8: Others

8.01 Occurrence of lymphangitis in serum-producing horses on São Joaquim farm of Instituto Butantan

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Introduction: Lymphangitis is an inflammatory process of the lymph vessels and lymph nodes that affect the limbs. It has a cosmopolitan distribution and has no predilections for age, breed or sex. This inflammation normally happens after bacteria and fungi have entered the body through a cut or wound distal to the limbs. There have been some reports of protein imbalance as a predisposing factor for its occurrence. It is more common in the hind limb especially in the distal hock-joint region. Inflammation occurs in lymph vessels with their total or partial obstruction, making it difficult or impossible to drain lymph fluid from the affected limb. The limb exhibits an increase in volume and temperature, pain and wounds that discharge purulent material. A life-threatening or inadequately treated wound can become chronic leading to fibrosis. **Objectives:** The aim of this investigation was to determine what happened with serum-producing horses. **Results and Discussion:** Two serum-producing horses, one belonging to the anti-rabies group and other to the anti-elapid venom group of the São Joaquim Farm. There were male, castrated, weighing approximately 450 kg, cross breed, and had lameness of the right hind limb, swelling of the coxal to the shank, no apparent wounds, sensitivity on palpation, hyperthermia (39 °C) and tachycardia (HR 50 beats / min). Lymphangitis was diagnosed. They were treated with hydrotherapy to reduce the swelling associated with administration of an anti-inflammatory and antibiotic for 28 days. After this period, the horses had resolution of symptoms and return to routine activities. This is the first reported case of lymphangitis at the São Joaquim Farm.

Supported by: Fundação Butantan.

8.02 Occurrence of *Bothrops jararaca* bite in a horse on São Joaquim farm of Instituto Butantan

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Introduction: In Brazil, 88-89% of the snake bites in human beings come from snakes of *Bothrops spp* genera. This type of accident also occurs in animals, showing more common reports in the canine species. There is considerable variation in effect of the several species of venomous snakes among domestic animals. In experiments carried out by Araujo & Belluomini (1962), horses, sheep and cattle showed a greater sensitivity to snake venom, and according to Rosenfeld (1971), carnivores seem to be more resistant to the venoms in relation to the other species. In the place of the bite a great edema occurs and spreads to other parts, and 12 h after its injection, necrosis can occur at the site of the bite. In general, blood coagulation time is increased, and because of that hemorrhages occur in body cavities and in the parenchyma of some organs, and it may happen even after appropriate therapy. In domestic animals such as horses and cattle, when the bite occurs in the limbs, the clinical signs are lameness, difficulty in locomotion, immobility, decubitus and inability to stand. Many snake bite accidents result in death. The sooner the specific serotherapy is started, the better the prognostic is for the animal's life. **Objectives:** The aim of this investigation was to determine what happened with the horse belonging to São Joaquim Farm. **Results and Discussion:** A horse, male, castrated, eight years old, showed sudden behavioral changes. An employee of the farm told that a *Bothrops jararaca* was next to him. Minutes after that, the horse showed lameness in the right front limb, edema in dorsal region of the hoof coronet, and increase in heart rate (90 beats per minute) and in respiratory movements (40 movements per minute). Coagulation test showed increased coagulation time (over 25 minutes). He was treated with six vials of Instituto Butantan specific intravenous antibothropic serum. Two hours later, he received three more vials of serum in the same way. Also, he was managed with antibiotic and anti-inflammatory for five days. After 15 days the horse had completely recovered, showing normalized vital functions being able to come back to carry out its normal function.

Supported by: Fundação Butantan.

