallions. Ten semen samples from different stallions were used. These samples were evaluated in a flow cytometer using the SCSA and Nicoletti methods and then a Pearson correlation test was carried out between the results of the two methods. The Nicoletti method showed a non-significant negative correlation (R=0.05, p=0.89) with SCSA, demonstrating at first that it is not a good alternative for identifying chromatin alterations in stallion sperm. However, we noticed that the application of Nicoletti method resulted a greater fluorescence intensity in some cells. Pearson's correlation test was then carried out between the SCSA and the percentage of cells with greater fluorescence intensity found in the Nicoletti method, which showed a positive and significant correlation (R=0.66, p=0.04). As the histones in sperm chromatin are replaced by protamines, which gives it very high compactness and great protection for the DNA, the sperm with alterations suffered a loosening of the chromatin, without loss of DNA, which made it easier for the IP to access the DNA, causing them to show greater fluorescence intensity. We conclude that this adaptation of the Nicoletti method is an alternative to SCSA for identifying chromatin alterations in stallion sperm. PARECER Nº 33/2023/CEUA/PROPP/REITO



Title	Maternal nicotine exposure induces changes in the anxiety- like behavior of adult male offspring
Authors	Silveira CCR¹ Mota APC¹ Cavalcante KVN¹ Ferreira-Junior MD¹ Xavier CH³ Lisboa PA² Gomes RM¹
Affiliations	<ul> <li>1 - Universidade Federal de Goiás, Laboratorio de Fisiologia Endocrina e Metabolismo, Brasil</li> <li>2- Universidade do Estado do Rio de Janeiro, Laboratorio de Fisiologia Endocrina, Brasil</li> <li>3- Universidade Federal de Goiás, Laboratoria de Neurobiologia de Sistemas, Brasil</li> </ul>
Session	28 - DOHaD desreguladores endocrinos

#### Abstract and Keywords

Nicotine is a highly addictive substance found mainly in tobacco. It has a significant impact on human behavior, influencing a variety of cognitive processes. Since its discovery, nicotine has been widely consumed in different forms, whether through traditional smoking, electronic cigarettes or other nicotine consumption devices. Its effect on human behavior is complex and multifaceted, affecting areas such as cognition, mood, motivation and even socialization. Accordingly, we sought to understand the role of nicotine in the behavior of adult offspring of rats induced to nicotine exposure through minipumps. The mothers were separated into two groups, Control (CO) where they underwent implantation of minipumps with 0.9% saline solution and Nicotine group (NIC, 6mg/kg). This exposure to nicotine was carried out from the first day of birth (PN1) until the 14th day of lactation (PN14). The offspring were monitored until PN180, when open field (OF) and elevated plus maze (EPM) tests were performed. Regarding the open field test, in NIC males we observed an increase in time in the center (CO 14.74  $\pm$  2.07 vs NIC 31.52  $\pm$  7.48 sec). However, no significant difference was observed in their time in the periphery (CO 284.7  $\pm$  2.18 vs NIC 268.5  $\pm$  7.49 sec). Regarding self-care, we observed a reduction in cleaning time (CO 39.94  $\pm$  6.28 vs NIC 22.28  $\pm$  5.21 sec). In the elevated plus maze test, there was no change in the time and entry of NIC males when compared to CO. In both tests, NIC females did not show significant changes when compared to the CO group. Based on these results, NIC animals had a less anxious profile than CO animals, and this response was sex specific, as females did not show any changes.

**Keywords:** Nicotine; DOHaD; Anxious-type Behavior.



Title	Perinatal exposure to bisphenol A induces morphological changes in the adult ovarian tissue
Authors	Isabella Barreto de Souza <sup>1</sup> , Isabella Barbosa Melvin <sup>1</sup> , Eloá Alves Bento <sup>1</sup> , Fernanda Cristina Alcântara dos Santos <sup>1</sup> , Manoel Francisco Biancardi <sup>1</sup> , Thalles Fernando Rocha Ruiz <sup>2</sup> , Sebastião Roberto Taboga <sup>2</sup> , Ellen Cristina Rivas Leonel <sup>1</sup>
	1 – Department of Histology, Embryology and Cell Biology Institute of Biological
Affiliations	Sciences, Federal University of Goiás, Goiânia, Brazil
	2 – Instituto de Biociências, Letras e Ciências Exatas, Universidade Estadual
	Paulista Júlio de Mesquita Filho, São José do Rio Preto, Brazil
Session	Poster/Oral Presentation

#### Abstract and Keywords

Bisphenol A (BPA), a polycarbonate monomer frequently used in the manufacture of plastics and resins, is an endocrine disruptor (ED), acting directly on cellular and tissue functions. The objective of this work was to describe the effects of this ED, when administered during the period of perinatal development, on the ovarian tissue composition of female Mongolian gerbils (Meriones unguiculatus). four groups of 5 pregnant females were formed (n=5). Two groups (↓BPA and ↑BPA) were treated orally with BPA (doses of 50 µg/kg and 5000 μg/kg), and an oil control group (OC) and an intact control group (IC) were established; treatments occurred from the 8th day of pregnancy until 21st postnatal day. The females of the litters were euthanized at 6 months of age and had their ovaries fixed and histologically processed. Analysis of the extracellular matrix components by stereology was performed after Gomori's trichrome, resorcin/fuchsin and Reticulin stainings. Data was subjected to ANOVA followed by Tukey's test to compare exposed groups. Results showed a greater deposition of collagen  $(38,0\pm7,5\%)$ , elastic  $(7,4\pm0,9\%)$ , and reticular fibers  $(31,5\pm1,8\%)$ in the ovarian stroma of the group treated with ↑BPA when compared to controls  $(20,7\pm6,2,2,3\pm0,5, and 14,1\pm2,3 in CO and 21,7\pm5,1,2,6\pm0,9, and 12,9\pm1,2$ in CI, respectively). It was also observed that the interstitial glands of the ovary reduced or delayed their development due to BPA exposure. Furthermore, in the control groups, the interstitial constitution of the ovary was related to steroids hormones-producing interstitial glands. In the BPA treated groups, the greater amount of connective tissue with less glandular tissue may be related to the progression of a pro-tumoral microenvironment, pointing to the deleterious effects of this ED on the morphophysiology of ovarian tissue. Such variation in the gland's incidence and establishment of ovarian fibrosis highlights the harmful impacts of BPA on the ovarian tissue.

Keywords: perinatal development, Mongolian Gerbils, steroid hormones



Title	Long-term effects of maternal dietary restriction during lactation on renal function in adult male rat offspring
Authors	Matos MO
	Cavalcante KVN
	Silva MER
	Soares RBA
	Ramiro MN
	Lima-Sobrinho JAB
	Ferreira-Junior MD
	Gomes RM
Affiliations	Laboratório de Fisiologia Endócrina e Metabolismo, UFG, Goiânia, Goiás
Session	DOHaD e Desnutrição

### and Kevwords

It is already known that protein restriction during pregnancy promotes a reduction in nephrogenesis in fetuses, posing a risk of damage to kidney function in the future. However, little is known about the impact of maternal caloric malnutrition during lactation on the renal function of these mothers offspring. The objective of this work is to evaluate the effects on renal function in offspring of mothers who underwent dietary restrictions during lactation. Pregnant Wistar rats were divided into two experimental groups: Control group (CO) fed with standard diet ad libitum and food restriction group (FR) fed with standard diet restricted to 50% of the daily food intake of CO mothers. The nutritional intervention was carried out from the post-natal day 1 (PN1) until PN14. The experimental protocols and sample collections were performed at PN120 (ethics committee 052/17). Male and female offspring did not showed difference in kidney weight (males CO 0.304±10.21 vs FR 0.290±4.03 mg/bw; females CO 0.368±26.96 vs FR 0.316±6.23 mg/bw), when compared to control groups. Despite no changes in female offspring, male FR offspring showed reduction in the glomerular filtration rate (CO 3.22±0.35 vs FR 2.25±0.12 ml/min) and increased total urinary protein (CO 82.82±11.79 vs FR 123.8±8.37 mg/dL), reduction in glomerular area in the male FR offspring (CO 7259±271.7 vs FR 6349±106.3 µm2) without changing the number of glomeruli per field (CO  $9.19\pm0.55$  vs FR  $7.85\pm0.71$   $n^{0}$ /field), in comparison to CO offspring. On the other hand, there was no difference in the fluorescence analysis for oxidative stress. In conclusion, maternal dietary restriction causes functional and morphological kidney damage in adult males, being a sex-specific response, since females did not present alterations, the alterations found were not caused by oxidative stress.

**Keywords:** Maternal dietary restriction, Kidney dysfunction, Lactation.



Title	Maternal dietary restriction during lactation promotes behavioral changes and impacts HPA axis in the offspring at adulthood
Authors	Cavalcante KVN <sup>1</sup> Ferreira-Junior MD <sup>1</sup> Silva MER <sup>1</sup> Ribeiro JVV <sup>2</sup> Pedrino GR <sup>3</sup> Xavier CH <sup>2</sup> Gomes RM <sup>1</sup>
Affiliations	<ul> <li>1 - Endocrine Physiology and Metabolism Laboratory, UFG, Goiânia, Goiás</li> <li>2 - Systems Neurobiology Laboratory, UFG, Goiânia, Goiás</li> <li>3 - Neuroscience and Cardiovascular Physiology Research Center, UFG, Goiânia, Goiás</li> </ul>
Session	29 DOHaD e Desnutrição

#### Abstract and Keywords

Hypothalamus-Pituitary-Adrenal (HPA) axis is sensitive to nutritional changes, since glucocorticoids participate in the regulation of metabolism and behavior. However, the role of nutritional stress in mothers during lactation in the HPA axis of their offspring at adulthood is poorly understood. In order to evaluate the behavioral and hypothalamic effects on the offspring of mothers in food restriction, pregnant Wistar rats were divided intro two groups (ethics committee 052/17): Control (CO) fed with standard chow ad libitum; and Food Restriction (FR), fed a standard diet, fixed at 50% of the CO group daily intake. The dietary intervention was carried out from the post-natal day 1 (PN1) until PN14. The offspring were monitored until PN120, when open field (OF) and elevated plus maze (EPM) tests were performed and brain samples were collected for molecular analyses, blood sample were collected to biochemistry and adrenal glands to morphological analyses. In the OF, FR male offspring spent more time in the center (CO  $34.14 \pm 3.70$  vs FR  $59.38 \pm 9.35$  seg) and increased number of crossings (CO  $51.94 \pm 4.29$  vs FR  $69.25 \pm 4.44$  events). However, we observed a reduction in the area of the adrenal zona fasciculata in FR male offspring (CO 944  $\pm$  2.67 vs FR 835  $\pm$  6.78  $\mu$ m2) and a reduction in ACTH levels (CO 59,38  $\pm$  15.89 vs FR 18.77  $\pm$  5.46 pg/ml) compared to their counterparts. Furthermore, although ACTH levels increased (CO 18 ± 3 vs RR  $53 \pm 2$  pg/ ml), FR female offspring showed reduced adrenal weight (CO 25  $\pm$  5.3 vs FR 21  $\pm$  3 g/pc) and reduced zona fasciculata area (CO 1166  $\pm$  10.0 vs FR  $1055 \pm 7.3 \mu m^2$ ), compared to CO equivalents. Taken together, restricted diet during lactation lead to reduced anxious behavior and increases exploratory activity in the male offspring at adulthood. However, both male and female offspring present reduced area of zona fasciculata in the adrenal gland, which together with behavioral tests suggest reduced stress signaling and a possible HPA axis unbalance.

**Keywords:** Food Restriction; Lactation; Behavior; HPA axis.



Title	Maternal dietary restriction during lactation impacts glucose homeostasis of their offspring at adulthood
Authors	Ramiro MN Cavalcante KVN Matos MO Soares RBA Lima-Sobrinho JAB Ferreira-Junior MD Gomes RM
Affiliations	Universidade Federal de Goiás, Laboratório de Fisiologia Endócrina e Metabolismo, Brasil
Session	29 DOHaD e Desnutrição

Abstract and Keywords Hunger is a global problem, which leads to long-term effects on metabolism. However, the impact of a restrictive maternal diet during lactation, which is also an important ontogenetic window for offspring health, is not well understood. The objective of this work was to evaluate the metabolic effects of maternal calorie restriction on offspring in adulthood. Pregnant Wistar rats were divided into two groups: Control (CO) and Food Restriction (FR) groups. FR mothers were fed a standard diet, fixed at 50% of the CO group daily intake. The nutritional intervention was carried out from the post-natal day 1 (PN1) until PN14. The offspring of both sexes were monitored until PN120 where the glucose tolerance test (GTT), insulin tolerance test (ITT), and pancreas collection were performed. All protocols were approved by an ethics committee (052/17). At weaning, both male (CO 41.77  $\pm$  2.34 vs FR 28.78  $\pm$  3.45 g) and female (CO 41.78  $\pm$  4.74 vs FR  $28.05 \pm 2.34$  g) FR offspring had reduced weight gain. However, at the end of the experimental period both male and female FR offspring showed similar weight than their CO counterparts. FR male offspring showed no change in the glucose homeostasis. However, FR female offspring presented higher area under curve (AUC) during GTT (CO 398  $\pm$  5.67 vs FR 433  $\pm$  6.32 a.u.) and lower AUC during ITT (CO 409  $\pm$  4.32 vs FR 327  $\pm$  2.34 a.u.). Despite no observable functional diferences, pancreatic islet hypertrophy was observed in FR male offspring (CO 12757  $\pm$  203 vs FR 17912  $\pm$  167  $\mu$ m<sup>2</sup>), but females presented no differences. Together, our results indicate that maternal caloric restriction is capable of altering body weight gain, and impacts glucose homeostasis of their offspring in a sex-specific manner at adulthood.

**Keywords:** Caloric Restriction; Lactation; Metabolic Programming; Glucose Homeostasis.



Title	Changes in the autonomic control of cardiovascular function and sodium sensitivity in adulthood as a result of gestational hypothyroidism
Authors	Cruz e Cruz BT¹, Alves JM¹, Balbi APC¹, Custódio CHX², Colombari E³, Pedrino GR², Ferreira-Neto ML¹.
Affiliations	<sup>1</sup> Laboratory of Electrophysiology and Cardiovascular Physiology, Department of Physiology, Biomedical
	<sup>2</sup> Institute of Biological Sciences, Federal University of Goias, Goiania, Brazil. <sup>3</sup> School of Dentistry, São Paulo State University (UNESP), Araraquara, Brazil.
Session	Poster Session

# and Keywords

Currently, it is known that internal and external stimuli to the intrauterine environment during fetal development can have important physiological consequences life during adulthood. Several studies have shown that different diseases in adulthood may have their origin in the early stages of life. A decrease in maternal thyroid hormone levels, as seen in gestational hypothyroidism (GHT) cases and during the lactation period, is related to problems known to affect fetal heart development, thus resulting in and long-term problems cardiovascular diseases. However, the mechanisms underlying these effects remain to be fully unraveled. Therefore, to this work was aimed at unveiling the autonomic implications of GHT during gestation and lactation periods upon cardiovascular systems during adulthood. For this purpose, we measured the arterial pressure, heart rate, baroreflex function and autonomic cardiovascular control of adult Wistar rats that underwent fetal and neonatal GHT (CEUA Ptortocol 071/18). We found that offspring underwent hypothyroidism during pregnancy and/or lactation showed significantly less weight gain in the first 90 days after birth. These rats also presented an increase in both mean arterial pressure and systolic blood pressure, in addition to an increase of the sympathovagal index when compared to the control group. Also, they presented a reduction in the barorreflex function as measured by pharmacological challenges, which allowed detecting a predominance of sympathetic influence on heart and vessels. In conclusion, our results showed that thyroid hormone deficiency at early stages of life can chronically impairs the autonomic control of cardiovascular function during adulthood.

**Keywords:** arterial pressure, baroreflex, heart rate, gestational hypothyroidism, cardiovascular diseases, sympathovagal index.



Title	Combined or isolated high-fat and high-sugar diets increase adiposity and liver lipid inclusion in juvenile mice
Authors	Soares RBA <sup>1</sup> Cavalcante KVN <sup>1</sup> Matos MO <sup>1</sup> Ramiro MN <sup>1</sup> Ferreira-Junior MD <sup>1</sup> Lima-Sobrinho JAB <sup>1</sup> Boer PA <sup>2</sup> Gomes RM <sup>1</sup>
Affiliations	<sup>1</sup> Laboratório de Fisiologia Endócrina e Metabolismo, UFG, Goiânia, Goiás <sup>2</sup> Laboratório de Programação Fetal e Metabolismo Hidroeletrolítico, UNICAMP, Campinas, São Paulo
Session	DOHaD e Hipernutrição

# and Keywords

Puberty is an ontogenetic window, in which important changes in the metabolism can be imprinted and impact health outcome throughout the life. Approximately 30% of adolescents in Brazil present metabolic syndrome, due to overweight and obesity. Since it has been proved that hypercaloric dietary patterns are linked to obesity in the puberty, and the obesity impacts overall health at adulthood, it is necessary to evaluate which obesogenic diet are more prone to induce metabolic impairments. To assess whether high-fat or high-sugar, or their combination, were more harmful to the health, we used a model of juvenile obesity to analyze metabolic parameters in male and female C57BL/6 mice (Ethics committee protocol 116/22). For this, we separated the animals into four groups: Control (CO); High-Fat (HF, 45% w/w fat chow); High-Sugar (HS, 20% w/v sucrose instead water); and High-Fat + High-Sugar (HFHS, HS plus HF diets). The dietary intervention was from postnatal day 30 (PN30) until PN60. At the end of the experimental period, there was an increase in body weight only in HFHS males (CO 22.91  $\pm$  2.5 vs HFHS 25.38  $\pm$  1.0 g) and females (CO 18.53  $\pm$  1.5 vs HFHS 20.27 ± 0.6 g), compared to CO counterparts. However, an increase in retroperitoneal adipose tissue were observed in both groups and sexes (male CO  $34 \pm 8.8 \text{ vs HS}$   $113.7 \pm 5 \text{ vs HF}$   $131.2 \pm 1.2 \text{ vs HFHS}$   $210 \pm 2.5 \text{ g}$ ) and (female CO 31.17±8.73 vs HS 64.50±2 vs HF 105.3±4.17 vs HFHS 92.80±2 q) compared to CO counterparts. Furthermore, HF and HFHS groups showed reduced normalized liver weight (CO 6.38  $\pm$  2.8 vs HF 4.08  $\pm$  2.5 vs HFHS 3.67  $\pm$  1.8 q/bw) and, on the other hand, increased hepatic lipid inclusion in both male and female experimental groups. Based on these data, despite overall body weight gain only in HFHS groups, both diets impact on adiposity and liver fat inclusion, which are hallmarks of metabolic syndrome.