8.03 Establishment of the comet assay for the freshwater snail *Biomphalaria glabrata* (Say, 1818)

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Introduction: The comet assay is a method developed to detect breaks in the DNA. In the test, cells are embedded in agarose and lysed by detergents in high salt and the DNA released electrophoresed. Cells with an increased frequency of DNA breaks display increased migration of DNA toward the anode. This is a promising test for studies on genotoxicity, DNA repair, environmental and human monitoring. **Objective:** This research aimed to establish the comet assay in hemocytes of *Biomphalaria glabrata* to assess the capability of this system to detect mutagenic pollutants. **Methods:** Twelve snails per group were selected (control and exposed to 60-Co gamma radiation). Snails were irradiated with single doses of 2.5, 5, 10 and 20 Gy at 2.82KGy/h. After the exposure, the hemolymph was collected and suspended in 0.5% of agarose low melting point (LMP), the mixture with low melting agarose was placed on a microscope slide previously coated with a 1.5 % normal melting agarose. The slides were placed in lysis solution of pH 10 for 600 min. After lysis, the electrophoresis tank was filled with slides and covered with the electrophoresis buffer to allow the expression of damage. The slides were kept in the buffer for 30 min, and then electrophoresis was carried out at 23V and 150mA for 30 min. After running, the cells were neutralized with buffer of pH 7.5. Next, the cells were stained with 0.002% ethidium bromide and visualized with a fluorescence microscope at 400x. The visual analysis was carried out by classifying comets in categories (0 to 3) according to the extent of DNA migration. **Results and Discussion:** The results showed a genotoxic effect of radiation at all doses in a dose-dependent manner. The comet assay in *B. glabrata* was demonstrated to be simple, fast and reliable in the evaluation of genotoxic effects of environmental mutagens.

8.04 Effect of methylecgonidine (AEME) on melatonin synthesis in the rat pineal gland

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Introduction: The pineal gland synthesizes the hormone melatonin at night induced by noradrenergic stimulation. The activation of α- and β-adrenoceptors increases cAMP, which increases activity of the enzyme arylalkylamine-N-acetyltransferase (AANAT). The gland also receives parasympathetic innervation, with acetylcholine being an inhibitor of melatonin synthesis. Methylecgonidine (AEME) is a byproduct of cocaine, when used in the form of crack. Its effects on the central nervous system are even more intense than those of cocaine itself. There is evidence showing the action of AEME on the cholinergic system, which could lead to an effect on melatonin synthesis. **Objectives**: The aim of the present work was to study the AEME effects on the melatonin synthesis in the rat pineal glands *in vitro* and *in vivo*. Also investigated was the possible cholinergic action of AEME and on the activity of AANAT. The nocturnal profiles of melatonin synthesis and of the expression of their receptors (MT1 and MT2) in the cerebellum were evaluated in *in vivo* experiments. **Methods:** Pineal glands were isolated from male Wistar rats and maintained in culture for 48 h in BGJb medium. The glands were stimulated by noradrenaline (1 μM) in combination with AEME at different concentrations (1 μM and 100 μM), that was added 30 min or 48 h before noradrenaline addition, or with 10 μM atropine. For the *in vivo* experiments, the rats were injected with AEME (2.0 mg/kg) for 7 consecutive days and then were sacrificed in order to evaluate the melatonin synthesis in the pineal gland and the expression of melatonin receptors (MT1 and MT2) in the cerebellum. Melatonin was quantified by HPLC, AANAT activity was determined by radiometric assay, and melatonin receptors were analyzed by RT-PCR. **Results and Discussion:** AEME reduced melatonin synthesis at every concentration used, independent of incubation time (Nor 1 μM = 10.56±0.61 ng/gland; Nor + AEME 1 μM = 7.56±0.69 ng/gland; Nor + AEME 100 μM = 7.96±0.78 ng/gland; Nor + AEME 100 μM (48 h) = 7.93±0.72 ng/gland). Atropine did not reverse the AEME effects on melatonin synthesis, showing that its actions seem to be independent of muscarinic cholinergic receptors. AANAT activity was not altered by AEME. The MT1 receptors in the cerebellum were increased by AEME treatment. However, no differences were observed for MT2 receptor expression. Melatonin was also reduced by AEME in the in vivo experiments, with a more marked reduction at the end of the dark period.