**Keywords:** Juvenile obesity; Obesogenic diets; Adiposity; Liver steatosis.



Title Authors	Maternal diets with different lipid levels and its gastrointestinal repercussions for the offspring: A model of fetal programming in Wistar rats  Fortunato-Silva, J¹; Rezende, LP¹; Ferreira-Neto, ML¹; Bispo da Silva, LB²; Balbi, APC¹.
Affiliations	1 Federal University of Uberlandia (UFU) - Department of Physiology; 2 Federal University of Uberlandia (UFU) - Department of Pharmacology.
Session	DOHaD and Hipernutrition.

### Abstract and

It is known that altered diets during pregnancy can result in diseases in the offspring's adult life. In this way, we investigate the gastrointestinal (GI) effects of a maternal high fat-diet on the offspring. Pregnant rats were divided into groups: C (control group: dams exposed to a normolipid diet - 3.5%), E1 (experimental group 1: dams exposed to 28% lipid diet) and E2 (experimental group 2: dams exposed to 40% lipid diet). Maternal parameters such as feed and fluid intakes and body weight variation were evaluated. The GI functional tests performed on the male offspring of 30 day-old were: ulcer index - UI, motility of small (S) and large intestines (LI) and SI enteropooling. Other evaluations were performed such as the relative weight and area of the stomach and SI, in addition to the its length. We also evaluated the total thickness of the stomach wall, the portions of the SI (duodenum, jejunum and ileum) and the LI (cecum, colon and rectum), as well as the thickness of each layer: mucosa, submucosa and external muscle. The number of villi/area (NV/A) was also studied in each portion of the SI. There were no differences between the groups for maternal weight gain and fluid intake during pregnancy, but dams from E1 and E2 groups ingested more lipids than control dams, although E2 group consumed less food, carbohydrates and proteins than dams from the other groups and dams from E1 consumed more calories. The E2 pups had lower body weight, stomach weight and area, while E1 had greater thickness of the gastric wall due to the increase in mucosa. Regarding SI, there was no difference between the groups for length, duodenal and jejunal measurements, as well as for the NV/A, but E2 presented a greater relative weight of this organ, despite having a smaller total thickness of the ileum, due to a smaller mucosal area. The total thickness of the colon was smaller, because there was a reduction in the thickness of the mucosa and muscular layers. The maternal high-fat diet caused important GI structural changes in the young offspring from Wistar rats, which could have a functional negative impact in the long term. It is possible that these changes are the result of an imbalance between macronutrients in the maternal diet, not just due to excess lipids.

**Keywords:** high-fat diet maternal, offspring, gastrointestinal changes, Wistar rats.



Title	Effect of high-intensity interval training on treadmill
	performance in a 2,400m track test: A case study
	Jefferson Fernandes de Sousa <sup>1</sup>
	Mário Eduardo Santos Rodrigues <sup>2</sup>
	Robson da Silva Medeiros <sup>3</sup>
	Fanny Gonçalves de Lima <sup>3</sup>
	Flander Diego de Souza <sup>1</sup>
Authors	Silvio Soares dos Santos <sup>2</sup>
	Guilherme Gularte de Agostini <sup>2</sup>
	Elmiro Santos Resende <sup>3</sup>
	Frederico Balbino Lizardo <sup>4</sup>
	Adriano Alves Pereira <sup>1</sup>
	Thiago Montes Fidale <sup>1,5</sup>
	1 - Postgraduate Studies in Biomedical Engineering - Federal University of
	Uberlândia - Uberlândia, Brazil;
Affiliations	2 – Physical Education College - Federal University of Uberlândia - Uberlândia,
	Brazil;
Amilacions	3 – Medical School - Federal University of Uberlândia - Uberlândia, Brazil;
	4 – Institute of Biomedical Sciences - Federal University of Uberlândia -
	Uberlândia, Brazil;
	5 – Biotechnology Institute, Federal University of Catalão, Goiás, Brazil.
Session	34 - Controle Motor e Biomecânica

# and Keywords

Running is a widely embraced adult physical activity popularity. Effective physical training programs tailored to the specificities of sports practices are integral to enhancing athletes' performance and recovery during competitive phases. High-Intensity Interval Training (HIIT) has emerged as a prominent training method, offering distinct advantages. The biomechanics of running, encompassing variables such as stride frequency and length, play a significant role in athletic performance. Despite differences between track and treadmill running, the latter is often employed in biomechanical and physiological studies due to its control over factors, space efficiency, and laboratory conditions. The study, approved by the Research Ethics Committee of the Federal University of Uberlândia through the Brazil Platform (Opinion Number: 3.397.582, CAE: 13624419.2.0000.5152), aimed to assess the impact of a HIIT protocol on a treadmill on the performance of a 2400m track test. A 33-year-old male volunteer, an experienced street runner with no reported health restrictions, participated. Following four weeks of HIIT, the volunteer demonstrated a remarkable 3.86% reduction in the 2400meter test time on a 200-meter asphalt track. Biomechanical analyses revealed notable improvements, including a 1.06% increase in stride frequency, indicating enhanced energy utilization efficiency. Additionally, there was a significant 7.59% increase in stride length, suggesting more effective biomechanics. These improvements, combined with a 4.13% increase in Vo2max, underscore the substantial biomechanical benefits achieved by the volunteer. In conclusion, the implemented HIIT protocol not only contributed to an overall enhancement in performance but also resulted in significant biomechanical improvements, showcasing its efficacy in optimizing athletic capabilities.

**Keywords:** Biomechanics, Street Running, High-Intensity Interval Training, Stride Length, Stride Frequency, and Performance.



Title	Influence of core fatigue on landing biomechanics in recreational runners with and without dynamic knee valgus
Authors	Gina Olívia Brigido da Costa Curi Franciele Dias da Costa Thiago Ribeiro Teles dos Santos Valdeci Carlos Dionisio
Affiliations	Faculdade de Educação Física e Fisioterapia, Universidade Federal de Uberlândia, Uberlândia, Brasil.
Session	Pôster

#### Abstract and Keywords

Inefficiency in neuromuscular control of the core has been linked to deficits in dynamic control of the lower extremities, predisposing runners to injuries. However, there is no study for female recreative runners with dynamic knee valgus (DKV). This study aimed to compare the lower limb and trunk biomechanical parameters during the single-leg drop landing test (SLDL) before and after muscle core fatigue of the female runners with and without DKV. Twenty-seven female recreative runners participated in this study approved by by the Human Research Ethics Committee (number 52473121.2.0000.5152). They were divided into the valgus (VG; n=14) and non-valgus (NVG; n=13) groups. During the SLDL, the movement was evaluated by a tridimensional system, force plate, and electromyographic activity of the major muscles of the dominant lower limb. Each variable was tested separately using repeated measures ANOVA. The results showed a reduction of minimum and maximum knee angles in the frontal plane and increased integral electromyography of gluteus medius (GMED) activity after core fatigue. Also, the VG group presented a greater hip flexion angle in the sagittal plane and vertical displacement of the shoulder compared to the NVG. Female recreative runners with and without DVK presented similar kinetic, kinematic, and electromyography parameters during SDLS. Still, the fatigue of the core increased the EMG activity of GMED in both groups, while there was a greater hip flexion angle in the sagittal plane, and greater vertical displacement of the shoulder in VG, suggesting that that group is more susceptible to core fatigue and, as a result, more risk of injury.

**Keywords:** biomechanics; drop landing; valgus knee; core muscles.



Title	Effect of trunk muscle fatigue on female novice runner's lower limb during the step-down test: A kinematic and electromyographic analysis
Authors	Vinícius Dias Barbosa Gina Olívia Brigido da Costa Curi Franciele Dias da Costa Victor de Souza Medeiros Valdeci Carlos Dionísio
Affiliations	Federal University of Uberlândia – UFU, Faculty of Physical Education and Physiotherapy- FAEFI, Uberlândia, Minas Gerais, Brazil.
Session	Poster presentation

#### Abstract and Keywords

Inexperience in running and trunk muscle fatigue could modify the dynamic posture and affect the lower limb alignment, leading to dynamic knee valgus. Regarding that, the step-down test (SDT) is a valuable functional evaluation that assesses the control of the trunk, pelvis, and lower limb. This study aimed to compare kinematic and electromyographic (EMG) patterns during the execution of the SDT in female novice runners before and after a trunk fatigue protocol. Twenty-five female runners participated in this study, approved by the Human Research Ethics Committee of the Federal University of Uberlândia (protocol 52473121.2.0000.5152). Volunteers were healthy, aged between 20 and 35 years, and ran up to 20 km/week, with no previous injuries in the last 3 months. The trajectory of segments was tracked by a three-dimensional system and synchronized with the electromyography activity of the tibialis anterior, gastrocnemius medialis, vastus medialis oblique, vastus lateral, biceps femoris, semitendinosus, soleus, gluteus medius, and gluteus maximus during three phases of movement: P1, 100ms before the beginning of the movement; P2, from the beginning to maximum knee flexion; and P3, from maximum flexion to the end of the movement. The paired t-test was used to compare pre- and postintervention. The results showed that the mean (p= 0.035) and maximum (p= 0.022) integrated EMG TA activity in P1 increased, while the mean (p= 0.034) integrated EMG GM activity in P2 was reduced post-intervention. Trunk muscle fatigue elicits a larger integrated EMG TA activity, which may indicate a preprogrammed reaction to initiate the movement. At the same time, it reduces the amount of integrated EMG GM activity in the eccentric phase during the SDT in female novice runners.

Keywords: Biomechanics; Trunk, Fatigue; Knee Dynamic valgus.



Title	Biomechanic analysis in amateur runners with and without dynamic valgus during the step-down test: Immediate effects of core muscle fatigue
Audiana	Franciele Dias da Costa
Authors	Gina Olívia Brigido da Costa Curi
	Thiago Ribeiro Teles dos Santos
	Valdeci Carlos Dionisio
Affiliations	Faculty Of Physical Education and Physiotherapy, Federal University Of Uberlândia, Uberlândia, Brazil
Session	Poster

#### Abstract and Keywords

Inadequate dynamic stability in the trunk and hips (core) can alter dynamic posture during running, but this topic must be addressed. The objective was to compare the kinematic and electromyographic (EMG) patterns during the stepdown test (SDT) in amateur runners with and without dynamic knee valgus (DKV) immediately after a core muscle fatigue (CMF) protocol. In this experimental and descriptive study approved by the Ethics Committee (nº 5,543,780), participated 34 amateur runners (between 20 and 35 years) were divided into DKV (n=17) and non-DKV (n=17) groups. The lower limb and trunk muscular strength were assessed. The kinematics and EMG activity of the anterior tibialis (TA), gastrocnemius medialis (GM), vastus medialis oblique (VMO), vastus lateralis (VL), biceps femoris (BF), semitendinosus (ST) and gluteus medius (GMED) and gluteus maximus (GMAX) were also evaluated, during SDT, before and after CMF. The integral of EMG activity (iEMG) was calculated in the before movement starts (P1), eccentric (P2) and concentric (P3) phases of SDT. After the intervention, for both groups, there was a greater shoulder mediolateral displacement (p<.05) towards the supporting limb, greater iEMG activity of the TA (p=.019), VMO (p=.022) in P1, and lower iEMG activity for the GM in P2 (p=.015). In P3, also was reduced the iEMG activity of GM (p=.006), VL (p=.012), ST (p=.014), BF (p=.032), and GMED (p=.014). The DKV group presented lower reduced hip extensor muscle strength (p=.049), greater iEMG activity for the VL (p=.043) in P3, and greater maximum hip angle in the frontal plane (p=.021). For both groups, the CMF led to greater shoulder displacement towards the supporting limb, producing a reduced activity of the main muscles of the lower limb. Still, the DKV group already had less strength in the hip extensors, which may have influenced the greatest VL activity and maximum hip angle in the frontal plane.

**Keywords:** running; EMG; dynamic valgus; functional test.



Title	Trunk fatigue effect on lower limb kinematic and electromyographic patterns during the single-leg drop-landing test in female novice runners
	Victor de Souza Medeiros
	Franciele Dias da Costa
Authors	Gina Olívia Brigido da Costa Curi
	Vinícius Dias Barbosa
	Valdeci Carlos Dionísio
Affiliations	Federal University of Uberlândia – UFU, Faculty of Physical Education and
	Physiotherapy- FAEFI, Uberlândia, Minas Gerais, Brazil.
Session	Poster presentation

#### Abstract and Keywords

The dynamic and high-impact nature of running and the grounding action of constant movement represent a significant postural control challenge and injuries. Trunk muscle fatigue could influence the dynamic control of the lower limbs in runners. The single-leg drop-landing (SLDL) is a scientifically tested assessment that can simulate the landing and help detect any neuromuscular control deficiencies in the lower limbs. This study aimed to compare the effect of trunk muscle fatigue on the kinematic and electromyographic patterns of the lower limb during SLDL in female novice runners. Approved by the Research Ethics Committee of University of Uberlândia (protocol 52473121.2.0000.5152), 18 female novice runners participated in the study. The fatigue protocol had three isometric exercises and three dynamic exercises performed consecutively until voluntary exhaustion, measured with a Borg scale. The kinematics was collected of the main lower limb joints, and electromyographic (EMG) of the muscles: tibialis anterior (TA), gastrocnemius medialis (GM), vastus medialis obliquus (VM), vastus lateralis (VL), biceps femoris (BF), semitendinosus (ST), soleus (SO) gluteus medius (Gmed) and gluteus maximus (Gmax) was collected during SLDL pre- and post-fatigue muscle trunk. The integrated EMG (iEMG) was calculated into 3 phases (F1, before movement starts; F2, eccentric; and F3, concentric). There was an increase in TA and SO mean iEMG in F1, a reduction in TA and GM maximum iEMG in F2, and an increase in the BF minimal iEMG in F2. Also, a reduced ankle angle and increased hip angle in the sagittal plane and a reduced knee angle in the frontal plane post-intervention. The induction of trunk muscle fatigue in female novice runners influenced the TA, SO, GM, and BF EMG changes, the kinematic pattern of the ankle and hip in the sagittal plane, and the knee in the frontal plane during the SDLD.



Title	Intra- and inter-examiner reliability of the forefoot-shank alignment analysis using Kinovea <sup>®</sup>
Authors	Victor Rodholfo de Oliveira Silva Ana Caroline Carvalho Rocha Ana Julya Santana Miranda Guilherme Naves Fernandes Lanna Rúbia Guimarães Azevedo Justino Oliveira Laura de Souza Mendes Leandro de Oliveira Câmara Marco Antônio Pereira Guimarães Galvão Valdeci Carlos Dionísio Thiago Ribeiro Teles Santos
Affiliations	Faculty of Physical Education and Physical Therapy, Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	Controle Motor e Biomecânica

#### Abstract and Keywords

Modifications of forefoot-shank alignment in the frontal plane impact the lower limb motion. For instance, forefoot varus was demonstrated to be associated with excessive pronation. The forefoot-shank alignment can be clinically measured using photos and 2D motion analysis software. Recently, Kinovea® has been used by a rising number of clinicians. The measurement properties of the procedures using Kinovea® to extract forefoot-shank alignment need to be determined to be able to consider this tool as an option. Thus, this study investigated the intraand inter-examiner reliability to analyze the forefoot-shank alignment using Kinovea®. A pilot study was conducted with five participants (24.2±3.5 years old, body mass: 69.0±14.8 kg, height: 1.70±0.10 m). The eligibility criteria were age ≥18 years, no history of surgery in lower limbs and spine and no pain or other musculoskeletal complaint. The participant was positioned in prone, a line was drawn bisecting the shank, and a rod with a strap was fastened to the metatarsal heads. Two trained examiners conducted the test and took bilateral photos with the ankle at 0°, capturing the foot and lower third of the shank. Three trials were performed on each side, resulting in six photos per participant. Then, eight examiners analyzed these photos independently and extracted the forefootshank alignment through Kinovea®. This procedure was performed two times, seven days apart. The mean angle was calculated for each side and used to calculate the intraclass correlation coefficient (ICC). The Ethics in Research Committee approved this study (CAAE 68074923.4.0000.5152). The results showed excellent intra-examiner (ICC<sub>3,k</sub>=0.805-1.000) and inter-examiner (Day 1  $ICC_{3,k}=0.998$  and day 2  $ICC_{3,k}=0.986$ ) reliability. Therefore, the findings of this pilot study revealed that extracting forefoot-shank alignment through Kinovea® presented proper reliability. This procedure showed the potential to be considered in the clinical setting. Keywords: Foot, psychometric properties, bidimensional analysis, biomechanics.



Title	Intra- and inter-examiner reliability of the pelvic transverse plane alignment measurement during the bridge test with unilateral knee extension using Kinovea®
Authors	Leandro de Oliveira Câmara; Marco Antônio Pereira Guimarães Galvão; Laura de Souza Mendes; Lanna Rúbia Guimarães Azevedo Justino Oliveira; Guilherme Naves Fernandes; Ana Julya Santana Miranda; Ana Caroline Carvalho Rocha; Victor Rodholfo de Oliveira Silva; Lilian Ramiro Felício; Thiago Ribeiro Teles Santos.
Affiliations	Faculty of Physical Education and Physical Therapy, Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	Controle Motor e Biomecânica

### and Kevwords

Core stability has been advocated as a crucial element during daily activities and sports motion. The bridge test with unilateral knee extension is a clinical tool to inform about the core capacity to maintain pelvic alignment when facing a load. The greatest misalignment in the transverse plane during this test is an outcome that can be measured through Kinovea®. Limited measurement properties were reported (only intra-examiner reliability) despite the use of Kinovea® for this purpose. Inter-examiner reliability is another important property since it informs about the agreement between different evaluators. Thus, this study investigated the intra- and inter-examiner reliability to analyze the transverse plane alignment during the bridge test with unilateral knee extension using Kinovea®. A pilot study was conducted with 10 participants (24.0±3.51years old, body mass: 72.6±13.8kg, height: 1.70±0.10m). The eligibility criteria were age ≥18 years, no history of surgery in lower limbs and spine and no pain or other musculoskeletal complaint. The participant was positioned in supine. Then, he was oriented to bend the knee, remaining the feet sole on the stretcher, raise his pelvis, and extend one of his knees, while keeping the trunk, hip, and lower limb in a straight line for 10 seconds. The test was recorded and repeated three times for each side, alternately. The greatest pelvic inclination in the transverse plane was extracted through Kinovea® independently by four examiners two times, seven days apart. The mean angle was calculated for each side and used to calculate the intraclass correlation coefficient (ICC). The Ethics in Research Committee approved this study (CAAE 68074923.4.0000.5152). The results showed excellent intra- (ICC<sub>3,k</sub>=0.910-0.999) and inter-examiner (Day 1  $ICC_{3,k}=0.986$  and day 2  $ICC_{3,k}=0.972$ ) reliability. Therefore, the findings of this pilot study revealed that the analysis through Kinovea® presented proper intraand inter-rater reliability.