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8.05 A bioinformatics tool to visualize peptides found by mass spectrometry in protein structures

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Introduction: Advances in mass spectrometry (MS) and in bioinformatics tools over the past decades have made it possible to characterize proteomes and peptidomes of complex biological samples such as venoms, blood sera and cell lysates, among others. Peptides found in biological fluids may result from proteolytic processing of proteins that may be triggered by endogenous mechanisms, or even by activation of proteases during manipulation in the laboratory. In some cases, the cleaved peptides come from a protein whose 3D structure is deposited in public databases and viewing the peptide positions may clarify if the peptide is internal or external or is located near some binding site in the structure, helping to build more realistic hypothesis about the mechanism of the proteolytic processing. However, it is not uncommon that the 3D structure (PDB) of the target protein is not available, as is the case of many venom toxins, making it necessary to search for a similar protein which is expected to have a similar structure. The peptides from the target protein cannot match exactly their homologs in the similar protein so a homolog peptide search must also be performed. Last, the homolog peptides must be highlighted in the PDB. Manually performing these tasks is very time consuming, while an automated computational tool may do it in few minutes. This work shows an automated method of finding the similar protein PDB and the homolog peptides and highlighting them on a 3D plot. It is available through an Intranet web server to which MS results processed by MASCOT are uploaded, processed and the resulting PDBs are downloaded. **Objectives:** The objective of this work was to develop a bioinformatics tool to extract peptide sequences obtained by MS and by the search engine MASCOT (Matrix Science), to find the most significant match of crystallographic protein structure by similarity search, and to align the peptides and redraw the structure highlighting the identified peptides. **Methods:** MS data analysis by MASCOT is processed to retrieve the match protein and identified peptide sequences. The match protein sequence is submitted to Blastp. The most similar protein with PDB is selected. The PDB is downloaded from the RCSB-PDB repository. The primary structure of the similar protein is retrieved from the PDB file. The homolog peptides are found by a Longest Common Subsequence (LCS) algorithm using Blosum62 matrix. The PDB is edited to highlight the homolog peptides in the similar protein. 3D view is obtained using PyMol or other protein viewer. **Results and Discussion:** The program can be accessed through http://172.25.60.101 IP address. We performed runs of peptide fractions of *Bothrops cotiara* venom analyzed by MS to evaluate the functionality of the tool. The resulting spectra were analyzed by MASCOT which identified several L-amino acid oxidase (LAAO) sequences, and one *Bothrops neuwiedi pauloensis* LAAO as the best match. There is no PDB file for this enzyme. Application of the program returned chain A, LAAO from *Agkistrodon halys pallas* as the most similar enzyme with PDB file and indicated the peptides found by MS.

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8.06 Participation of neutrophils in antinociception induced by glycogen, evaluated by the paw pressure model

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Introduction: Several pieces of evidence demonstrate the involvement of neutrophils in the modulation of painful response during the acute inflammatory process. However, the participation of these cells in pain generation is still conflicting since neutrophils have been proposed to play a role in either induction or inhibition of painful response. Glycogen-induced peritonitis has been shown to induce antinociception in mice submitted to the abdominal contortion test, and secretion of calcium-binding protein S100A9 by neutrophils mediated this effect. **Objectives:** To investigate involvement of neutrophil in nociception control after administration of glycogen in rats pre-treated with fucoidan, a selectin adhesion molecule inhibitor. In addition, the leukocyte migration profile was investigated in several periods of time, in which painful response was assessed after intraplantar injection of glycogen in rats pre-treated with fucoidan. **Methods:** Male Wistar rats were injected intravenously with fucoidan (5 mg/kg, 500 μL/animal) 15 min before intraplantar injection of 5% (w/v) glycogen solution (100 μL/animal) or saline, and after different periods of time the paw pressure test was evaluated. In another experimental procedure, rats were injected with fucoidan 15 min before intraplantar injection of glycogen, and after different periods of times euthanized in order to the collect the plantar tissue for histological analysis. **Results and Discussion:** Glycogen induced antinociception in rats at 2, 4, 6, 8 and 12 h. Pre-treatment with fucoidan reversed the antinociception observed at 2, 4 and 6 h after glycogen injection and induced hyperalgesia at the same periods of time. Rats pre-treated with fucoidan and tested 8 h after glycogen injection only showed reversal of the antinociceptive effect. When the nociceptive test was evaluated 12 h after glycogen administration, the pre-treatment with fucoidan failed to diminish antinociception. Histological analysis demonstrated an increase in migration of polymorphonuclear cells between 2 and 8 h after glycogen administration, but migration of polymorphonuclear cells was inhibited in rats pre-treated with fucoidan and injected with glycogen over the initial periods of time after glycogen injection. The predominant cells accumulated in the footpad after glycogen administration were neutrophils. Pre-treatment with fucoidan, a selectin inhibitor, not only reversed glycogen-induced antinociception but also induced hyperalgesia. Besides, fucoidan reduced neutrophil migration in rat footpads after glycogen injection. Thus, glycogen induces antinociception in rats evaluated by the paw pressure model, likely by accumulation of polymorphonuclear cells, particularly neutrophils. The possible participation of S100A9, found in high concentrations in neutrophil cytosol, in this phenomenon is under investigation.

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8.07 Development of a MATLAB application for processing kinetic data of animal cell experiments

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Introduction: Experimental raw data is normally difficult to be analyzed without some processing. In general, spreadsheets like Microsoft Excel are used to do this treatment. **Objectives**: The aim of this work was to uniformize and speed up data treatment using an application developed in MATLAB. **Methods**: Starting from original data of experiments with animal cells, a MATLAB application was developed to import data from Microsoft Excel spreadsheets with these data. It is important to emphasize that the data are imported to MATLAB directly from Microsoft Excel and after the treatment the resulting data of that analysis can be exported back to an Excel spreadsheet so that the user can use them in the way he/she is used to, as in making plots. In this MATLAB application, the source data are fitted and smoothed with automatic generation of a specific growth rate plot, using a spline algorithm. The application still allows the user to make small adjustments in the experimental points. **Results and Discussion**: The use of that application results in less time spent for this treatment and also in a standardization of results. This new method, using a MATLAB application, shows better results in its analysis, besides presenting the information in a clearer way to be understood by anybody. Besides, the results are standardized and obtained in a shorter time, using a single program.

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8.08 Ethnoherpetology in the Vale do Paraíba, São Paulo State, Brazil: relationship between rural populations and the *Bothrops jararaca* species