**Keywords:** Pelvis, core, psychometric properties, bidimensional analysis, biomechanics.



Title	Games and toys to stimulate the sensorimotor skills of children with low vision: low-cost materials
Authors	Gabriella Araújo Silva; Ingrid Aparecida Reis Castro; Karina Pereira
Affiliations	1 – Universidade Federal do Triângulo Mineiro, Uberaba, Brasil 2 –Universidade Federal do Triângulo Mineiro, Uberaba, Brasil 3- Universidade Federal do Triângulo Mineiro, Uberaba, Brasil
Session	Apresentação de Pôster

#### Abstract and Keywords

Introduction: Being born with congenital or early acquired visual impairment affects and impairs the child's typical overall development, altering learning and recreation. Thus, the study aims to build activities, games and toys that stimulate the sensorimotor actions of children with low vision from 2 to 3 years old, using low-cost and easily accessible materials. Materials and Methods: Two studies were used as the basis for this study, which aimed to: identify, describe and verify the frequency of manipulative actions of children with low vision, in the age group of 2 to 3 years. Based on the results of these studies, low-cost games, games and toys were developed for this age group. Results: 5 games, toys and/or games were developed for children with low vision from 2 to 3 years old, which stimulate: -Play 1: tactile recognition and stimulation, global motor coordination, proprioception and balance. -Play 2: fine motor skills, recognizing colors, motor development, promoting visual stimuli and stimulating organization. -Play 3: fine motor development, recognition and Learning of colors and geometric shapes, normal motor development, cognition, visual stimulation, and organization. -Play 4: stimulate fine motor skills, object recognition, touch, fine motor coordination and socialization. -Play 5: stimulate auditory perception, imagination, rhythmic awareness and reasoning. Conclusion: The games, toys and/or games are made of low-cost materials, using various stimuli and facilitating access for children with moderate visual impairment.

**Keywords:** Games and toys, children, low vision, motor skills, low-cost materials.



Title	Stimulation of gross motor skills in autism spectrum disorder and the work of a multidisciplinary team: a case report
Authors	Gabriella Araújo Silva; Alice Martins Ribeiro
Affiliations	1 – Universidade Federal do Triângulo Mineiro, Uberaba, Brasil 2- Centro Universitário do Triângulo - UNITRI, Uberlândia, Brasil
Session	Poster Presentation

### Keywords

Abstract and Introduction: Autism Spectrum Disorder is classified as a neurodevelopmental disorder according to the following criteria: deficits in communication and social interaction and the existence of repetitive and restricted patterns of behavior, as well as activities or interests. Methodology: The present study is a case report of a 5-year-old and 2-month-old female patient diagnosed with autism spectrum disorder who was followed up at the Autism Spectrum Disorder Reference Center (CRTEA) in Uberlândia, Minas Gerais, Brazil. After evaluation of gross motor skills, it was found that the behavior of going up and down stairs was in deficit, compared to typical development. Muscle weakness, muscle hypotonia, and alterations in sensory processing were observed. The goal set for the child was to go up and down stairs without support during the period of 8 weeks. Results: The stimulation of the gross motor skills of the patient E. E. S. was performed with the psychology and physiotherapy professional through the science of Applied Behavior Analysis (ABA). ABA consists of behavior changes, those who are in excess decrease them, and those in deficit increase them. Training in this behavior was carried out in 8 weeks, in addition to sensory stimulation, training to strengthen the lower and upper limbs and balance in sessions of 2 times a week lasting 1 hour, and parental training for the family. Conclusion: The performance of the multidisciplinary team in the application of training with the active participation of the family contributed significantly to the improvement of the child's motor skills assisting in their coordination capacities and reducing their limitations in the execution of functional activities, achieving the proposed objective.

**Keywords:** Autism Spectrum Disorder, Child or Child, preschool, motor skills.



Title	Exploring the <i>in vitro</i> antiparasitic potential of BthTX-I and BthTX-II: Phospholipases A2 isolated from <i>Bothrops jararacussu</i> venom against <i>Toxoplasma gondii</i>
Authors	Vinícius Queiroz Oliveira <sup>12</sup> , Samuel Cota Teixeira <sup>2</sup> , Luísa Carregosa Santos <sup>3</sup> , Tassia Rafaella Costa <sup>1</sup> , Lorena Pinheiro Morais <sup>1</sup> , Emanuelle Lorrayne Ferreira <sup>1</sup> , Guilherme de Souza <sup>2</sup> , Marina Paschoalino <sup>2</sup> , Luana Carvalho Luz <sup>2</sup> , Eloisa Amália Vieira Ferro <sup>2</sup> , Daiana Silva Lopes <sup>3</sup> , Veridiana de Melo Rodrigues Ávila <sup>1</sup>
Affiliations	1- Laboratory of Biochemistry and Animal Toxins, Institute of Biotechnology - Federal University of Uberlândia, Uberlândia, Brazil. 2- Laboratory of Reproductive Immunophysiology, Institute of Biomedical Sciences - Federal University of Uberlândia, Uberlândia, Brazil. 3- Multidisciplinary Institute of Health, Federal University of Bahia - Anísio Teixeira Campus, Vitória da Conquista, Brazil.
Session	35- Parasitologia

#### Abstract and Keywords

Congenital toxoplasmosis is a zoonosis caused by the parasite *Toxoplasma* gondii, affecting women during pregnancy and resulting in severe consequences for both the mother and fetus. The ineffectiveness of conventional treatments, coupled with adverse effects such as severe toxicity, underscores the need to explore new therapeutic methodologies. Thus, we have considered a series of therapeutic approaches, including phospholipases A2, enzymes found in snake venom with potential antiparasitic effects. In this study, we investigated the antiparasitic effect of BthTX-I and BthTX-II, phospholipases A2 isolated from Bothrops jararacussu venom, on crucial processes of T. gondii infection. We assessed the viability of host human trophoblastic cells (BeWo) using the MTT method and conducted adhesion assays with anti-T. gondii antibodies. Additionally, we examined invasion and intracellular proliferation of the parasite through a β-Galactosidase reaction under distinct experimental conditions: postinfection and pre- treatment of parasites and pre-treatment of cells. The toxins demonstrated cytotoxic concentrations affecting 50% of cells (>100µg/mL for BthTX-I and 17.53  $\pm$  1.09 $\mu$ g/mL for BthTX-II) in BeWo viability. The IC50 value for parasite proliferation inside BeWo cells was 13.38  $\pm$  1.57 $\mu$ g/mL for BthTX-I and 20.1  $\pm$  1.13 $\mu$ g/mL for BthTX-II, resulting in a superior selectivity index for BthTX-I of >7.47 compared to BthTX-II of 0.87. Subtoxic concentrations of toxins significantly reduced the percentage of intracellular proliferation, adhesion and invasion of *T. gondii* in BeWo cells in all three experimental. When compared to the standard treatment (Sulfadiazine and Pyrimethamine) the toxins were more effective. In light of these findings, the antiparasitic effect of BthTX-I and II is emphasized, highlighting biotechnological potential for the potential development of new therapies for toxoplasmosis.

**Keywords:** Snake Venom, parasite, toxoplasmosis.



Title	Neospora caninum induces lethal pancreatitis in mice experimentally infected with high doses of tachyzoites
Authors	Ana Paula Navarro Gonçalves <sup>1</sup> Tiago Wilson Patriarca Mineo <sup>1</sup> Caroline Martins Mota <sup>1</sup> Flávia Batista Ferreira <sup>2</sup> José Roberto Mineo <sup>1</sup>
Affiliations	1 – Laboratório de Imunoparasitologia "Dr. Mário Endsfeldz Camargo",
	Departamento de Imunologia, Instituto de Ciências Biomédicas, Universidade
	Federal de Uberlândia, Uberlândia, Brasil
	2 – Laboratório de Biotecnologia em Modelos Experimentais, Universidade
	Federal de Uberlândia, Uberlândia, Brasil
Session	Parasitology

# and Keywords

Neospora caninum is an obligate intracellular parasite that causes neuromuscular disease in dogs and abortion in cattle. It is distributed worldwide, leading to significant economic losses to cattle raisers. Experimental models in mice have been used to further understand the pathogenesis of this disease, although the cause of death in these animals is still unknown. In the present work we aimed to evaluate the cause of death in mice experimentally infected with *N. caninum*. All experiments were done using WT female mice, infected with a lethal dose (3x10<sup>7</sup>) of tachyzoites of the N. caninum Liverpool (Nc-Liv) isolate (CEUA protocol 109/16). First, we observed through histopathological evaluation that the pancreas of the infected mice presented severe macroscopic and microscopic alterations. Based on experiments with sequential sampling of the infected mice, we found that lesions in the pancreas were already evident at day one after the infection, confirming the acute characteristic of the pancreatitis. We also determined that this phenomenon was only present after parenteral infections, since the lesions could only be observed in mice that received an intraperitoneal inoculation, and not on those that received parasite by gavage (oral). In order to comprehend the mechanism behind the causa mortis, we sought to determine if the crucial cytokine IFNy played a role in the onset of the fatal pancreatitis. We found that Ifng-/- mice did not develop the pancreatic lesions found in WT mice, evidencing the role of IFNy in the pathogenesis. In conclusion, we have determined that lethal N. caninum infections in the mouse model are induced by an acute pancreatitis, that depends on the parenteral route of infection and IFNy pathway. Keywords: Neospora caninum, pancreatitis, causa mortis, experimental models, mice.



Title	Comparative bioinformatic analysis of the dense granule proteins (GRAs) of Neospora caninum and Toxoplasma gondii
Authors	Ruth Opeyemi Awoyinka Jhoan David Aguillón Torres Tiago Wilson Patriarca Mineo
Affiliations	Laboratório de Imunoparasitologia "Dr. Mário Endsfeldz Camargo", Universidade Federal de Uberlândia, Uberlândia. Brazil
Session	Poster Presentation

#### Abstract and Keywords

Neospora caninum and Toxoplasma gondii are closely related parasites within the Apicomplexa phylum, with a significant impact on human and veterinary medicine. Both parasites have complex life cycles, and a major aspect of their pathogenicity is the secretion of dense granule proteins (GRAs). GRAs are important for the intracellular survival of these parasites, as they influence hostparasite interactions, acquisition of nutrients and modulation of the host's immune responses. Despite the great similarity between these parasites, little research has focused on N. caninum GRAs (NcGRAs), and their comparative analysis with T. gondii GRAs (TgGRAs) remains poorly studied. Carrying out a bioinformatic analysis, the study aims to fill this gap, supplying data on the conservation and divergence of Tg&NcGRAs. Using open-access databases, a list of Tg&NcGRAs are generated. Bioinformatic analyses, including orthology, synteny, and evaluation of structural features such as domain analysis and conserved motifs, are performed. The 3D structures of the GRAs are predicted, allowing a detailed comparison of their tertiary structures. The main findings of the study include the identification of 185 TgGRAs and 146 orthologous NcGRAs. The proteins have distinct identities, with most falling within the 61-70% range, suggesting variations in their functional roles. Furthermore, variations in signal peptides and transmembrane domains suggest that GRAs may differ in subcellular localization and activity. The existence of other conserved domains reveals functional components that these GRAs share in common. The 3D structural analysis also highlights the differences and similarities in the molecular architecture of these GRAs. The development of treatments and therapies for neosporosis and toxoplasmosis infections could be impacted by this research, which advances the understanding of the molecular features, functional variety, and evolutionary history of TgGRAs & NcGRAs.

Keywords: GRAs, conserved domanis, 3D structures, Neospora caninum, toxoplasma gondii.



Title	The hydroalcoholic extract treatment of <i>Punica granatum</i> L. fruit controls <i>Toxoplasma gondii</i> infection in extravillous human trophoblastic cells (HTR-8/SVneo)
Authors	Izadora S. Damasceno, Juliana G. de Oliveira, Guilherme de Souza, Samuel C. Teixeira, Marina Paschoalino, Luana C. Luz, Bellisa de F. Barbosa, Eloisa Amália V. Ferro.
Affiliations	Laboratório de Imunofisiologia da Reprodução, Universidade Federal de Uberlândia, Uberlândia, Brazil.
Session	35 Parasitologia

#### Abstract and Keywords

gondii is an obligate intracellular parasite that causes Toxoplasma toxoplasmosis. In immunocompetent individuals, this infection is generally asymptomatic. However, for pregnant women, tachyzoites can pass through the placental barrier and reach the fetus, risking the embryo development. Conventional treatment involves a combination of sulfadiazine and pyrimethamine, but is impaired by its toxicity and side effects. Therefore, the search of new therapeutic approaches is mandatory, and studies involving natural compounds from plant species are showing its diverse medicinal potentials. Pomegranate (Punica granatum L.) has antifungal, antiviral, antimetastatic, and anti-inflammatory activities; however, there is no published research presenting its activity against T. gondii, neither in the maternal-fetal interface. Thus, this study aimed to evaluate the in vitro effects of hydroalcoholic extract from the peel of P. granatum fruit at early and late of experimental infection of T. gondii in extravillous human trophoblastic cells (HTR-8/SVneo). Cytotoxicity of the compound was evaluated at different concentrations through the MTT assay. Parasite invasion, intracellular proliferation and the treatment effect in parasite reversibility were measure by the  $\beta$ -galactosidase assay. T. gondii morphology was verified by the scanning electron microscopy (SEM). As results, only the highest concentration of the extract was toxic. The non-toxic concentration reduced the invasion and intracellular proliferation of *T. gondii* in HTR-8/SVneo cells, and the parasitic activity was irreversible. Pre-treatment of tachyzoites reduced invasion and intracellular proliferation. SEM results showed ultrastructural changes on the parasite surface, torsion and pores, demonstrating cytoskeleton damage. Therefore, this study concluded that the treatment with the P. granatum extract was effective against T. gondii infection, proofing its potential therapeutic in the treatment of this infection.

**Keywords:** Congenital toxoplasmosis. Herbal treatment. *Toxoplasma gondii*. *Pomegranate* extract. HTR-8/SVneo.



Title	New compounds derived from protoporphyrin IX associated with photodynamic therapy: Cytotoxicity in <i>Leishmania</i> promastigotes
Authors	Cíntia de Campos Chaves¹ Mônica Soares Costa Veiga¹ Tássia Rafaella Costa¹ Maynne Duarte Suriani Franco² Thibault Joseph William Jacques Dit Lapierre² Júlia Maria Costa e Silva¹ Veridiana de Melo Rodrigues¹ Renata Santos Rodrigues¹ Celso de Oliveira Rezende Júnior² Tayana Mazin Tsubone² Kelly Aparecida Geraldo Yoneyama¹
Affiliations	<ul> <li>1 - Laboratório de Bioquímica e Toxinas Animais, Instituto de Biotecnologia,</li> <li>Universidade Federal de Uberlândia, Minas Gerais, Brasil.</li> <li>2 - Instituto de Química, Universidade Federal de Uberlândia, Minas Gerais,</li> <li>Brasil.</li> </ul>
Session	Parasitology

#### Abstract and Keywords

Leishmaniasis caused by Leishmania parasite is considered a neglected disease. Leishmaniasis represents a set of diseases with distinct clinical manifestations. The Cutaneous Leishmaniasis (CL) that affects the skin and/or mucous membranes presents therapy associated with high toxicity and cases of resistance. Thus, the development of new compounds is essential for improvement of disease therapy. Photodynamic Therapy (PDT) that consists on administration of a photosensitizer (PS) compound that becomes active after exposure to light, it can be considerer an interesting therapeutic approach for the treatment of localized infections, thus it could be an important tool for CL treatment. Porphyrins and their derivatives constitute a group of tetrapyrrolic compounds that are among the PSs capable of positively interacting with PDT. In this regard, two new compounds derived from protoporphyrin IX (PpIX), called mpPpIX and mPpIX, were analyzed for their cytotoxic effect on Leishmania amazonensis, L. braziliensis, and L. guyanensis. Briefly, promastigotes were placed on 96 well plates (5 x  $10^5$  parasites/well) in Schneider medium with 10%of SBF containing different concentrations of PSs in quadruplicates and exposed or not to LED green light for 5 min. Control parasites were included in assays and mantained in absence of PSs. The plates were incubated at 23°C for 48h. MTT assay was performed to determined of citotoxicities. At least two independent assay were developed. Results showed that mpPpIX and mPpIX at concentrations of 10  $\mu M$  and 5  $\mu M$ , in the presence of light, drastically reduced the cell viability of the three species analyzed. There was no citotoxicity in absence of light for all concentrations PSs assayed. Therefore, considering the results, mpPpIX and mPpIX can be important compounds for the development of a new therapeutic approach to CL.

Keywords: Leishmania, Photodynamic Therapy, Protoporphyrin IX, cytotoxicity.



Title	Intercellular adhesion molecule (ICAM)-1 is essential to control the adverse pregnancy outcomes during maternal infection in mouse model of congenital toxoplasmosis
Authors	Rafaela José da Silva¹ Guilherme de Souza¹ Marcos Paulo Oliveira Almeida¹ Iliana Claudia Balga Milián¹ Alessandra Monteiro Rosini¹ Samuel Cota Teixeira¹ Priscila Silva Franco¹ Angelica Oliveira Gomes² Neide Maria Silva³ José Roberto Mineo⁴ Eloisa Amália Vieira Ferro¹ Bellisa Freitas Barbosa¹
Affiliations	<ul> <li>1 - Laboratory of Immunophysiology of Reproduction at the Federal University of Uberlândia, Uberlândia, Brazil</li> <li>2 - Laboratory of Cell Interactions at the Federal University of Triângulo Mineiro, Uberaba, Brazil</li> <li>3 - Laboratory of Immunopathology at the Federal University of Uberlândia, Uberlândia, Brazil</li> <li>4 - Laboratory of Immunoparasitology "Dr. Mário Endsfeldz Camargo" at the Federal University of Uberlândia, Uberlândia, Brazil</li> </ul>
Session	35 - Parasitology

#### Abstract and Keywords

Congenital toxoplasmosis is caused by transplacental transmission of Toxoplasma gondii parasite from mother to embryo/fetus, leading to adverse effects to the offspring such as visual lesions, cranioencephalic malformations, mental retardation, and, in severe cases, miscarriage. Several factors are involved in the dissemination of T. gondii throughout the host tissues, which includes the intercellular adhesion molecule (ICAM)-1, expressed in models of human placental tissues and trophoblast cells in the context of *T. gondii* infection. Despite the recognized influence of ICAM-1 in the systemic toxoplasmosis, its contribution in the course of *T. gondii* infection during pregnancy are unknown, being this scenario the subject of investigation here. In this sense, C57BL/6 mice, Wild Type (WT) and knockout for ICAM-1 ( $^{-/-}$ ), were used according the ethical committee (CEÚA/UFU protocol number 142/16). Pregnant females from both genetic lineages were infected by 8 days (di), or not, with 5 cysts of T. gondii (ME49 strain) at the days 1 or 12 of pregnancy (dp), euthanized at 8 or 19dp, respectively, and analyzed in relation to pregnancy outcomes, parasite burden, and cytokine production. We evidenced that T. gondii-infected ICAM- $1^{-/-}$  females had a severe embryonic loss at 8dp, as well as increased fetal loss at 19dp, in relation to the respective WT counterparts. Despite at 19dp the parasitism was not altered neither in the placenta nor in the fetus under ICAM-1 depletion, at 8dp the parasite burden was significantly increased in the uterus of ICAM-1<sup>-/-</sup> females in relation to the WT control. Finally, females at 8dp from both lineages significantly increased the levels of IFN-y, IL-6, IL-10, and TNF-a cytokines in response to infection. In conclusion, our data demonstrate that ICAM-1 protects the pregnancy course against adverse gestational outcomes due to infection, especially at early pregnancy (8dp) in which the presence of ICAM-1 promotes a reduced uterine parasite burden.

**Keywords:** ICAM-1; *Toxoplasma gondii*; Congenital Toxoplasmosis; Mouse Pregnancy; Gestational Outcomes.



Title	Therapeutic Potential of Chalcones in <i>Toxoplasma gondii</i> Infection at Human Materno-fetal Interface
Authors	Marina Paschoalino <sub>1</sub> , Samuel C. Teixeira <sub>1</sub> , Guilherme De Souza <sub>1</sub> , Luana C. Luz <sub>1</sub> , Alessandra M. Rosini <sub>1</sub> , Joed P. De Lima Júnior <sub>1</sub> , Natalia Carine L. Dos Santos <sub>1</sub> , Rafael M. De Oliveira <sub>1</sub> , Daniel Pereira Sousa <sub>1</sub> , Matheus C. Barbosa <sub>1</sub> , Vilson Serafim Júnior <sub>2</sub> , Luís Octávio Regasini <sub>2</sub> , Eloisa Amália V. Ferro <sub>1</sub> , Bellisa F. Barbosa <sub>1</sub> .
Affiliations	<ul> <li>1 - Laboratório de Imunofisiologia da Reprodução, Universidade Federal de Uberlândia, Uberlândia, MG, Brasil.</li> <li>2 - Instituto de Biociências, Letras e Ciências Exatas, Universidade Estadual Paulista Júlio Mesquita Filho, São José do Rio Preto, SP, Brasil.</li> </ul>
Session	Parasitology

#### Abstract and Keywords

Toxoplasma gondii is an obligate intracellular parasite that causes toxoplasmosis.

The congenital transmission is one of the most severe form of this desease and occurs when a woman acquires the infection before or during pregnancy and T. gondii tachyzoites cross the placental barrier, affecting the fetus. In this case, the first-choice treatment involves the combination of sulfadiazine and pyrimethamine. However, these drugs are associated with intense adverse effects, leading to treatment discontinuation. In this sense, chalcones are a class of compounds belonging to the flavonoid family, easily found in products common to human consumption that have gained prominence due to their bioactive properties, including antiparasitic. The objective of this work was investigate the anti-T. gondii activity of synthetic compounds analogous to chalcones in a human maternal-fetal interface model. Human trophoblastic cells (BeWo) infected or not with *T. gondii* tachyzoites were treated with different concentrations of chalcones to evaluate cell viability (MTT assay), intracellular proliferation, parasite invasion, as well as the potential irreversible treatment effect in trophoblast (reaction of  $\beta$ -galactosidase). Scanning electron microscopy (SEM) was performed to verify morphological changes in T. gondii. As results, non-cytotoxic concentrations of chalcones irreversibly reduced the proliferation and invasion of *T. gondii* in BeWo cells. In addition, pre-treatment of the parasite was able to reduce intracellular proliferation and invasion, suggesting a mechanism of action related directly to parasite damage. The SEM analysis showed substantial ultrastructural changes on the parasites surface, such as plasma membrane rupture and torsion, demonstrating cytoskeleton impairment. Therefore, it can be concluded that chalcones may represent a promising alternative therapeutic approach in congenital toxoplasmosis.

**Keywords**: congenital toxoplasmosis; treatment; flavonoids.



Title	Evaluation of the regenerative process in neurons of the enteric nervous system of Chagasic patients with megacolon
Authors	Michelle Aparecida Ribeiro de Freitas <sup>1</sup> , Enio C. de Oliveira <sup>2</sup> , Axel Brehmer <sup>3</sup> , Alexandre Barcelos Morais da Silveira <sup>4</sup>
Affiliations	<ul> <li>1 - Departamento de Parasitologia, ICBIM, Universidade Federal de Uberlândia,</li> <li>Brasil</li> <li>2 - Departamento de Cirurgia, Faculdade de Medicina, Universidade Federal de</li> </ul>
	Goiás, Brasil
	3 - Institute of Anatomy I, University of Erlangen–Nuremberg, Erlangen,
	Germany
	4 - Laboratório de Neurociências, ICBIM, Universidade Federal de Uberlândia,
	Brasil
Session	

## Abstract and

Chagas disease, since its discovery in mid-1908, has infected thousands of people across the American continent, and even today, despite all the environmental and sociopolitical advances, it continues to affect new individuals. This disease, caused by Trypanosoma cruzi, can cause pathological changes in both the heart and gastrointestinal tract. The most common gastrointestinal changes are megaesophagus and megacolon, which occur due to the inflammatory process and consequent neuronal loss in the enteric nervous system (ENS). These neurons, damaged by the parasite and/or inflammatory process, may present a process of neuronal regeneration in response, which is still obscure. Therefore, the objective of this work was to evaluate the neuronal sub-classes of the ENS regarding their regenerative capacity in chagasic patients with megacolon and in non-chagasic individuals. For this, colon samples were used, subjected to fluorescence immunohistochemistry with a series of neuronal markers (Peripherin - Pan-neuronal marker; Calretinin - Primary afferent neurons; NPY -Interneurons; Ach and Substance P - Excitatory motor neurons; NOS and VIP inhibitory motor neurons). The results demonstrated that the immunoreactive area of neurons is much larger in uninfected individuals than in chagasic patients with megacolon and that there is a selective destruction of inhibitory motor neurons (VIP and NOS) and a consequent increase in the regeneration process of these neuronal classes in chagasic patients with mega-colon compared to the uninfected group. We believe that the regenerative process of these neurons may be decisive for the development or not of chagasic megacolon, and further work must be carried out to characterize the exact way in which this re-generation occurs.

Keywords: Neurotrophins; regeneration; Enteric nervous system; Chagas disease



Title	Detection of IgG antibodies against <i>Toxoplasma gondii</i> in pig serum samples using immunoassays based on synthetic peptides
Authors	Débora Karolla de Freitas Oliveira <sup>1</sup> , Bruna Barbosa de Sousa Simamoto <sup>1</sup> , Ricardo Toshio Fujiwara <sup>2</sup> , João Luis Garcia <sup>3</sup> , Tiago Wilson Patriarca Mineo <sup>1</sup> , José Roberto Mineo <sup>1</sup>
	1 – Laboratory of Immunoparasitology "Dr. Mário Endsfeldz Camargo", Institute
	of Biomedical Sciences, Federal University of Uberlândia, Uberlândia, Minas
	Gerais, Brazil.
Affiliations	2 – Department of Parasitology, Institute of Biological Sciences (ICB), Federal
	University of Minas Gerais, Belo Horizonte, Minas Gerais, Brazil.
	3 - Protozoology Laboratory, Preventive Veterinary Medicine Department, State
	University of Londrina, Londrina, Paraná, Brazil.
Session	Parasitology

## and Keywords

Toxoplasma gondii, a zoonotic parasite with worldwide distribution, is a major public health concern since it may be transmitted through consumption of undercooked meat. Surveillance of the disease in pigs is a crucial effort to undermine the spread of the infection and to effectively control, prevent, and minimize economic losses in the meat industry. To date, the serological surveillance continues to pose significant challenges in these settings, specially due to the expensive and laborious assay procedures. Therefore, this study aims to develop immunoassays based on synthetic peptides predicted from major immunodominant antigens of *T. gondii*. For that purpose, we used serum samples from pigs experimentally infected with either tachyzoites (n= 22) and oocysts (n= 14) of *T. gondii* and performed ELISAs and Dot Blots using 27 selected peptides. We first observed that the assays were able to clearly distinguish positive and negative samples. We also observed that peptides derived from some SRS, GRA and MIC protein showed better reactivity in the ELISA, although most of the tested peptides showed reasonable detection in Dot Blots. On the other hand, neither assay, regardless of the peptides used, were able to distinguish in antibody reactivy in pigs infected with different infective forms. Based on these results, we were able to optimize and validate serological assays that can be an essential step in developing new rapid, accessible and practical tests.

**Keywords:** *Toxoplasma gondii*, serological diagnosis, pigs, peptides, immunoassays.



Title	Evaluation of protective role of anti-glycosylphosphatidylinositol antibody fragment in murine cerebral malaria
Authors	Sandra Gabriela Klein <sup>1</sup> ; Kelem Cristina Pereira Mota <sup>2</sup> ; Bruna Cristina Borges <sup>1</sup> ; Mylla Spirandelli Vieira <sup>2</sup> ; Matheus Morais Neves <sup>1</sup> ; Ludmilla Silva Mendes <sup>1</sup> ; Fabiana de Almeida Araújo Santos <sup>2</sup> ; Flávia Batista Ferreira <sup>1</sup> ; Isabela Lemos de Lima <sup>1</sup> ; Wânia Rezende Lima <sup>3</sup> ; Murilo Vieira da Silva <sup>1</sup>
Affiliations	<ul> <li>1 - Laboratory of Biotechnology in Experimental Models, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>2 - Nanobiotechnology Laboratory, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>3 - Biotechnology Institute, Federal University of Catalão, Catalão, Brazil</li> </ul>
Session	35 - Parasiltology

### and

Malaria, a parasitic disease transmitted by Anopheles mosquitoes, primarily affects children under five, pregnant women, and immunosuppressed individuals, caused by protozoa of the Plasmodium genus. It presents as uncomplicated malaria, with symptoms like fever, headaches, and body aches, and complicated malaria, characterized by metabolic acidosis, respiratory distress, and neurological signs, with cerebral complications responsible for most deaths and disease-related sequelae. Despite treatment protocols targeting parasite replication, inefficacy in managing neurological symptoms underscores the need for investigating novel drugs. Thus, this study aimed to determine the therapeutic single-chain potential οf variable fragment (scFv) targeting glycosylphosphatidylinositol (GPI) from Plasmodium falciparum in murine cerebral malaria. Experiments were approved by the Ethics Committee on Animal Use (CEUA) of the Federal University of Uberlândia (UFU) under protocol 153/16. C57BL/6J mice infected with *Plasmodium berghei*-ANKA (PbA) were divided into groups treated with anti-GPI scFv or phosphate-buffered saline (PBS) on days 0, 3, and 6 post-infection, alongside uninfected groups receiving the same treatments. Evaluation included mortality, parasitemia, and cerebral microhemorrhages. Our results show that anti-GPI scFv treatment in PbAinfected animals rescued 71.4% from mortality, significantly reduced parasitemia with more pronounced reductions on the 5th and 6th days post-infection, and decreased cerebral hemorrhages on the 6th day post-infection. Thus, P. falciparum anti-GPI scFv demonstrates significant therapeutic efficacy in experimental cerebral malaria, though further studies are needed to validate its

**Keywords:** nanobiotechonoly, antibody, plasmodium berghei, treatment.



Title	Histological alteration of glycogen trophoblastic Ccells in congenital malaria
Authors	1- Duarte Eduardo Pereira 2- Débora de Oliveira Santos 3- Neide Maria Silva
Affiliations	Universidade Federal de Uberlandia, Uberlandia- Minas gerais, Brasil.
Session	Parasitology

#### Abstract and Keywords

Gestational malaria is an important public health problem that occurs mainly in tropical and subtropical regions. It is defined as the presence of plasmodium in maternal peripheral blood or the finding of the parasite in the placenta, which can result in harmful consequences for the pregnant woman, the fetus and the newborn.

The mouse placenta accumulates glycogen stores in a subtype specialized in trophoblastic cells called glycogen trophoblastic cells (CTG), some of which interact closely with the maternal decidua, whose function is poorly under-stood. Like mouse CTG, they are considered analogous to human extravillous cytotrophoblasts responsible for the invasion of the decidua and myometrium, mainly due to the fact that they also contain large amounts of glycogen. For this reason, mice establish as an ideal model to elucidate the function(s) of placental glycogen stores.

Studies in animal models have led to the hypothesis that placental glycogen stores provide a source of glucose to support fetal growth during late gestation. This study aimed to evaluate the volume, number of placental CTG and placental glycogen stores, in pregnant mice on gestational day (dg) 19, 6 days after infection.

The experimental procedures were approved by the Ethics Committee on the Use of Animals at UFU, (CEUA protocol 060/19). Eight-week-old C57BL/6 mice were intravenously infected with P. berghei ANKA expressing GFP. at 13 dg, fetuses were euthanized, weighed, the placenta extracted for histological analysis (Periodic Acid Shiff Staining) and measurement of the uteroplacental layers using the "brush tool" of the QuPath software.

Note that infected animals that developed gestational malaria had changes in the quantity and color of CTG in relation to their control, being directly proportional to parasitemia. Plasmodium, the parasite that causes malaria, can cause changes in the accumulation of glycogen in CTG and the formation of glycogen vacuoles. These changes may have impacted levels of fetal growth and development.

**Keywords:** gestational malaria, fetal growth and glycogen cells



Title	Use of kDNA molecular markers for detection and genotyping of Leishmania species
Authors	Beatriz R Ferreira <sup>1</sup> ; Marcos V B Santos <sup>1</sup> ; Mariana S Cardoso <sup>1</sup> ; João L R Cunha <sup>3</sup> , Laila V Almeida <sup>1</sup> ; Ricardo T Fujiwara <sup>1</sup> ; Célia Gontijo <sup>2</sup> ; Gabrielle A Bento <sup>1</sup> ; Daniella C Bartholomeu <sup>1</sup>
	1 – Federal University of Minas Gerais, Departament of Parasitology, Brazil
Affiliations	2 – René Rachou Research Center, Oswaldo Cruz Foudation, Brazil
	3 - University of York Heslington,York,England
Session	35 - Parasitologia

#### Abstract and Keywords

Leishmaniases, neglected tropical diseases caused by obligatory intracellular parasites of the genus Leishmania, present diverse manifestations of clinical importance in both human and veterinary medicine. Despite being extensively studied, there are gaps in species-specific identification and diagnosis, highlighting the need and urgency to develop a robust panel of genetic markers allowing for the identification of Leishmania species with high sensitivity and specificity. kDNA sequences, the parasites' mitochondrial DNA, exhibit a high number of copies and are promising targets to enhance the sensitivity of PCR reactions. In this project, we identified species-specific markers derived from kDNA sequences of the maxicircle of L. amazonensis, L. braziliensis, L. guyanensis, L. infantum, L. lainsoni, L. major, L. mexicana, L. panamensis, and L. shawii species through sequence alignment and identification of polymorphic regions. The specificity of the markers was verified by standardizing annealing temperatures using gDNAs from 9 Leishmania species as targets. So far, markers for L. amazonensis, L.braziliensis, L. guyanensis, L. infantum, L. major, L. mexicana, L. panamensis, and L. shawii have been standardized with annealing temperatures ranging from 53°C to 66°C. The detection threshold was determined through serial dilution of gDNAs, ranging from 50 nanograms to 1 femtogram. In the future, we will expand the panel of species used in standardization to include Trypanosoma cruzi strains, Babesia sp., 12 different Leishmania species, and 5 other trypanosomatids (Crithidia, Endotrypanum, Herpetomonas, Leptomonas, and Phytomonas), as well as testing samples from naturally infected sand flies, dogs, and humans. Thus, it is believed that an efficient, rapid, and low-cost technique for the diagnosis and genotyping of Leishmania species associated with infection can be established.

Keywords: PCR, kDNA, Leishmania



Title	Effect of helminthic antigenic extracts during <i>Toxoplasma gondii</i> infection in human trophoblastic cells
Authors	Fabíola Teixeira Alves Bhrenda Carolliny Guardieiro Jesus Maria Eduarda Silva Diniz Luana Carvalho Luz Priscila Silva Franco Vanessa da Silva Ribeiro Bellisa de Freitas Barbosa Henrique Tomaz Gonzaga
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brazil
Session	Parasitology

# Abstract and Keywords

Neglected tropical helminthiasis and congenital toxoplasmosis are worldwide public health problems. Epidemiological surveys found between 50-90% of positive serology for toxoplasmosis in the studied populations. It is estimated that more than 2 billion people worldwide are infected with helminths, including Ascaris lumbricoides, species of the Taenia genus, and Strongyloides stercoralis, most of them in tropical regions. Coinfection between intestinal helminths and the protozoan Toxoplasma gondii is certainly not uncommon. There are no studies in the literature evaluating use of antigenic extracts from A. lumbricoides, Taenia crassiceps and S. venezuelensis during Toxoplasma gondii infection in human trophoblastic cells. In this context, this study aimed to evaluate the effects of helminths saline extracts (SE) on human trophoblastic cells infected with *T. gondii* regarding cell viability and parasite proliferation. Human villous trophoblastic cells (BeWo lineage) were used as a host cell model. Initially, the cytotoxicity of treatment with SE at serial concentrations of 64, 32, 16, 8, 4, 2, 1, 0.5, 0.25, and 0.125  $\mu$ g/ml was assessed using MTT assays. Different concentrations of SE exhibited no toxicity when compared to the control. Subsequently, the intracellular replication index of T. gondii was evaluated through a proliferation assay, analyzed by  $\beta$ -galactosidase reaction. In this assay, BeWo cells infected with T. gondii were treated with concentrations of 32, 8, and 2 µg/ml for each stimuli. Our data from a maternal-fetal interface model of toxoplasmosis showed: the absence of cytotoxic effects on host cells and noninterference of helminth antigen extracts, when tested individually, in the proliferation rate of *T. gondii*. The possible effects on the risk of the congenital form are within our scope horizon, including combinations of SE, or even with medications such as anthelmintics and corticosteroids.