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Introduction: The "Vale do Paraiba" is considered a geological fault sided by the "Serra do Mar" and "Serra da Mantiqueira," which contain the "caipira" culture. The prefix *Ethno*, before the name of a discipline, such as herpetology, means the relation between the man and the environment, that is, the understanding of herpetology in a community. Ethnobiological approaches occurred in Brazil starting in the 1980s, presenting themselves as a recently discussed subject in the country. The species in question is one of the most important epidemiologic agents in the area, whose genus is responsible for 90% of the ophidian accidents in Brazil's southwest and one of the most aggressive snakes of the Viperidae family. **Objective**: The objective of this study is to analyze the regional and scientific knowledge about the natural history of the *Bothrops jararaca* species in some cities in the Vale do Paraíba, in the state of São Paulo. **Methods**: The choice of the study areas follows historical logic. Lagoinha preserves the agropastoral activity derived from the period of coffee plantation in the Valley. Roseira has been clearly impacted by the construction of President Dutra Highway, with industries that are near the traditional rural properties. Taubaté is the most ancient representative in the Valley and was the departure center for exploratory expeditions**.** The research was divided into two different phases, from March to November, 2009. First, a semi-structured questionnaire was applied, with a recorded interview, containing 20 questions about ecology and biology of *B. jararaca*. Afterward, some photographs were shown so that the interviewees were able to identify some snakes in order to map possible mistakes in the recognition of the species in question. A previous survey of the snakes from the sampled area has been verified in the books of the Supplier Register of Animal Reception of Instituto Butantan, over the last 20 years. **Results and Discussion**: To date, 80% of the interview activities have been made, as well as the survey from 1988 to 2003, which will be complemented up to 2008. We have determined the addition of 29 species, up to now: 1.3% to the Boidae family; 20.8% Colubridae; 0.4% Elapidae and 77.5% Viperidae. Of all those species, 20% are represented by *B. jararaca*. The killing of those snakes seems to be an attribute of courage in the area. Knowledge about viviparity and predatory activity seems to be mysterious to some rural residents. However, seasonality and the search for shelter are more clarified, justified by the unimodal reproductive pattern and the accumulation of rubble in the surroundings of the houses, creating microhabitats for the *jararacas*, known by the inhabitants of the area as *urutu*. The *jaracussu* nomenclature may occur in the more melanic individuals. *Urutu-mestiço* classifications have also occurred in Roseira, which would be a supposed cross between the rattlesnake and the *urutu*, known popularly as *cascaverana* in Lagoinha. With the development of this methodology, we intend to have a better interaction between the rural population and the scientific community, being able using this approach to map the origins and characteristics of the traditional herpetological knowledge of this sampled area.

8.09 Effect of Piperaceae amide on schistosomiasis vector *Biomphalaria glabrata* (Say, 1818)

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Introduction: Schistosomiasis occurs in 54 countries mainly in South America, the Caribbean, Africa and east of the Mediterranean. In Brazil, 5-6 million people are infected and 30 thousand are exposed to risk of infection. One of the more efficient methods to control this disease is the application of molluscicides which eliminates or reduces the intermediate host population. Concerning environmental preservation, the high cost and recurrent resistance of snail to the synthetic molluscicide have stimulated the study of molluscicides of plant origin. The species from the Piperaceae family has a diversified and bioactive compounds such as essential oils, unsaturated amides, pyrones, flavonoids, monoterpenes, sesquiterpenes, arylpropanoids and lignoids. **Objective:** In the present study the molluscicide and ovicide actions of an amide provided by the *Piper* genus (Piperaceae) were determined against the adult snail *Biomphalaria glabrata* and embryos of blastula, gastrula, trocophore and veliger stages. **Methods:** The Piperaceae amide was evaluated at concentrations lower than 10 ppm in snail to obtain LC_{90} (lethal concentration causing 90% mortality) and then submitted to evaluation of ovicidal action. **Results and Discussion:** The amide showed 100% molluscicide effect at concentrations lower than 8 ppm and 100% ovicide effect at concentrations lower than 3 ppm in all the stages of embryonic development.

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8.10 Quantification of thimerosal in vaccines without pretreatment, using flame atomic absorption spectrometry (FAAS)