**Keywords:** Parasite-host interaction; toxoplasmosis; tropical helminthiasis.



Title	Assessment of exposure to <i>Ascaris lumbricoides</i> and <i>Trichuris trichiura</i> in individuals with systemic lupus erythematosus
Authors	Vinícius Campos Miranda <sup>1</sup> ; Jordânia Costa Pinto <sup>3</sup> ; Lilian Lacerda Bueno <sup>3</sup> ; Ricardo Toshio Fujiwara <sup>3</sup> ; Cristiane Guimarães Morais <sup>1,2</sup> ; Rodrigo Rodrigues Cambraia de Miranda <sup>1</sup> ; Natália Berne Pinheiro <sup>4</sup> ; Humberto Machado Resende <sup>5</sup>
Affiliations	<ol> <li>Laboratório de Diagnóstico, Epidemiologia e Controle de Helmintos (LADECH), Departamento de Parasitologia (DEPAR), Instituto de Ciências Biomédicas (ICBIM), Universidade Federal de Uberlândia (UFU), Uberlândia/MG, Brasil.</li> <li>Campus Avançado Uberaba Parque Tecnológico (CAUPT), Instituto Federal do Triângulo Mineiro (IFTM), Uberaba/MG, Brasil.</li> <li>Laboratório de Imunobiologia e Controle de Parasitos (LICP), Instituto de Ciências Biológicas (ICB), Universidade Federal de Minas Gerais (UFMG), Belo Horizonte/MG, Brasil.</li> <li>Instituto de Biologia (IB), Universidade Federal de Pelotas (UFPel), Pelotas/RS, Brasil.</li> <li>Hospital de Clínicas da Universidade Federal de Uberlândia</li> </ol>
Session	35 - Parasitologia

# Abstract and Keywords

Systemic lupus erythematosus (SLE) is a multisystemic autoimmune rheumatic disease characterized by an abnormal immune system self-attack. The treatment with immunosuppressive, makes individuals with this condition more susceptible to other diseases and infections. Geohelminthiasis are diseases caused by a group of soil-transmitted helminths and are currently the most widespread parasitic infections. A 100 blood samples from patients diagnosed with SLE were collected at HC-UFU and they were determined as the SLE group. For a control group (CT Group), samples were collected from individuals with corresponding age and sex and free of chronic diseases. This project was approved by the CEP (Research Ethics Committee of the Federal University of Uberlândia, CAAE: 63571422.1.0000.5152). Data on the degree of disability caused by the autoimmune disease were collected using the Health Assessment Questionnaire (HAQ-DI) and correlated with the presence of IgG antibodies against soiltransmitted helmiths. For immunoassays to quantify IgG antibodies, as an indirect ELISA, plates were sensitized with a pool of synthetic peptides predicted as B-cell epitopes of A. lumbricoides or T. trichiura. The Reactivity Index (RI) of IgG antibodies was obtained with a 3-fold cutoff. Data were analyzed using GraphPad Prism 8.0 software. The SLE group and the CT group demonstrated non-parametric distribution and no significant difference between them with regard to IgG IR (p>0.05). The HAQ-DI between seroreactive and nonseroreactive individuals showed a significant difference between them (p < 0.05), suggesting that seroreactivity to parasites influences the patient's clinical condition. There was no correlation between the HAQ-DI values and the reactivity index of the seroreactive samples from the SLE group. These results contribute to understanding the relationship between helminthiasis and autoimmune diseases.

**Keywords:** Immune Response, Geohelminthiasis, IgG, autoimmune diseases



Title	Environmental contamination with helminths in soils of public areas in the western zone of Uberlândia/MG
Authors	Ana Júlia Machado Farnezi¹ Rebeca Barbassa Ching Zanoni¹ Eduardo Ramalho Orlando¹ Wellington Barbosa de Paiva¹ Isadora Aparecida Ferreira Silva¹ Lara Cardoso Rabello¹ Mariana de Magalhães Maximiano¹ Natália Berne Pinheiro² Rodrigo Rodrigues Cambraia de Miranda¹
Affiliations	<ul> <li>1 - Laboratório de Diagnóstico, Epidemiologia e Controle de Helmintos (LADECH),</li> <li>Departamento de Parasitologia (DEPAR), Instituto de Ciências Biomédicas</li> <li>(ICBIM), Universidade Federal de Uberlândia (UFU), Uberlândia/MG, Brasil</li> <li>2 - Instituto de Biologia (IB), Universidade Federal de Pelotas, Pelotas/RS, Brasil</li> </ul>
Session	35 – Parasitologia

## and Keywords

Geohelminths are intestinal parasites belonging to the phylum Nematoda that develop in warm and humid soil during a portion of their life cycle. Soil contamination by feces from animals and humans in public areas poses a public health risk, especially for children. The objective of this study was to assess the presence of helminth eggs and larvae in soil samples from the western zone of Uberlândia/MG. Soil samples were collected from 13 squares in the western zone of the city. In each square, five soil samples were obtained, each weighing approximately 250g. Soil analysis was conducted using the Caldwell & Caldwell technique adapted by Gallina et al. (2011) for egg concentration through centrifugal flotation in a sodium dichromate solution. For each collected sample, six grams of soil were used for the technique processing. Briefly, 10 mL of 4% sodium hypochlorite was added to the samples, which were homogenized and filtered through gauze. The filtered material was centrifuged, and the supernatant was discarded. The precipitate was then resuspended in 10 mL of sodium dichromate solution (1.35 mg.d $L^{-1}$ ) and centrifuged again. Subsequently, to form a meniscus, the sodium dichromate solution was added. Then, a coverslip was placed over the meniscus to collect the floating material. Each sample was analyzed in quintuplicate, using an optical microscope at magnifications of 100x and 400x. Among the 13 squares analyzed, seven (53.8%) were contaminated with parasite eggs. A total of 65 points were analyzed, of which 12 (18.4%) showed parasite eggs. The most prevalent parasites were from the Strongylida order (12.3%), the Ascaridida order (3%) and Toxocara spp. (3%). These data contribute to monitoring the occurrence and distribution of parasites with zoonotic significance in the municipality of Uberlândia/MG.

**Keywords:** soil-transmitted helminths (STH's); zoonosis; public health; Toxocara spp.; Ancylostoma spp.;



Title	Identification of antibodies against hookworm and Strongyloides stercoralis B cell epitopes in patients with systemic lupus erythematosus
Authors	Daniela da Silva Guimarães <sup>1</sup> ; Vinícius Campos Miranda <sup>1</sup> ; Humberto Machado de
	Resende <sup>2</sup> ; Jordânia Costa Pinto <sup>4</sup> ; Lilian Lacerda Bueno <sup>4</sup> ; Ricardo Toshio
	Fujiwara <sup>4</sup> ; Cristiane Guimarães Morais <sup>1,3</sup> ; Natália Berne Pinheiro <sup>5</sup> ; Rodrigo
	Rodrigues Cambraia de Miranda <sup>1</sup>
	1 – Laboratório de Diagnóstico, Epidemiologia e Controle de Helmintos (LADECH),
	Departamento de Parasitologia (DEPAR), Instituto de Ciências Biomédicas
	(ICBIM), Universidade Federal de Uberlândia (UFU), Uberlândia/MG, Brasil.
	2 – Hospital de Clínicas (HC/UFU), Universidade Federal de Uberlândia (UFU),
	Uberlândia/MG, Brasil.
Affiliations	3 – Campus Avançado Uberaba Parque Tecnológico (CAUPT), Instituto Federal
Allillations	do Triângulo Mineiro (IFTM), Uberaba/MG, Brasil.
	4 – Laboratório de Imunobiologia e controle de parasitos, Instituto de Ciências
	Biológicas (ICB), Universidade Federal de Minas Gerais (UFMG), Belo
	Horizonte/MG, Brasil.
	5 - Instituto de Biologia (IB), Universidade Federal de Pelotas (UFPel),
	Pelotas/RS, Brasil.
Session	35 - Parasitologia

#### Abstract and Keywords

Hookworm and strongyloidiasis are diseases transmitted by contact with soil contaminated with nematode larvae. Systemic Lupus Erythematosus (SLE) is a chronic autoimmune disease that is characterized by the loss of tolerance to selfantigens and the synthesis of different autoantibodies, with the formation and deposit of immune complexes. The immunological response to the presence of these helminths presents a T helper 2 (Th2) pattern mediated by cytokines that consequently stimulate IgG specific to the antigen. Blood samples were collected from 154 people who consented to participate in the study. Of these, 97 were diagnosed with SLE and 57 were free of autoimmune and chronic diseases. For those positive for SLE, a socioeconomic and Health Assessment - Disability Index (HAQ-DI) questionnaire was also applied. This project was approved by the CEP (Research Ethics Committee of the Federal University of Uberlândia, CAAE: 63571422.1.0000.5152). For all samples, the immunological response was evaluated using the indirect ELISA technique in order to detect total serum immunoglobulins G (IgG). The serological detection of anti-hookworm and anti-Strongyloides IgG was carried out with synthetic peptides, with specific action for interaction with B cells. For this serological detection, a pool of peptides (P),



specific for hookworms and Strongyloides stercoralis, was used in equimolar quantities: P1, P2, P8, P9, P13 and P18 at final concentration of 2  $\mu$ g/well and the serum at a dilution of 1/100. The frequency of seroreactive individuals is higher in the SLE group (37.11%) compared to the control group (19.29%). The mean HAQ-DI score does not present a statistical difference between the immunosuppressed and control groups. Our data suggest that immunosuppression caused by SLE favors greater exposure to hookworms and Strongyloides stercoralis.

Keywords: soil-transmitted helminths (STHs); autoimmune diseases; ELISA;



Title	Synthetic Compounds Derived from Phenolic Plant Metabolites as Potential Antiviral Agents Against Mayaro Virus <i>In Vitro</i>
	Nancy Araceli Juárez Contreras <sup>1</sup> , Natasha Marques Cassani <sup>1</sup> , Alvaro Luiz
Authors	Helena <sup>2</sup> , Patrick Rômbola Ozanique <sup>2</sup> , Chenglin Deng <sup>3</sup> , Bo Zhang <sup>3</sup> , Luis
	Octávio Regasini <sup>2</sup> , Ana Carolina Gomes Jardim <sup>1,2</sup> .
	<sup>1</sup> Laboratory of Antiviral Research, Institute of Biomedical Science (ICBIM),
Affiliations	Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil.
	<sup>2</sup> Institute of Biosciences, Humanities and Exact Science (IBILCE), São Paulo
	State University (UNESP), São José do Rio Preto, SP, Brazil.
	<sup>3</sup> Key Laboratory of Special Pathogens and Biosafety, Wuhan Institute of Virology,
	Center for Biosafety Mega-Science, Chinese Academy of Sciences, Wuhan, China.
Session	Poster

Abstract and

Mayaro virus (MAYV), the etiological agent of Mayaro fever, has shown an increase in reported cases in recent decades. This raises concern due to its potential to cause epidemic outbreaks, along with its clinical similarity to other mosquito-borne diseases. The absence of approved antiviral therapies for its treatment highlights the importance of exploring new therapeutic approaches to address this emerging disease. Psoralea corylifolia, a medicinal plant rich in phenolic metabolites, has been used in traditional medicine, and its extracts have shown antibacterial and antifungal activities. Therefore, in this study, we evaluated the *in vitro* antiviral activity of six synthetic compounds inspired by the phenolic metabolites present in Psoralea corylifolia against Mayaro virus infection. Vero-E6 cells were infected with MAYV-nanoluc at a multiplicity of infection (MOI) of 0.1 and were treated with the highest non-cytotoxic concentrations of each compound for 24h, previously determined by MTT viability assays. Virus replication levels were quantified by measuring nanoluciferase activity using the Renilla luciferase Assay System. The preliminary results show that the compounds P1, P2, P3, P4, P5 and P6 inhibited MAYV replication in vitro in 58.3%, 70.8%, 61.5%, 95.6%, 57.5% and 68.6%, respectively. These results point to the possibility of using Phenolic compounds derived from Psoralea corylifolia as candidates for the development of new antivirals against MAYV. Further studies are in progress to better understand the mechanisms of antiviral action that might be involved.

**Keywords:** Antivirals, Alphaviruses, Mayaro, Synthetic compounds, Psoralea corylifolia.

Financial support: FAPEMIG, CNPq, CAPES.



Title	Metallodrugs as a new potential class of antivirals against Mayaro virus
Authors	Ana Laura Costa Oliveira <sup>1</sup> ; Igor Andrade Santos <sup>1</sup> ; Antonio Augusto de Oliveira Neto <sup>2</sup> ; Igor Santos Oliveira <sup>2</sup> ; Jennyfer Castro da Silva <sup>2</sup> ; Camilla Abbehausen <sup>2</sup> ; Ana Carolina Gomes Jardim <sup>13</sup> .
Affiliations	<ol> <li>Universidade Federal de Uberlândia (UFU), Instituto de Ciências Biomédicas, Uberlândia, MG, Brasil;</li> <li>Universidade Estadual de Campinas (Unicamp), Instituto de Química, Campinas;</li> <li>Universidade Estadual Paulista (UNESP), Instituto de Biociências, Letras e Ciências Exatas, Campus SãoJosé do Rio Preto, SP, Brasil.</li> </ol>
Session	Virology/Microbiology

#### Abstract and Keywords

Mayaro virus (MAYV) is an arbovirus responsible for Mayaro fever, a neglected emerging disease that causes hyperthermia, muscle pain, and skin rashes. MAYV is mainly transmitted through Haemogogus genus mosquitoes, and in less incidence, by Aedes sp. Aggravatingly, the infection can develop into a chronic arthralgia and polyarthralgia. Currently, there are no vaccines or specific drugs for the treatment of MAYV. In this context, we evaluate the potential of new and unique metallodrugs with the aim of finding antiviral candidates against MAYV. In this research, 21 compounds were screened towards its cytotoxicity in Vero E6 at concentrations of 50, 10, and 2  $\mu$ M cells thought MTT assay after 24h. The highest non-cytotoxic concentration of each compound was used to treat Vero E6 cells in the presence of MAYV expressing the nanoluciferase gene (MAYVnanoluc) at a Multiplicity of infection of 1. Viral replication was measured by luciferase activity 24h post-infection. Among them, 11 were able to inhibit MAYV ≥ 70%: Met-a (95.5%), Met-c (90.6%), Met-A (96.8%), Met-9H (71.7%), and Met-9R (77.3%) at 50  $\mu$ M. On the other hand, at 10  $\mu$ M Met-b and Met-I inhibited 76.6% and 76.2%, respectively. Further, at 2 µM the molecules Met-E, Met-F, Met-G, and Met-H, impaired viral replication in 76%, 80.9%, 89.6%, and 77,7%, respectively. Thus we conclude that metallodrugs showed here demonstrated potent anti-MAYV activity in vitro, making it an interesting source of molecules for the development of antivirals against MAYV.

**Keywords:** Antiviral, metallodrugs, Mayaro virus, Mayaro fever.



Title	Molecular detection of <i>Borrelia</i> sp. in <i>Ornithodoros cavernicolous</i> (Acari: Argasidae) in midwestern Brazil
Authors	Mayara Garcia Polli <sup>1,2</sup> Maria Marlene Martins <sup>3</sup> Vinícius da Silva Rodrigues <sup>2,3</sup> Laís Miguel Rezende <sup>3</sup> Adriane Suzin <sup>3,4</sup> Rodrigo da Costa Maia <sup>2,3</sup> Ana Carolina Prado Souza <sup>2,3</sup> Sebástian Muñoz-Leal <sup>5</sup> Matias Pablo Juan Szabó <sup>3</sup> Jonny Yokosawa <sup>1*</sup>
Affiliations	<sup>1</sup> Laboratory of Microorganisms of Cerrado (Brazilian Savannah), Department of Microbiology, Instituto de Ciências Biomédicas (ICBIM), Universidade Federal de Uberlândia (UFU), Uberlândia, Brazil; <sup>2</sup> Post-Graduation Program in Applied Immunology and Parasitology, UFU, ICBIM, Brazil. <sup>3</sup> Laboratory of Ixodology (LabIx), Faculdade de Medicina Veterinária (FAMEV), UFU, Brazil. <sup>4</sup> Post-Graduation Program in Ecology and Conservation of Natural Resources, Instituto de Biologia, UFU, Brazil. <sup>5</sup> Departamento de Ciencia Animal, Facultad de Ciencias Veterinarias, Universidad de Concepción, Chillán, Ñuble, Chile.  *Electronic address: jonny.yokosawa@ufu.br
Session	36 Virologia/Microbiologia

#### Abstract and Keywords

Ticks are obligate hematophagous parasites that can transmit to vertebrates several pathogens, including viruses, bacteria, protozoa and helminths. Among these agents, some Borrelia species cause disease in humans and other vertebrate animals; therefore, they have human and veterinary health importance. To gather additional information on Borrelia species in Brazil, the current study aimed to detect the presence of these species in *Ornithodoros* cavernicolous ticks collected in September 2019 from cement pipes that were used by bats as shelter in a farm located in the midwestern region of Brazil. DNA samples obtained from 18 specimens of O. cavernicolous were subjected of two polymerase chain reactions targeting a segment of the Borrelia flaB gene. Of the samples tested, only one (6 %, 1/18) showed amplification. The nucleotide sequence of the amplified DNA showed 97.7 % (293/300) identity with a sequence of a Borrelia sp. detected in blood collected from a bat from Macaregua Cave, Colombia, and 97.3 % (292/300) with a Borrelia sp. Sequence detected in lungs from vampire bats from northeastern Brazil. The deduced amino acid sequences were identical to each other. Phylogenetic analysis indicated that these sequences formed a group of Borrelia species putatively associated with bats that is closely related to sequences of Borrelia species of the Lyme borreliosis



group. Further investigations should be carried out in order to determine whether the sequence of the *Borrelia* sp. we found belongs to a new taxon. It will also be of great importance to determine which vertebrate hosts, besides bats, *O. cavernicolous* ticks can parasitize in order to investigate whether the *Borrelia* sp. we found may be transmitted and cause disease to the other vertebrate hosts.

**Keywords:** Bats; *Borrelia* sp.; Brazil; Molecular detection; *Ornithodoros* cavernicolous.



Title	Prospection of Klebsiella quasipneumoniae phages
Authors	Pedro Antonio Moraes Souza <sup>1,2</sup> Lara Silva Reis <sup>1</sup>
	Tatiana Maria Lopes Canuto de Souza¹ Sabrina Royer³
	Deivid William da Fonseca Batistão <sup>4</sup> Jonny Yokosawa <sup>1</sup>
	1 - Laboratory of Cerrado (Brazilian Savannah) Microrganisms, Department of
	Microbiology, Instituto de Ciências Biomédicas (ICBIM). Universidade Federal de
	Uberlândia (UFU), Uberlândia, Brazil.
Affiliations	2– Post-Graduation Program of Applied Immunology and Parasitology.
	ICBIM/UFU.
	3 – Laboratory of Molecular Microbiology (MICROMOL) ICBIM/UFU
	4 - Departamento de Clínica Médica da Faculdade de Medicina FAMED/UFU,
	Uberlândia, Brazil
Session	Session 36 – Virology/Microbiology

#### Abstract and Keywords

In the post-antibiotic era, there's a need to discover or develop other antimicrobial drugs or methods which surpass bacteria multi-drug resistance. These pathogens keep on acquiring more resistances as time passes, and the highlights are on bacteria commonly found in healthcare associated infections, such as Klebsiella spp. Therefore, phage therapy can be researched to deal with these organisms, given the wide variety of phages found in almost any environment and the possibility of finding phages to almost any bacteria. This study aimed to detect lytic phages capable of infecting pathogenic Klebsiella quasipneumoniae (ATCC 700603). Five samples were collected from rivers and sewage in the cities of Uberlandia and Sao Paulo. After chloroform treatment, centrifugation, concentration and filtration with 0.22 µm filters, samples were individually incubated with of pathogenic K. quasipneumoniae. To verify whether there were bacteriophages, dilutions of these cultures were deposited over lawns of *K. quasipneumoniae* made on LB-agar plates. Lysis was only observed with the Sao Paulo sample, collected from Aricanduva stream. Five different lysis plaques were picked and subjected to three purification steps. All five bacteriophage clones showed to lyse the K. quasipneumoniae strain used for their isolation. The isolation and purification of bacteriophages of K. quasipneumoniae from sewage and river samples were successful. In the next steps, genomic and morphologic characterizations of these phages are going to be carried out, as well as their efficiency in lysate other strains of Klebsiella spp., including those that have been shown to be resistant to antibiotics.