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Introduction: Since the 1930s, thimerosal at high concentrations is used to prevent microbial growth in biological compounds, including many vaccines. It is an organomercurial molecule, and its composition is about 50% mercury in weight. The metabolism or degradation of this molecule results in ethylmercury whose environmental or clinical toxicity is not firmly established yet, but it bears a chemical similarity to methylmercury. **Objectives:** This work proposed a methodology for high concentration organic mercury analysis using the flame atomic absorption technique, without any sample pretreatment in DTP, Td and DT vaccines, Diphtheria and tetanus toxoids, and whole cell pertussis vaccine. **Methods:** The experiments were carried out in a HR-CS-AAS ContrAA 700 (Analytikjena) equipped with a 2D FFT-CCD detector and Echelle grating double monochromator. As continuum light source, a shot-arc xenon lamp was set for 253.65 nm (Hg line). The C_2H_2/air flame worked at 40 L h^{-1} flow rate. A stock solution of 1000 ppm thimerosal USP-RS was used as standard for the assays. All dilutions were performed only with Milli-Q water. All samples were provided by Instituto Butantan. **Results and Discussion:** The aspiration was studied in different rates, ranging the burner height as well. The signals were analyzed in a response surface graph and the best parameters were selected. The analytical curve showed linear correlation ($r^2 > 0.995$) and the standard solutions were stable for up to three days. For the analysis of selectivity and recovery assays, the standard additions method was employed for each sample, and the slopes of the curves obtained were compared to the slope of the analytical curve, using a *t*-test and hypothesis theory. This approach can give information about interferences of matrices, but results did not show such interferences. The developed method showed good precision and recovery rates, ranging below 5.0%, which is required for the AAS technique itself. The LOD and LOQ were, respectively, 1.27 mg L^{-1} and 4.63 mg L^{-1}.

Supported by: Fundação Butantan.

8.11 Validation of sterilization process by ionizing radiation of bromobutyl rubber stopper used as a component of the primary packaging of vaccines produced by Instituto Butantan

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Introduction: The sterilization by ionizing radiation of medical products and pharmaceuticals is being increasingly used. This is due to logistical reasons, or aimed to optimize and increase the efficiency of processes. Similarly, the use of such technique can be justified on the incompatibility of the material in relation to other sterilizing methods. The Instituto Butantan adopted this method for the sterilization of bromobutyl rubber stoppers, used in the primary packaging of multi-dose vaccine, which was made possible through the outsourcing process in a company that has a multipurpose radiator. This work describes the validation of the sterilization process by ionizing radiation of bromobutyl rubber stopper component of the primary packaging of vaccines produced by the Instituto Butantan. **Objective:** To perform the sterilization process validation of bromobutyl rubber stopper used as a component in the primary packaging of vaccines produced by Instituto Butantan, in accordance with requirements in the Resolution RDC no. 210/03 of the Brazilian Health Surveillance Agency (ANVISA). **Methods**: Three lots of bromobutyl rubber stoppers near the expiration date were sterilized. After determining the total microbial load "bioburden" which challenged the process, the stoppers were submitted to a radiation dose of 25 ± 5 kGy, and finally tested in the fungal and bacterial sterility test by the technique of direct inoculation in culture fluid (*fluid thioglycollate* medium and soybean-casein). Twenty different points were determined for the dosimetric mapping (use of dosimeters to evaluate the received dose) positioned by geometric analysis, as: 15 points for distinct biological evaluation using the bioindicator *Bacillus pumilus* (1.17×10^6 CFU/strip) and 5 samples at different points for the sterility test. All items were distributed in three sequential containers. Three cycles of sterilization were performed on three different days (one cycle for each batch of stopper). **Results and Discussion:** All samples tested were satisfactory in the bacterial and fungal sterility test, bioindicator showed no growth and the dose received by the product was within the stipulated acceptance criteria; the highest dose received was 28.8 kGy, the lowest 21.0 kGy and the average of 25.1 kGy sterilized in three lots. The results showed reproducibility between the cycles, obtaining the average coefficient of variation of 4.00% and an average standard deviation of 1.00 kGy. Therefore, the sterilization process of bromobutyl rubber stoppers by ionizing radiation is effective and reproducible, and it is concluded that this process is validated and can be routinely performed under the conditions in which it was evaluated.

Supported by: Fundação Butantan.