Keywords: Phage therapy, bacteriophages, Klebsiella



Title	Lectins isolated from tropical plants strongly inhibit Chikungunya virus infection
	Lara Cristina Ferreira Freitas¹
	Igor Andrade Santos <sup>1</sup>
	Bruna Coelho Sandim <sup>1</sup>
Authors	Renato Rodrigues Roma <sup>3</sup>
	Romério Rodrigues Santos Silva³
	Claudener Souza Teixeira <sup>3</sup>
	Ana Carolina Gomes Jardim <sup>1,2</sup>
	1 – Institute of Biomedical Sciences, Federal University of Uberlandia, Uberlândia, Minas Gerais, Brazil;
	2 – Institute of Biosciences, Languages and Exact Sciences, Paulista State
Affiliations	University, São José do Rio Preto, São Paulo, Brazil;
	3 – Center of Agrarian Science and Biodiversity, Federal University of Cariri, Juazeiro do Norte, Ceará, Brazil

#### Abstract and Keywords

The Chikungunya virus (CHIKV) is a member of the Alphavirus genus that is transmitted through Aedes aegypty and A. albopictus mosquitoes, and is the etiological agent of chikungunya fever disease. Its symptoms include fever, joint pain, as well as arthralgia and polyarthralgia, and CHIKV infection can progress to a chronic condition for months or years, resulting in a disabling disease. Up to date, there is no specific antivirals approved to treat CHIKV infection and therefore, currently threatens public health systems. Lectins isolated from plants have been demonstrating several biological activites, becoming an interesting source of molecules with antiviral activity, due to their ability to bind to specific sugar regions in viral particles. There is lack of data comphreending the effect of lectins against CHIKV replication. Therefore, the aim of this work was to evaluate the anti-CHIKV activity of five lectins extracted to: C. ensiformi; C. maritima; P. platycephala; M. acutifolium; V. macrocarpade. To this, the lectins L1 to L5 were screened towards their cytotoxicity in BHK-21 cells at concentrations of 50, 10, and 2µg/mL employing MTT assay after 16h. The highest non-cytotoxic concentration of each lectin was selected to be evaluated against CHIKV expressing the nanoluciferase gene (CHIKV-nanoluc) in a multiplicity of infection of 0.1. Viral replication was measuraed by nanoluciferase levels after 16h postinfection. The lectins L1, L3, L4, and L5 were viable at 50µg/mL (94.8%, 97.4%, 92.7%, and 91%, respectively), while L2 was viable at 10µg/mL (107.4%). When evaluated against CHIKV-nanoluc the lectins L1 and L2 demonstrated to be active inhibiting 95.7% and 93.4%, in this order. On the other hand, L3 and L5 were able to inhibit only 16.8% and 13.2%, respectively, while L5 was inactive. In conclusion, the data here shows that L1 and L2 possess potent antiviral activity, being promising compounds for the development of a specific antiviral against CHIKV.

**Keywords:** Antiviral, Chikungunya virus, Lectins, Natural compounds.



Title	Evaluation of Viral Protein 2 (VP2) of Mogiana tick virus (MGTV) for the development of an immunoenzimatic assay and its application in the prevalence of MGTV Infection in cattle of farms in Brazilian savannah (Cerrado Biome)
Authors	Amanda Branquinho de Oliveira Cunha <sup>1,3</sup> Maria Tereza R. Oliveira <sup>1,4</sup> Ana Paula Mendes Muniz <sup>1,2</sup> Vinicius da Silva Rodrigues <sup>2,5</sup> Laís Miguel Rezende <sup>5,6</sup>
	Matias Pablo Juan Szabó <sup>5</sup> Jonny Yokosawa <sup>1</sup> Mayara Garcia Polli <sup>1,2</sup>
Affiliations	<ul> <li><sup>1</sup> Universidade Federal de Uberlândia (UFU), Instituto de Ciências Biomédicas (ICBIM), Laboratório de Microrganismos do Cerrado (LaMiCe), Uberlândia-MG;</li> <li><sup>2</sup> UFU, ICBIM, Programa de Pós-graduação em Imunologia e Parasitologia Aplicadas (PPIPA), Uberlândia-MG;</li> <li><sup>3</sup> UFU, Instituto de Biotecnologia (IBTEC), Curso de Biotecnologia;</li> <li><sup>4</sup> UFU, Faculdade de Medicina (FAMED), Curso de Enfermagem;</li> <li><sup>5</sup> UFU, Faculdade de Medicina Veterinária (FAMEV), Laboratório de Ixodologia (LabIx), Uberlândia-MG;</li> <li><sup>6</sup> FAMEV, Programa de Pós-graduação em Ciências Veterinárias (PPGCV), Uberlândia-MG.</li> </ul>
Session	35 Parasitologia

## and Keywords

Presently, diseases caused by virus transmitted to humans and other animals by arthropods cause big impact in public health. The main arthropod vectors are insects and ticks. Ticks are hematophagous ectoparasites, highly specialized and by having a large variety of vertebrate hosts may transmit to them a series of pathogens, including viruses. Recently, a new group of viruses belonging to the *Flavivirus* genus has gained prominence: the Jingmenvirus. Mogiana tick virus (MGTV), which belongs to this group, has been identified in *Rhipicephalus microplus* ticks, known as cattle ticks. Although MGTV has also been detected in cattle, the infection and the possible impact it causes in the animals are not known. In order to evaluate the prevalence of the MGTV infection in cattle in three farms of Cerrado biome of southeastern Brazil, we cloned the partial coding sequence of the viral protein two (VP2) of MGTV into a vector for expression in bacteria. The expression of the truncated VP2 (trVP2) was observed by sodium dodecyl sulfate – polyacrylamide gel electrophoresis and Enzyme-Linked



Immunosorbent Assay (ELISA) using the cell lysate and anti-6xHis monoclonal antibody. Then, trVP2 was purified using Ni-NTA columns under denaturing conditions and used as an ELISA antigen for testing bovine sera. The percentage of reactive samples was higher from cattle from the Gloria farm (100%, 31/31), which has mainly Holstein breed cattle for milk production, followed by those from the Lago Verde farm (50%, 26/52), with cattle of Girolando breed, and from the Capim Branco farm (49%, 20/41), which has mainly Nelore breed cattle for meat production. These results suggest a higher prevalence of MGTV infection in the former farm, which has cattle breed that is more resistant to ticks. More tests are needed to further evaluate the assay in order to be used as a diagnostic test to detect MGTV infection in cattle. This project was approved by the Ethics Committee on the Use of Animals (CEUA) of the Federal University of Uberlândia in accordance with protocol 001/21.

Keywords: VP2, MGTV, ELISA, Diagnostics



Title	Chalcone-like drugs as antiviral against Zika virus
Authors	Júlia Amaral Vieira d'Almeida <sup>1</sup> Natasha Marques Cassani <sup>1</sup> Letícia Ribeiro de Assis <sup>2</sup> Luis Octávio Regasini <sup>2</sup> Ana Carolina Gomes Jardim <sup>1,2</sup>
Affiliations	<ul> <li>1 - Institute of Biomedical Science (ICBIM), Federal University of Uberlandia ^ (UFU), Uberlandia, Minas Gerais, Brazil.</li> <li>2 - Institute of Biosciences, Humanities and Exact Sciences, São Paulo State University (UNESP), São José do Rio Preto, SP, Brazil.</li> </ul>
Session	34: Virology/Microbiology

### Abstract and Keywords

Zika fever is a disease caused by the Orthoflavivirus zikaense (Zika virus, ZIKV), mainly transmitted by mosquitoes from the genus Aedes. The symptoms are classified as dengue-like, however, more severe cases can present neurological disorders, such as Guillain-Barré syndrome and microcephaly in newborns of infected pregnant women. ZIKV belongs to the family Flaviviridae, that is characterized by viruses with a positive sense single stranded RNA. There are no vaccines and antivirals available against ZIKV, and therefore, the search for compounds with antiviral activity is mandatory. Natural and synthetic compounds derived from hidro- and nitrochalcones have shown promising antimicrobial and antiviral activity, highlighting the interest in investigating their potential against ZIKV. Vero E6 cells were treated with 11 natural or synthetic chalcones for 72 hours to established non-cytotoxic concentrations, performing MTT assay. DMSO was used as untreated control. The antiviral assay was performed infecting Vero cells with ZIKVPE243 at a multiplicity of infection of 0.01 in the presence of each compound at the established non-cytotoxic concentration. immunofluorescence assay was performed, and focus of infection (FFU) were counted. Cell viability and replication rate were calculated according to the equation  $(T/C) \times 100\%$ , where T and C represent the mean optical density of the treated group and vehicle control group, respectively. The results demonstrated that at the concentration of 50  $\mu$ M, the compounds E27, E31 and E40 inhibited 72%, 75% and 35% of ZIKV infection, respectively, while at 10  $\mu$ M, E1 and E2 inhibited 71% and 42%, respectively. At the concentration of 2  $\mu$ M, E13 and T10B were able to inhibit ZIKV in 32% and 49.1%, respectively. Our data shows that chalcone-like drugs may be useful templates in the development of potent antiviral drugs against Zika fever.

**Keywords:** Zika virus, antiviral, chalcones.



Title	Antimicrobial and antibiofilm activities of the proteolytic enzyme mixture bromelain extracted from Amazonian pineapples
Authors	Camargo GCB <sup>1</sup> , De Oliveira RC <sup>2</sup> , Pontes GS <sup>3</sup> , Do Carmo EJ <sup>4</sup> <sup>1</sup> UFAM Manaus, PPG Biotecnologia, Brasil; <sup>2</sup> UFAM Manaus, PPG Imunologia básica, Bra
Affiliations	<sup>1</sup> UFAM Manaus, PPG Biotecnologia, Brasil; <sup>2</sup> UFAM Manaus, PPG Imunologia básica, Brasil; <sup>3</sup> INPA Manaus, Lab Virologia e Imunologia, Brasil; <sup>4</sup> UFAM Manaus, Dpt de Parasitologia e Microbiologia, Brasil
Session	

# and Keywords

The pineapple, Ananas comosus, is a tropical fruit commonly known for its unique and acidic taste caused by the mixture of proteolytic enzymes called Bromelain which are present in all parts of the fruit. The Brazilian variety "Turiaçu" of pineapple is known for its low acidity and sweetness, being the most produced variety in the state of Amazonas. This study aims to extract and purify the Bromelain present in Amazonian Turiaçu pineapples and test its proteolytic derived antimicrobial and antibiofilm activities against clinical interest bacteria such as S. aureus, S. epidermidis and C. acnes. The extraction of Bromelain was done through blending, the purification of the extract was done via an in-house method utilizing different concentrations of acetone and proteolytic testing was done through casein assay. Minimum inhibitory concentration (MIC) and Minimum bactericidal concentration (MBC) were used to evaluate the Antimicrobial and antibiofilm activities of Bromelain through 96-well plate assay and disc-diffusion test. Through testing, the pineapples were divided into two groups: Mature (N) and Spoiled (P), signifying the fruit's state. The casein assay showed that the Bromelain-acetone purified extract had higher enzymatic activities at 90% concentration in both groups. MIC and MBC testing done in 96-well plate against S.aureus and S.epidermidis showed that both groups had significant antimicrobial activity (p<0.05), with the N group having an IC<sub>50</sub> of 2.67 mg/mL against S.aureus and of 2.54mg/mL against S.epidermidis and a IC90 of 1.45 mg/mL and 1.34 mg/mL, respectively; the P group had an IC<sub>50</sub> of 2.73mg/mL against S.aureus and 3.09mg/mL against S.epidermidis and a IC90 of 1.50 mg/mL and 2.07 mg/mL, respectively. The antibiofilm assay done via disc diffusion method showed no statistically significant results. This study showed that Bromelain has significant antimicrobial activity against clinical bacteria, but that this activity is greatly harmed by biofilm.

Keywords: Bromelain; MIC; MBC; Clinical interest bacteria; Amazon



Title	Development of a new platform for the detection of avian pathogenic Escherichia coli (APEC), using peptides selected by phage display
Authors	Iara Pereira Soares <sup>1</sup> Mário Machado Martins <sup>1</sup> Fabiana de Almeida Araújo Santos <sup>1,2</sup> Belchiolina Beatriz Fonseca <sup>1,2</sup>
Affiliations	1 – Laboratory of Nanobiotechnology Prof. Dr. Luiz Ricardo Goulart Filho, Institute of Biotechnology, Federal University of Uberlandia, Minas Gerais, Brazil 2 – School of Veterinary Medicine, Federal University of Uberlandia, Uberlandia, Brazil
Session	Virology/Microbiology

### Abstract and Keywords

The avian pathogenic E. coli (APEC) is considered a sub-group of extra-intestinal pathogenic E. coli (ExPEC). APEC can include the ST131 sequences that may be a multiresistent foodborne zoonotic pathogen and it has the potential to cause urinary tract infections. APEC strains constitute a heterogeneous group. This diversity makes it difficult to identify an avian E. coli isolate as a pathogenic or non-pathogenic strain. Thus, tools such as sequencing, phylogeny, Multilocus Sequence Typing and analysis of virulence factors are used to characterize the isolates. The Phage display is a process that uses high-affinity molecules that interact with targets and can therefore be used as a marker for diagnosis. The objective was to select molecules capable of recognizing APEC using Phage Display technology. The phage library was composed of 7 random amino acids expressed in the pIII region of the bacteriophage. Three negative selections were performed by incubating the phage library with commensal E. coli, and unbound particles were transferred for positive selection with APEC ST131. The Phage-ELISA assay was performed to analyze the reactivity of clones with APEC, after 3 rounds of biopanning. The DNA of the selected phages was subjected to DNA sequencing to determine the peptide sequences. Only 11 samples presented valid sequences, of these, 8 samples presented the same sequence, and presented a greater phage-ELISA reactivity ≥2.7 compared to commensal *E. coli*. These phages were subjected to another round of phage-ELISA and proved to be selective, recognizing only APEC ST131. In this context, the application of Phage-ELISA appears to be a potential marker for the diagnosis of the APEC ST131 strain.

Keywords: Phage Display, Phage-ELISA, avian pathogenic Escherichia coli, APEC



Title	Antiviral activity of coordinated Ruthenium-nitric oxide complexes against Zika And Chikungunya viruses
Authors	Natasha Marques Cassani¹  Uriel Enrique Aquino Ruiz¹  Giovanna André Antoniucci¹  Igor Andrade Santos¹  Daniel Oliveira Silva Martins¹,²  Ana Laura Costa Oliveira¹  Evelyn Christine de Souza Arantes³  Renata Galvão de Lima³  Ana Carolina Gomes Jardim¹,²
Affiliations	<ol> <li>Laboratory of Antiviral Research, Institute of Biomedical Science (ICBIM), Federal University of Uberlândia (UFU), Uberlândia, MG, Brazil.</li> <li>Institute of Biosciences, Humanities and Exact Science (IBILCE), São Paulo State University (UNESP), São José do Rio Preto, SP, Brazil.</li> <li>Institute of Exact and Natural Sciences (ICENP), Federal University of Uberlândia, Ituiutaba, MG, Brazil.</li> </ol>
Session	Virologia/Microbiologia

# Abstract and Keywords

Since its discovery and association with human infection, Chikungunya virus (CHIKV) and Zika virus (ZIKV) have shown potential health concern, unleashing serious outbreaks throughout the world. The need to find treatment that can help abrogate these viral infections of extreme clinical concern instigate the search for potential molecules that can act as potent antiviral drugs. In this sense, Ruthenium (Ru) complexes have shown interesting activity against RNA viruses, based on their chemical and biological properties. Here, the antiviral activity of four Ruthenium and nitric oxide (NO) coordinated molecules were evaluated against CHIKV and ZIKV. For that, Vero E6 were infected with ZIKVPE243 at a multiplicity of infection (MOI) of 0.01, and BHK-21 with CHIKV nanoluc at a MOI of 0.1, in the presence or absence of compounds for 72 and 16 hours, respectively. For replicon assays, BHK21-RepZIKV\_IRES Neo cells and BHK-CHIKV-NCT were treated non-cytotoxic concentrations of each compound, previously determined by MTT viability assays, and Renilla-luciferase activity levels were measured 72h later. The results demonstrate that the compounds Ru-1, Ru-2, Ru-3, and Ru-4 were able to inhibit CHIKV and ZIKV in vitro, with a dose-dependent activity, resulting in the selective index (SI) of 24.8, 1.7, 6.9, and 1.8, respectively for CHIKV, while 12.2, 3.3, 14.5, and 3.2 for ZIKV, respectively. Additionally, the treatment of stable cell lines harboring replicons with Ru-1 and Ru-2 inhibited 81.4% and 39% of CHIKV replication, respectively, and 72% and 84.2% for ZIKV, respectively. In conclusion, these data demonstrate the potential of these molecules as potential inhibitors of CHIKV and ZIKV infections.

**Keywords**: Zika, Chikungunya, Ruthenium, antiviral.



Title	Development of an immunoassay for confirmation of Mogiana tick virus (MGTV) infection in cattle on farms in the Triângulo Mineiro region
Authors	Guilherme Brandão Mamede Mayara Garcia Polli
Affiliations	Universidade Federal de Uberlândia, Uberlândia, Brasil
Session	36 - virology and microbiology

# Abstract and Keywords

The objective of the study is to evaluate, in Western Blotting, bovine sera previously confirmed positive in an ELISA test in order to verify their performance for the potential development of laboratory diagnosis. The study addressed the classification and characterization of the Mogiana tick virus (MGTV), identified in samples from cattle infected by Rhipicephalus microplus ticks in Brazil. The taxonomic challenge of the virus revealed its inclusion in the Jingmenvirus group, segmented and multicomponent, related to the Orthoflavivirus. The study involved the selection of positive bovine sera for MGTV by ELISA and their subsequent analysis by Western Blotting. The results indicated a clear reaction in the positive control, while bovine sera showed weak and nonspecific spots, pointing to possible failures in the detection method as proposed. Despite the limitations, MGTV is recognized as a potential economic and public health risk, demanding further research and improvement of diagnostic methods. The use of specific antibodies for cattle is proposed to enhance the sensitivity and specificity of the tests. In conclusion, the study underscores the ongoing importance of investigating MGTV and its interaction with cattle, aiming to develop effective strategies for prevention and control of outbreaks and epidemics associated with this emerging virus. This project was authorized to obtain blood samples from cattle from Fazenda Experimental Glória and Fazenda Experimental Capim Branco, both from UFU, and Fazenda Lago Verde, in Itumbiara, Goiás, according to protocol 001/21 approved by the Ethics Committee on the Use of Animals (CEUA) of the Universidade Federal de Uberlândia.

Keywords: Mogiana tick virus, tick, western blotting



Title	The resistance of bacillus spores: implications for the strain- specific response to high-performance disinfectants
Authors	Simone Sommerfeld Lidianne Fabricia dos Santos Tomais Isabelle Ezequiel Pedrosa Lara Reis Gomes Maysa Vitória Cunha Silva Danielle Silva Vieira Arthur Campoi Peluco Belchiolina Beatriz Fonseca
Affiliations	Graduate Program in Animal Science, Federal University of Uberlândia, Uberlândia, MG, Brasil
	Virology/Microbiology

## Abstract and Keywords

The presence of bacterial spores in materials and equipment can pose risks to the biosecurity. The spores are the best indicator of good disinfection. Although formoldehyde studies mention that (FOR), glutaraldeide Orthophthalaldehyde (ORT) are sporicidal, there have been relatively few recent studies on this subject to new or reclassified species. Then, we aimed to evaluate the efficacy of ORT and a solution of quaternary ammonia and glutaraldehyde (AG) inactivating spores of Bacillus thuringiensis (BT), B. cereus (BC) and two strains of B. velezensis (BV1 and BV2). The spores of BV2 and BT were disinfected with 22.5mg/m<sup>3</sup> of ORT by dry fumigation or 1mg/mL of AG by spray for 20 minutes, according to the manufacturer's recommendation. The use of AG did not reduce BV2 or BT spores. However, BV2 and BT treated with ORT showed 82.01% and 99.09% respectively. As there was no sporocidal effect we tested ORT at 112.5mg/m³ for 40 minutes and ORT showed a sporicidal effect for BT but not for BV2 (93.5%). The determination of Minimum Bactericidal Concentration (MBC) was carried out allowing the assessment of the sporicidal effect. The MBC of ORT was 11.25 mg/mL and 5.62 mg/mL for BV2 and BT spores, respectively. The MBC of AG was 15.62 mg/mL and 7.81 mg/mL (dose for glutaraldehyde in the solution) for BV2 and BT spores, respectively. Since is difficult associate the MBC in liquid and gas media, we tested the previous dose of ORT and a greater dose of AG (50mg/mL) overnight. FOR was used as control by dry fumigation. AG reduced all strains, but the sporicidal effect was limited. ORT was sporicidal to BT and BV1 but not to BC and BV2. The formaldehyde showed better performance, but the sporicidal effect was not complete for BV2. Our results show that high-performance disinfectants have sporicidal effects strain-dependent. ORT and AG can be safely used in disinfection processes in laboratories, industries or hospitals as a substitute for toxics disinfectants such as FOR.

**Keywords:** orthophthalaldehyde, glutaraldehyde, formaldehyde



Title	Portable and sustainable biophotonic platform coupled with artificial intelligence algorithms for salivary detection of Chikungunya virus infection in a mouse model
Authors	Marco Guevara-Vega <sup>1</sup> Rafael Borges Rosa <sup>2</sup> Douglas Carvalho Caixeta <sup>1</sup> Mariana Araújo Costa <sup>1</sup> Rayany Cristina de Souza <sup>1</sup> Giulia Magalhães Ferreira <sup>3</sup> Anagê Calixto Mundim Filho <sup>4</sup> Murillo Guimarães Carneiro <sup>4</sup> Ana Carolina Gomes Jardim <sup>3</sup> Robinson Sabino-Silva <sup>1</sup>
	1 - Innovation Center in Salivary Diagnostic and Nanobiotechnology,
Affiliations	Department of Physiology, Institute of Biomedical Sciences, Federal University of Uberlandia, Minas Gerais, Brazil.  2 - Rodents Animal Facilities Complex, Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil & Department of Virology, Aggeu Magalhães Institute (IAM), Oswaldo Cruz Foundation (Fiocruz), Recife, Brazil.  3 - Laboratory of Antiviral Research, Institute of Biomedical Science, Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil.  4 - Faculty of Computing, Federal University of Uberlandia, Minas Gerais, Brazil.
Session	34 Virologia/Microbiologia

# Abstract and Keywords

The detection of Chikungunya Virus (CHIKV) currently relies on an invasive and expensive molecular biology procedure, considered the gold standard diagnostic approach. In this sense, there is a compelling need to explore alternative methodologies that are non-invasive, cost-effective, reagent-free, and sustainable for the detection of CHIKV infection, with paramount implications for public health. The portable Fourier-transform infrared coupled with the Attenuated Total Reflection (ATR-FTIR) platform is a reliable alternative in discriminating systemic diseases using saliva. However, the application of salivary diagnostics in the context of viral diseases remains underexplored. The present study aimed to identify unique vibrational modes of salivary infrared profiles to detect CHIKV infection using chemometrics and artificial intelligence algorithms. The procedures were approved by the Ethics Committee for Animal Research at UFU (License CEUA-UFU#023/2021). To achieve this, we subcutaneous challenge interferon-gamma gene knockout C57/BL6 mice with CHIKV (20  $\mu$ l, 1 X 10<sup>5</sup> PFU/ml, n=6) and vehicle (20  $\mu$ l, n=7). Saliva and serum samples were judiciously collected on day 3, strategically chosen to coincide with



the peak of viremia. The confirmation of CHIKV infection in the serum of CHIKV-infected mice through Real-time PCR. For this purpose, the best artificial intelligence strategy was the support vector machine (SVM) algorithm for pattern classification obtaining a sensitivity of 83%, specificity of 86%, and an accuracy of 85% for CHIKV detection in saliva. These findings strongly suggest that the salivary ATR-FTIR platform holds the potential to discriminate CHIKV infection and emerge as a non-invasive, sustainable, and cost-effective tool for the detection of this emerging disease. This breakthrough not only addresses the current challenges associated with CHIKV detection but also opens doors for potential applications in the diagnosis of other viral diseases, marking a significant advancement in the landscape of diagnostics.

**Keywords:** ATR-FTIR; Machine learning algorithms; Chikungunya virus; Salivary diagnosis.



Title	Molecular Detection of <i>Ehrlichia</i> spp. in ticks from Cerrado
Authors	Maria Tereza Resende de Oliveira <sup>1,2</sup> Mayara Garcia Polli <sup>1,3</sup> Jonny Yokosawa <sup>1</sup>
Affiliations	<ul> <li>1 - Laboratory of Microorganisms of Cerrado (Brazilian Savannah), Department of Microbiology, Instituto de Ciências Biomédicas, Universidade Federal de Uberlândia, Uberlândia, Brazil.</li> <li>2 - Nursing course, Faculdade de Medicina, Universidade Federal de Uberlândia, Uberlândia, Brazil.</li> <li>3 - Post-Graduation Program in Applied Immunology and Parasitology, Instituto de Ciências Biomédicas, Universidade Federal de Uberlândia, Uberlândia, Brazil.</li> </ul>
Session	36 - Virologia/Microbiologia

### Abstract and Keywords

Ticks are hematophagous ectoparasites known to carry viruses and bacteria, which can be transmitted to vertebrate hosts during blood feeding. Several of the transmitted bacteria, including those of the genus Ehrlichia, may cause serious diseases or may even be fatal to humans and to other vertebrates. Examples of the diseases caused by Ehrlichia are human monocytic ehrlichiosis, human granulocytic ehrlichiosis and canine ehrlichiosis. In addition, there has been evidences of human ehrlichiosis in Brazil since 2001, although there are limited studies. With all this in mind, this study aimed to investigate the presence of Ehrlichia spp. in ticks collected in the Cerrado Biome (Brazilian Savannah)., using the Polymerase Chain Reaction (PCR). , but has also been reported to parasitize humans. For PCR, primers based on scientific articles were modified in an attempt to increase the range of detection of dsb gene sequences of different species of Ehrlichia. Amblyomma sculptum and Amblyomma dubitatum ticks were collected from wild dogs (Cerdocyon thous) from the region of Cumari, Goiás, midwestern Brazil, and 18 DNA samples were tested. Seven of the DNA samples tested showed amplification of the expected size, four from A. sculptum (28.6%, 4/14) and three from A. dubitatum (75%, 3/4). The intensity of the DNA bands in the electrophoresis gel was weak and, to improve the amount of the products formed, PCR conditions are going to be modified. Subsequently, nucleotide sequencing are going to be carried out for confirmation of the amplified sequences. Additional samples are also going to be tested.

**Keywords:** *Erlichia* spp, Human monocytic ehrlichiosis, Canine ehrlichiosis, Ticks, Polymerase chain reaction.



Title	A mouse model for Mayaro virus infection
Authors	Rafael Borges Rosa <sup>1</sup> ; Emilene Ferreira de Castro <sup>2</sup> ; Débora de Oliveira Santos <sup>3</sup> ; Anaíra Ribeiro Guedes Fonseca Costa <sup>3</sup> ; Willyenne Marília Dantas <sup>1</sup> ; Camila Ayumi Tanaka <sup>4</sup> ; Renata Pessôa Germano Mendes <sup>1</sup> ; Ronaldo Celerino da Silva <sup>1</sup> ; Cláudio Antônio de Moura1 Pereira <sup>1</sup> ; João Paulo Silva Servato <sup>5</sup> ; Lindomar José Pena <sup>1</sup>
Affiliations	<ol> <li>Department of Virology and Experimental Therapy (LAVITE), Aggeu Magalhães Institute (IAM), Oswaldo Cruz Foundation (Fiocruz), Recife, Brazil.</li> <li>Faculty of Medicine, Federal University of Uberlandia (UFU), Uberlandia, Brazil.</li> <li>Department of Oral and Maxillofacial Pathology, Federal University of Uberlândia, Uberlândia, Brazil</li> <li>Clinical Hospital, Faculty of Medicine, University of São Paulo (FMUSP), São Paulo, Brazil.</li> <li>Department of Clinical Dentistry, University of Uberaba, Uberaba, Minas Gerais, Brazil</li> </ol>
Session	Panel Session

# and Keywords

Mayaro virus (MAYV) infection is an emerging arbovirus disease that causes nonspecific febrile illness or arthralgia syndromes. Animal models can help clarify the pathogenesis of disease and contribute to the advancement of prevention and treatment. Thus, we carried out the characterization of a new murine model for investigations with the MAYV (MT/SINOP/210/2011, GenBank: KF305672.1) (Protocol CEUA 124/2021). Female (F) and male (M) mice wild-type and knockouts for the IFN type I route were inoculated with the MAYV (experimental groups= MAYVF WT/MAYVM WT/ MAYVF KO/ MAYVM KO, n=6/13 per group) or PBS (control groups, n=6/13 per group) subcutaneously in the footpad region(right). In general, the animals felt pain, lordosis, lethargy, bristly hair, ears placed back and eyes closed with eye irritation. After 3 and 6 days of infection (d.p.i) evaluated the presence of mechanical sensitivity (electronic von Frey method) that was better evidenced in MAYVM KO mice (p<0.0001, 3 and 6 d.p.i) and MAYVF WT and KO (p<0.0001, 3 and 6 d.p.i.) that the controls groups; assessment of muscle strength (Kondziella method) demonstrate loss of muscle strength was evidenced only in MAYVF KO (p<0.05, 3 and 6 d.p.i.); histopathological changes in the tibiotarsal



joints area demonstrated that MAYV WT and MAYV KO mice have an inflammatory response, more pronounced in knockout mice at 3 d.p.i. The presence of plantar edema was monitored for 10 days and was evident in KO mice on all days post-infection (p<0.0001). The viral load test demonstrated a systemic spread of the virus in the organs, muscles, joints and serum, with the muscle and joints being the largest viral loads found. When comparing genetic status, viral load was significantly higher in knockout animals (Joint (p<0.0001) and muscle (p<0.05), 3 d.p.i.). We demonstrated that mice knockouts to the IFN-I pathway act as an appropriate experimental model showing phenotypes like the symptoms of Mayaro fever in humans.

Keywords: Mayaro virus; Animal model; Arbovirus;



Title	Environmentally sustainable approach of Brazilian propolis through in vitro biological applications against phytopathogen
	Gabriel Guimarães Calefi¹
	Nagela Bernadelli Sousa Silva¹
	Jairo Kenupp Bastos <sup>2</sup>
Authors	Sergio Ricardo Ambrósio <sup>3</sup>
	Rodrigo Cássio Sola Veneziani <sup>3</sup>
	Robinson Sabino da Silva <sup>4</sup>
	Carlos Henrique Gomes Martins <sup>1</sup>
	1 - Laboratory of Antimicrobial Testing, Institute of Biomedical Sciences, Federal
	University of Uberlândia, Uberlândia, Brazil.
	2 – Faculty of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo,
	Ribeirão Preto, Brazil.
	3 - Nucleus of Research in Sciences and Technology, University of Franca, Franca,
Affiliations	Brazil.
	4- Innovation Center in Salivary Diagnostic and Nanobiotechnology, Department
	of Physiology, Institute of Biomedical Sciences, Federal University of Uberlandia,
	Uberlândia, Brazil
Session	36 - Virologia/Microbiologia

# Abstract and Keywords

Phytopathogens infect plants and croplands, causing worldwide economic losses every year. Furthermore, phytopathogens biofilm formation makes it harder to control and treat bacteria in agricultural lands. In this sense, Brazilian Green Propolis (BGP), Brazilian Brown Propolis (BBP) and Brazilian Red Propolis (BRP) becomes an option against phytopathogens due to its numerous biological properties in an approach not threatening to the environment. Thus, this study aimed to evaluate the potential antibacterial and antibiofilm of the of Brazilians propolis against phytopathogens. The crude extracts of Brazilian propolis were evaluated against phytopathogens Pantoea ananatis, isolated from corn leaf; Ralstonia solanacearum and Pectobacterium carotovorum isolated from potatoes and Burkholderia cepacia from onion bulb at Minas Gerais and Goiás, Brazil, using broth microdilution technique to determine the Minimal inhibitory concentration (MIC) at concentrations ranging from 0.00098 mg/mL to 2 mg/mL. Biofilm inhibition was evaluated, with the strain that demonstrated the lowest value of MIC, by quantifying biomass (Violet crystal technique) through the determination of the Minimum Inhibitory Concentration of Biofilm (MICBso), and by assessing cell viability through the counting of colony forming units (CFU/mL). The extracts demonstrated values of MIC that varied from 1 mg/mL to 2 mg/mL. Besides that, the BRP showed an MICBso of 2 mg/mL for R. solanacearum and 1 mg/mL for P. ananatis. The lowest number of viable cells was observed in the concentration at 2 mg/mL (6 and 8 Log10CFU/mL for R. solenacearum and P. ananantis, respectively). Thus, Brazilian propolis showed antibacterial activity and inhibited biofilm formation against all evaluated strains. Therefore, Brazilian propolis extracts may be a promising alternative for controlling phytopathogenic bacteria in agriculture.

**Keywords:** Antibiofilm activity, Antimicrobial Activity, Brazilian propolis, Phytopathogens.



Title	Identification of the antimicrobial resistance mechanisms and virulence factors of <i>Bradyrhizobium sp.</i> prophages
Authors	Matheus Cesar Rodrigues Garcia <sup>1</sup> ; Hugo Martins Correia <sup>2</sup> ; Richard Costa Polveiro <sup>34</sup> ; Flávio Tetsuo Sassaki <sup>1</sup>
Affiliations	<ul> <li>1 - Institute of Biotechnology, Universidade Federal de Uberlândia, Uberlândia, MG, Brazil</li> <li>2 - Universidade Estadual De Campinas, São Paulo, SP, Brazil</li> <li>3 - Laboratory of Bacterial Diseases, Sector of Preventive Veterinary Medicine and Public Health, Department of Veterinary Medicine, Universidade Federal de Viçosa, Viçosa, MG, Brazil.</li> <li>4 - Laboratory of Biotechnology in Experimental Models, Uberlândia, Universidade Federal de Uberlândia, MG, Brazil</li> </ul>
Session	36 – Virologia/Microbiologia

#### Abstract and Keywords

Bacteria of the genus Bradyrhizobium sp. are crucial for their ecological role in nitrogen fixation by plants, making them significant agro-economic and environmental interests. Bacteriophages are viruses that infect bacteria and can integrate into bacterial genomes, influencing the evolutionary process of bacteria. Additionally, they act as horizontal transmitters of genes, including those related to antimicrobial resistance and virulence factors. Our objective was to identify bacterial resistance and virulence genes in prophage genomes obtained from Bradyrhizobium sp. genomes. Bacterial genomes were collected and selected from the NCBI Assembly database, and prophages were identified using Virsorter2/2.2.3 and Checkv/1.0.1 software. To characterize the bacterial and prophage resistome, ABRICATE/1.0.0 software was utilized with all available databases. Additionally, individual genome testing was performed using the online software CARD-RGI/6.0.3 with database version 3.2.8. For virulence factor studies, PathoFact/1.0 software was employed, with a specific focus on the Virulence Factor Database (VFDB). In 54 Bradyrhizobium sp. genomes, we identified 21 viral regions of high completeness. Genes related to resistance, such as clbA (associated with antimicrobials like lincosamide, streptogramin, streptogramin A, oxazolidinone, phenicol, pleuromutilin), AAC(6')-Ib-SK, and AAC(6')-II (associated with the class of antimicrobials called aminoglycosides), were characterized using the CARD-RGI software. Furthermore, the PathoFact software annotated 120 genes related to virulence factors, of which 15 are identified as secreted virulence factors and 105 as non-secreted virulence factors. Therefore, prophages may play a key role in the diversity of Bradyrhizobium genomes and in carrying genes associated with antimicrobial

**Keywords:** Bacteriophages; *Bradyrhizobium*, nitrogen fixation, antimicrobial resistance



Title	In vitro activity of green, red and brown Brazilian propolis against Xylella fastidiosa
Authors	Andreia Marques de Oliveira <sup>1</sup> Nagela Bernardelli Sousa Silva <sup>1</sup> Jairo Kenupp Bastos <sup>2</sup> Sérgio Ricardo Ambrósio <sup>3</sup> Rodrigo Cássio Sola Veneziani <sup>3</sup> Carlos Henrique Gomes Martins <sup>1</sup>
Affiliations	Federal University of Uberlândia, Uberlândia, Brazil <sup>1</sup> Federal University of São Paulo <sup>2</sup> University of Franca <sup>3</sup>
Session	34- Virologia/Microbiologia

## Abstract and Keywords

Xylella fastidiosa is a phytopathogen that affects economically important agricultural crops, such as orange and lemon trees. One of the most alarming problems caused by this bacteria in Brazil is Citrus Variegated Chlorosis (CVC), which develops in citrus crops through the formation of biofilm in the plant's xylem vessels, reducing its nutrition. Brazilian propolis has already demonstrated potential antibacterial activity against various microorganisms. Therefore, the objective of this study was to evaluate the antibacterial and antibiofilm activity of green, red, and brown propolis against Xylella fastidiosa (9a5c). The crude extract of green, red, and brown propolis was evaluated against X. fastidiosa, the strain of which was collected from branches of orange trees affected by CVC in Macaubal (São Paulo, Brazil), using the broth microdilution technique to determine the Minimum Inhibitory Concentration (MIC), at concentrations ranging from 2000 to 0.98 µg/mL. The Minimum Bactericidal Concentration (MBC) was determined to evaluate whether the propolis extracts tested in the study exerted bactericidal or bacteriostatic activity against the studied bacterium. The antibiofilm activity was assessed using 2% crystal violet for the determination of the Minimum Inhibitory Concentration of biofilm (MICB50) and microorganism counting. The MIC and MBC values for all samples were 2000 μg/mL, demonstrating bactericidal activity against the evaluated strain. In the biofilm evaluation, the MICB50 of green propolis was 15.62 µg/mL, red propolis was 2000 μg/mL, and brown propolis was 62.5 μg/mL. Considering the issues caused by Xylella fastidiosa and the results obtained in this study, Brazilian green, red, and brown propolis show promising antibacterial and antibiofilm activity, promising to improve the quality of plantations affected by this bacterium and potentially leading to the control of CVC incidence in the future.