8.12 New occurrence of *Amblyomma romitii* Tonelli-Rondelli, 1939 (Acari: Ixodidae) in Pará State, Brazil

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Introduction: *Amblyomma romitii* Tonelli-Rondelli was described based on six females, one male and seven nymphs collected on a capybara (*Hydrochaeris hydrochaeris* Linnaeus), from British Guyana (now Guyana). The area of distribution of the species includes Guyana, French Guyana, Suriname, Venezuela and Brazil. In 1955, it was synonymized with *A. extraoculatum* Neumann; however, based on the type species it was redescribed and validated recently. **Objective:** The aim of this study was to record the second occurrence of this species in Brazilian territory. **Methods**: In April 2009, specimens were sent to the Laboratorio de Parasitologia do Instituto Butantan: one male, two females and two nymphs, collected on a capybara from the city of Rurópolis, southwestern Pará State (04°05'45 "S and 54°54'33" W). Based on an illustrated key, the ticks were identified as *A. romitii*. One of the females was engorged and it was kept in an incubator (BOD) at 27°C and humidity of 80-90%, where it got the posture. The other specimens were fixed in 70% alcohol and were deposited in the Acari collection from Instituto Butantan. **Results and Discussion:** The only confirmation of *A. romitii* in Brazil was a male with hypostome fractured, captured on humans in the state of Pará in 1957. No other report of its occurrence was published since that time in Brazil. This is then the first record of this species found on capybara in this country. Larvae and nymphs were obtained including an engorged female, and studies about the life cycle of this species are in progress.

Suported by: FAPESP and CNPq.

8.13 Ticks of the Neotropical region: illustrated key for immature stages of the genera Argasidae and Ixodidae

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Introduction: Determination of the genera of the tick families Argasidae and Ixodidae in the larval and nymphal stages have been a taxonomic problem, mainly because the few keys for the Neotropical region are chaetotaxic and rarely show illustrations. **Objective:** Considering the importance of the immature stages in the transmission from pathogenic agents for human and animals, the aim of this study was to help the classification of the genus of larvae and nymphs of ticks. **Methods:** Classification of the immature stages, larvae and nymphs, was based on optical and scanning electron microscopy. **Results and Discussion:** Illustrated keys for immatures of the different tick genera of both families that occur in the Neotropics were proposed. Generic diagnosis and updated listings of species for each genus were also included.

Supported by: CNPq and FAPESP.

8.14 Occurrence of heart failure with presence of ascites in a horse on São Joaquim farm of Instituto Butantan

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Introduction: Congestive heart failure (CHF) is a rare disease. It is known as a clinical syndrome characterized by limited cardiac, neuro-humoral activity increased, sodium retention, tissue edema and the presence of transudate in serous cavities. Most cardiac lesions in horses are not serious enough to cause CHF. However, it can occur in foals and adult horses as a result of severe degenerative valvular disease, valvulitis, dilated cardiomyopathy, myocarditis or myocardial necrosis, bacterial endocarditis, effusion, congenital malformation and vascular rupture or obstruction of the pulmonary artery. The most common cause of CHF in horses is valvular heart disease. It can develop quickly or gradually. The most common clinical signs are ventral, preputial, chest and limb edema, indicating widespread venous distension that is a sign of CHF on the right. On physical examination of the animal, there is elevated jugular venous pressure, increased pulse and pathological jugular filling, tachycardia at rest (over 60 beats per minute), tachypnea, pericardial effusion, ascites, lethargy and weight loss. **Objectives:** The aim of this investigation was to determine what have happened to a horse belonging to the São Joaquim Farm. **Results and Discussion:** A male horse, castrated, was found in the paddock with edema in the chest, in the abdomen and of the foreskin. On physical examination there were pale mucous membranes, tachycardia (65 beats per minute), weight loss and apathy. Specific therapy to decrease the edema and to improve the clinical condition of the animal was done. He showed reduced edema, but the clinical picture remained the same. After four days, the horse died, and was necropsied. The internal organs were jaundiced, and there were large amounts of fluid in the abdominal cavity, and presence of fibrin in many arteries, in veins and in the pericardium. It was determined that the fluid in the abdomen was ascites, which is a rare event in horses.

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