**Keywords:** Antibacterial activity, antibiofilm activity, Brazilian propolis, citrus variegated chlorosis, *Xylella fastidiosa* 



	A lectin from Luetzelburgia auriculata (LAA) effectively inhibits
Title	infection of SARS-CoV-2 variants of concerns
Authors	Leandro Peixoto Ferreira de Souza¹ Victória Riquena Grosche²,³ Marco Guevara-Veja¹ Tamara Carvalho³ Romério Rodrigues dos Santos Silva⁴ Karla Lilian Rodrigues Batista⁵ Rodrigo Paolo Flores Abuna João Santana Silva⁶,7 Marília de Freitas Calmon³ Paula Rahal³ Luis Cláudio Nascimento da Silva⁵ Bruno Silva Andrade® Claudener Souza Teixeira⁴ Ana Carolina Gomes Jardim²,³ Robinson Sabino-Silva¹
Affiliations	1– Innovation Center in Salivary Diagnostic and Nanobiotechnology, Institute of Biomedical Science (ICBIM), Federal University of Uberlândia (UFU), Uberlândia 38405-317, Brazil 2– Laboratory of Antiviral Research, Institute of Biomedical Science (ICBIM), Federal University of Uberlândia (UFU), Uberlândia 38405-317, Brazil 3- Institute of Biosciences, Languages, and Exact Sciences (Ibilce), São Paulo State University (Unesp), São José do Rio Preto 15054-000, Brazil 4- Center of Agrarian Science and Biodiversity, Federal University of Cariri (UFCA), Crato 63130-025, Brazil 5- Laboratory of Microbial Pathogenesis, CEUMA University, São Luís 65045-380, Brazil 6- Department of Biochemistry and Immunology, Ribeirão Preto Medical School, University of São Paulo,Ribeirão Preto 14049-900, Brazil 7- Oswaldo Cruz Foundation (Fiocruz), Bi-Institutional Platform for Translational Medicine,Ribeirão Preto 14049-900, Brazil 8- Laboratory of Bioinformatics and Computational Chemistry, State University
Session	Virology/Microbiology

# Abstract and Keywords

The year 2020 will always be remembered for the start of the COVID-19 pandemic. The development of safe and effective new therapies against severe acute respiratory syndrome-coronavirus 2 (SARS-CoV-2) still is necessary. The vaccines was a major turning point in the fight against the COVID-19 pandemic, however some mutations forms of SARS-CoV-2 had enhanced rates of transmission, adaptation to antibody neutralization and reduction performance of vaccines. In late 2020, the emergence variants of concern (VOCs) that posed an increased risk to global public health prompted Word Health Organization (WHO), here we present a new biotechnological tool against tow special VOCs, Gamma and Omicron. Our data demonstrated the capacity of galactose/lactose legume lectin Luetzelburgia auriculata (LAA) blockade SARS-CoV-2 VOCs cell entry. The LAA reduce more than 90% of virus infectivity in Vero E6 cells and presents low levels of cytotoxicity. We confirm that LAA virus neutralization effect occurs after virus attachment and different moments of virus infection. Even after the entry of the virus into the cells, LAA proved to be effective in post-infection test. Our findings suggest that LAA lectin could be in the future a alternative tool against SARS-CoV-2 VOCs.

Keywords: Lectin, SARS-CoV-2, lectin from Luetzelburgia auriculata (LAA),



Title	Anatomical description of the heart of the puma (Puma concolor, Linnaeus 1771)
Authors	Cassiane Júlia Machado da Silva <sup>1</sup> ; Geovanna Gonçalves Ribeiro <sup>2</sup> ; Marina Fernandes Arbués Mota <sup>2</sup> ; Carolina Freitas da Silva <sup>2</sup> ; Matheus da Costa Matutino Martins <sup>2</sup> ; Zenon Silva <sup>1</sup> ; Juan Fernando Vélez García <sup>3</sup> ; Deusmar Machado da Silva <sup>3</sup> ; Klayton Marcelino de Paula <sup>2</sup> ; Roseâmely Angélica de Carvalho Barros <sup>2</sup> ; Romeu Paulo Martins Silva <sup>2</sup>
Affiliations	<ul> <li>1-Federal Institute Goiano, Professionalizing Secondary Education, Catalão, GO, Brazil.</li> <li>2-Federal University of Catalão (UFCAT), Institute of Biotechnology, Department of Biological Sciences, Laboratory of Comparative Anatomy of Wild Animals, Catalão-GO, Brazil.</li> <li>3-Universidade del Tolima, Veterinary Medicine, Ibague, Colombia</li> </ul>
Session	FeSBE Jovem

#### Abstract and Keywords

The heart is a vital organ in the vertebrate organism and the analysis of its morphology is capable of offering information about the physiology and adaptation of a species to the environment. The study of the anatomy of the heart of the Puma (Puma concolor) can provide valuable information for understanding the anatomy of felines, in general, and guide conservation measures for the species. Aiming to describe the anatomy of the Puma heart (PUMA CONCOLOR, LINNAEUS 1771). The methodology used was the dissection and analysis of two Puma concolor hearts, taken from carcasses donated by the Wild Fauna Research Institute (IPEVIS) and the Wild Animal Triage Center (CETAS) of Catalão (GO), (SISBIO 37072 - two). Opinion of the ethics committee on the use of animals/CEUA-UFCAT Protocol 22/01. To fix the material, a 10% aqueous solution of formaldehyde was used via perfusion and the vascular system was stained with latex. Macroscopic analysis was performed accurately and photodocumented. The results found in these studies were that the hearts of the specimens have an elongated shape, measuring approximately 8.0 cm in length and 5.5 cm in the transverse direction; They are located in the mediastinum of the thorax and are surrounded by the pericardium. The right ventrolateral and left lateral dorsal surfaces produce right and left cardiac images in the lungs. In conclusion, the results obtained allowed a greater understanding of the cardiac morphology of the species, and we hope that they will serve as a basis for research into its biology and ecology

**Keywords:** Anatomy; heart; Puma concolor.



Title	Antioxidant potential and effect on LDL oxidation of aqueous and n-Butanolic extracts from Cagaiteira leaves ( <i>Eugenia dysenterica</i> DC.)
Authors	Juliana Mateus Gomes Vidigal <sup>1</sup> Raylla Emanuelle de Jesus Silva <sup>1</sup> Renato Gois Sossa <sup>1</sup> , Françoise Vasconcelos Botelho <sup>2</sup> .
Affiliations	1 – Escola Estadual Américo René Giannetti, Uberlândia, Brazil 2 – Instituto de Biotecnologia, Universidade Federal de Uberlândia, Brazil
Session	FeSBE Jovem

# and Keywords

Atherosclerosis is a chronic inflammatory condition characterized by oxidative modifications in low-density lipoproteins (LDL), contributing to the development of atherosclerotic lesions. Plant-derived bioactive compounds have been investigated for their potential role in preventing and mitigating atherosclerosis. This study aimed to assess the antioxidant potential and impact on LDL oxidation of aqueous and n-butanolic extracts from cagaiteira leaves (Eugenia dysenterica DC.). Total phenolic compounds were quantified using the Folin-Ciocalteu reagent, flavonoids using the aluminum chloride, and the antioxidant potential was evaluated by the ability to reduce Fe<sup>3+</sup> to Fe<sup>2+</sup> (FRAP assay), all by spectrophometry. LDL oxidation was monitored by measuring conjugated diene formation at 234 nm and quantifying LDL lipid peroxidation using the thiobarbituric acid-reactive substances (TBARS) method. All experiments were conducted in triplicate. Results showed that both extracts exhibited similar levels of total phenolic compounds, flavonoids, and antioxidant potential. The mean content of total phenolics for the aqueous and n-butanolic extracts was 0.189 mg.g-1 and 0.162 mg.g-1 gallic acid equivalent, respectively, with antioxidant potential exceeding 500 µmol trolox eq.g-1 for both extracts. However, regarding the effect on LDL oxidation, no significant differences were observed among the various concentrations tested, and LDL oxidation inhibition was modest compared to the positive control (quercetin 2.0 µg/mL). These findings suggest that aqueous and n-butanolic extracts from cagaiteira leaves possess antioxidant potential, but their impact on LDL oxidation warrants further investigation for conclusive insights.

**Keywords:** Atherosclerosis, Low-density lipoproteins (LDL), Antioxidant potential, *Eugenia dysenterica* DC.



Title	Effect of polyphenols from Cagaiteira leaves ( <i>Eugenia dysenterica</i> DC.) on low-density lipoprotein (LDL) oxidation <i>in vitro</i>
Authors	Renato Gois Sossa <sup>1</sup> ,
	Juliana Mateus Gomes Vidigal <sup>1</sup> ,
	Raylla Emanuelle de Jesus Silva <sup>1</sup> ,
	Françoise Vasconcelos Botelho <sup>2</sup> .
Affiliations	1 – Escola Estadual Américo René Giannetti, Uberlândia, Brazil
	2 - Instituto de Biotecnologia, Universidade Federal de Uberlândia, Brazil
Session	FeSBE Jovem

# Abstract and Keywords

Atherosclerosis, a chronic inflammatory disease, involves modifications in lowdensity lipoproteins (LDL), contributing to atherosclerotic lesions. Studies show that bioactive compounds present in plants can be important allies in the prevention and progression of atherosclerosis. Cagaiteira (Eugenia dysenterica DC.) contains antioxidants, but the impact of its ethyl acetate fraction on LDL oxidation remains unexplored. The study aims to quantify the total phenolic compounds and flavonoids of the ethyl acetate fraction of cagaiteira leaves (Eugenia dysenterica DC.); evaluate the antioxidant potential and the effect on LDL oxidation in vitro. We quantified the content of phenols and flavonoids through colorimetric assays. Antioxidant capacity was assessed by the potential to reduce  $Fe^{3+}$  to  $Fe^{2+}$  (FRAP), also by spectrophotometric assay. LDL oxidation was monitored by the formation of conjugated dienes at 234 nm and the quantification of LDL lipid peroxidation by the TBARS method with different concentrations (0.5, 1.0, and 2.0  $\mu$ g/mL) of the ethyl acetate fraction. All experiments were conducted in triplicate. The ethyl acetate fraction of cagaiteira leaves had an average concentration of 1.47 mg.g-1 of gallic acid equivalent and 1.09 mg/mL flavonoids. Such content of antioxidant compounds reflected in a high antioxidant capacity, FRAP >1200 µmol trolox eq.g-1, and the different concentrations tested (0.5, 1.0, and 2.0 µg/mL of the extract significantly reduced LDL oxidation in vitro and the production of thiobarbituric acid-reactive species. The results indicate that the ethyl acetate fraction of cagaiteira leaves has a strong antioxidant potential, may play a protective role in atherosclerosis, and further studies need to be conducted to evaluate this protective effect.

**Keywords:** Atherosclerosis, polyphenols, cagaiteira leaves (*Eugenia Dysenterica* DC.)



Title	Analysis of scorpionism incidence in urban sectors: A Study from 2020 to 2022 in Uberlândia-MG
Authors	Ronan Marques Teixeira¹; João Paulo Ferreira Campos¹; Adalberto Teixeira de Matta Flora Neto¹; Fábio Tonissi Moroni¹; Juliana Junqueira da Silva²; Raquel Borges-Moroni³; José Fausto de Morais⁴; Elton Nunes Britto⁵.
Affiliations	<ul> <li>Faculdade de Medicina, Universidade Federal de Uberlândia, Uberlândia, Brasil.</li> <li>Centro de Controle de Zoonoses, Prefeitura Municipal de Uberlândia-Uberlândia, Uberlândia, Brasil.</li> <li>Instituto de Ciências Biomédicas, Universidade Federal de Uberlândia, Uberlândia, Brasil.</li> <li>Faculdade de Matemática, Universidade Federal de Uberlândia, Uberlândia, Brasil.</li> <li>Instituto Federal de Educação, Ciência e Tecnologia, Santarém, Brasil.</li> </ul>
Session	38 - Epidemiologia.

#### Abstract and Keywords

Scorpions are the main etiological agent of accidents caused by venomous and poisonous animals in Brazil. Consequently, preventing scorpion incidents in cities is important for public health. The objective of this study is to assess the sectorspecific occurrence of such poisonings in the municipality of Uberlândia between 2020 and 2022. In this context, submission to the Ethics Committee is not required, according Brazilian law. The study was descriptive, retrospective, and based on secondary data from the Health Department of the Municipal Government of Uberlândia. Data related to monthly notifications and various city sectors (central, north, south, east, west, rural zone) were analyzed. Statistical data analysis was conducted using R software and based on a significance level of 5%. There was a difference in the number of occurrences in the years 2020, 2021, and 2022, with 2392, 2050, and 3213 registered occurrences, respectively. The months of January (2020), November (2021), and July (2022) recorded the highest incidents. Concerning sector distribution in 2020, the central region had the significant difference (p<0.05), averaging 53.5 cases/month. In 2021, the average decreased by 20.93% (42.33 cases/month). In 2022, there was an increase of 57.48% (73.58 cases/month). The sector with the lowest incidence in 2020 and 2021 was the West, with averages of 32.25 and 28.08 cases, respectively. In 2022, the rural sector had the lowest record, followed by the South and West, with averages of 0.92, 40.25, and 41.17, respectively. Consequently, these data may indicate the necessity for epidemiological studies, coupled with georeferencing technologies, to identify priority areas and months for conducting campaigns to prevent scorpion-related accidents in the municipality of Uberlândia.

**Keywords:** scorpion accidents, scorpionism, epidemiology, toxicology.



Title	Emerging challenges: Multidrug resistance in neonatal infections by gram-negative bacteria
Authors	Mallu Santos Mendonça Lopes¹; Isadora Caixeta da Silveira Ferreira²; Izabella Clara de Brito Machado¹; Thiago Alves de Jesus¹; Ralciane de Paula Menezes³; Denise Von Dolinger de Brito Röder⁴.
	1 - Undergraduate Course in Biomedicine, Institute of Biomedical Sciences, Federal University of Uberlândia, Minas Gerais, Brazil.
	2 - Postgraduate Program in Health Sciences, Faculty of Medicine, Federal University of Uberlândia, Minas Gerais, Brazil.
Affiliations	3 - Technical Course in Clinical Analysis, Technical School of Health, Federal University of Uberlândia, Minas Gerais, Brazil.
	4 - Department of Microbiology, Institute of Biomedical Sciences, Federal University of Uberlândia, Minas Gerais, Brazil.
Session	Epidemiological

# Abstract and Keywords

The growing increase in bloodstream infections (BSI) by multidrug-resistant Gram-negative bacteria (MDR) has a negative impact on morbimortality in Neonatal Intensive Care Units (NICU), representing a critical and urgent public health challenge. We therefore sought to analyze the incidence and lethality of BSI-GNB in a Brazilian NICU. This is a retrospective cohort study, approved by the Research Ethics Committee (No. 2.173.884/2017), and conducted through the medical records of neonates admitted to the NICU between January 2019 and June 2022. Epidemiological surveillance was conducted daily using forms from the National Healthcare Safety Network. Of 208 BSI, 43 (20.7%) were attributed to GNB, distributed over the years as follows: 2019 (7; 20.6%), 2020 (5; 14.7%), 2021 (10; 29.4%) and 2022 (12; 35.3%). Among the GNB isolates, 9 (20.93%) were MDR, with 6 (66.67%) detected in 2021 and 3 (33.33%) in 2022. Of the GNB-MDR, 6 (66.7%) were Klebsiella pneumoniae, 2 (22.2%) Escherichia coli and 1 (11.1%) Enterobacter cloacae. With regard to resistance, 6 (66.7%) of the GNB-MDR were ESBL. The lethality rate among neonates with BSI-GBN was 20% in 2019, 13.3% in 2020, 20.8% in 2021 and 12.3% in 2022. The growing incidence of BSI-GNB MDR in the NICU in recent years is alarming and presents challenges due to the therapeutic difficulties and the associated high lethality rate. It is therefore crucial to implement comprehensive prevention and control strategies, with continuous epidemiological surveillance, and to adopt preventive approaches, including the promotion of rational antimicrobial use. Keywords: Drug Resistance; Gram-Negative Bacteria; Infection Control; Neonate.



Title	COVID-19: hospitalization and the impact of social determinants in the elder mortality
	Victor Antônio Ferreira Freire <sup>1</sup>
Authors	Gustavo Siconello dos Santos <sup>2</sup>
	Luciana Karen Calábria <sup>3</sup>
	Alexandre Azenha Alves de Rezende <sup>3</sup>
	1 – Federal University of Uberlândia, Uberlândia-MG, Brazil.
Affiliations	2 – Amaral Carvalho Hospital Foundation, Jaú-SP, Brazil.
	3 – Federal University of Uberlândia, Ituiutaba-MG, Brazil.
Session	Epidemiology

# and Keywords

COVID-19-related social determinants such as age, occupation, educational attainment, race, housing status and food security, could have an underestimated effect in mortality. This is further worsened in the presence of chronic diseases. We evaluated the association between social determinants, clinical variables and the mortality of the elder patients hospitalized due to SARS-CoV-2 between 1<sup>st</sup> and 39<sup>th</sup> epidemiological weeks of 2021 in Ituiutaba-MG. Data collected between January and October 2021 (n=425) in the municipality's health service. The frequencies of positive COVID-19 cases and mortality were estimated according to age, sex, occupation, educational attainment, skin color categories, noncommunicable diseases, time of hospitalization and intensive care (ICU). The association between categorical and metric variables were analyzed by Chi-square and Mann-Whitney tests, respectively. Prevalence ratio was obtained using Poisson regression with robust variance and 95% confidence interval (CAAE 56051721.2.0000.5152). Patients with advanced age (70-79 years) (n=78, 33.5%, p<0.01), diabetes mellitus (DM) (n=65, 42.8%, p=0.01), cardiovascular diseases (n=113, 72.9%, p=0.05), total time of hospitalization (median=10; IQR=11, in days) were associated to mortality. The death probability increased 2% (PR adjusted: 1.01, 95% CI 1.01-1.02) for each year of life, and self-reported worker (PR adjusted: 1.25, 95% CI 1.02-1.51), DM (PR adjusted: 1.30, 95% CI 1.05-1.61) and total time of ICU hospitalization (PR adjusted: 1.03, 95% CI 1.01-1.04) were associated with higher death prevalence for COVID-19. The death prevalence was lower to self-reported housewife and/or husband or unemployed (PR adjusted: 0.63, 95% CI 0.39-0.96). In addition to age, the elder who works, diabetics, and higher time of ICU hospitalization contributes to mortality. These results reveal a complex challenge for the public healthcare due to social inequalities, reverberating in the outcome of the illness.

Keywords: SARS-CoV-2; Public healthcare; Mortality.