## Vetores - Vectors

### VE01 - IDENTIFICATION OF DIFFERENTIALLY EXPRESSED GENES IN *ANOPHELES AQUASALIS INFECTED WITH PLASMODIUM VIVAX*

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Malaria affects 300 million people worldwide every year and 450.000 only in Brazil. In the Brazilian coast, the main malaria vector is *Anopheles aquasalis*. Understanding the interaction mechanisms between mosquito vectors and plasmodia is important for the development of malaria control strategies. We have performed subtraction experiments to identify *Anopheles* molecules potentially important during the initial steps of infection by *Plasmodium vivax*. A total of 2638 clones were sequenced and 981 high quality unique sequences were obtained, with the identification of groups of genes induced by blood and parasite challenge. In insects infected by *P. vivax*, sequences related to embryogenesis, replication, transcription and translation were down-regulated, while sequences related to energy and conversion were up-regulated. Expression of some of the annotated genes was analyzed by real time PCR, which in most cases corroborated the subtraction results. The expression of actin increased with infection time (24 and 36h). A serine protease was down-regulated 24hours after infection in females, while no expression was seen in males and immature stages. On the other hand, there was an increased expression of genes for fibrinogen and bacteria responsive protein in males, in relation to infected females. There were no significant differences in expression of carboxypeptidase and fibrinogen. The bacteria responsive protein gene expression increased with time after infection (2, 24 and 36 hours). Cecropin mRNA was negatively regulated 24h after infection while serpin was up-regulated at 36 hours of infection. These subtraction experiments revealed differentially expressed genes that can be important in the *A. aquasalis* - *P. vivax* interactions, and thus contribute to the development of new malaria transmission-blocking strategies.
Support: CNPq, CAPES, Fapemig, PRONEX

### VE02 - EFFECT OF THE INSECTICIDES DELTAMETRIN AND MALATHION ON THE PROTEIN PROFILE OF THE PHYTOPHAGOUS INSECT *ONCOPELTUS FASCIATUS*

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The hemipteran insect *Oncopeltus fasciatus* is a model for various studies. Deltamethrin and malathion are insecticides that act by direct contact or ingestion. Deltamethrin is an insecticide that reduces and delays the conductance of sodium into the cells and removes the efflux of potassium; it can also inhibit ATPases, resulting in a decrease of potential action and generation of repetitive nerve impulses. Malathion is an organophosphate that inhibits the enzyme acetylcholinesterase in an irreversible manner, causing a severe form of cholinergic poisoning. In the present study we have analyzed the protein profile of several organs of *O. fasciatus* both resistant and sensitive to deltamethrin and malathion. Groups of sensitive adults were treated for 6 and 24 hours by adding 1-µl aliquots of each of the insecticides at several concentrations (serial dilution) onto the ventral region. Untreated adults were used as controls. The initial concentration for malathion was 3 µg / ml and 2.5 µg / ml for deltamethrin. These adults were carefully dissected and the salivary glands, fat bodies and ovaries were removed at 4°C. Homogenates of these organs were obtained at 4°C in the presence of protease inhibitors. The proteins were then analyzed by SDS-PAGE. Resistant insects had previously been obtained in our laboratory; sensitive and resistant adults, as well as their offspring have also been analyzed in this study. These organs, whole first instar nymphs, as well as both immature (yellow) and mature (red) eggs were prepared as described above; the protein profiles were then analyzed by SDS-PAGE. Significant quantitative and qualitative differences in the protein profiles were observed both when sensitive and resistant insects and their offspring were compared, both for insects treated with deltamethrin and malathion. The identification of the proteins of interest is currently under investigation, using proteomics analysis.
Supported by: FAPERJ-PESAGRO, INCT-CNPq/FAPERJ (INCT Molecular Entomology),CAPES, CNPq and PIBIC/CNPq.

## VE03 - POSSIBLE ROLE OF MONOOXYGENASES ON THE RESISTANCE TOWARDS THE INSECTICIDE MALATHION IN THE PHYTOPHAGOUS INSECT *ONCOPELTUS FASCIATUS*

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Chemical insecticides are important tools for controlling vectors of human and veterinary diseases, as well as in agriculture. Many of these compounds act in the central nervous system of the insects. Malathion is an organophosphate that inhibits the enzyme acetylcholinesterase in an irreversible manner, causing a severe form of cholinergic poisoning. The ***indiscriminate*** use of insecticides has led to the selection of insect lineages resistant to many classes of these compounds. Chemical resistance towards insecticides represents a threat to human health and agropecuary, including products for human consume. In general, insecticide resistance can occur by two major mechanisms. The first involves alteration of the binding sites for the insecticides and the second requires mechanisms of detoxification. This last mechanism is denominated metabolic resistance and is due to the increase of detoxifying enzymes, such as monooxygenases. The hemipteran insect *Oncopeltus fasciatus* is a model for various studies. In this study we compared the levels of monooxygenases in sensitive and malathion-resistant *Oncopeltus fasciatus*. Groups of sensitive adults were treated for 5 and 16 hours by adding 1-µl aliquots of 3 µg / ml malathion onto the ventral region. Untreated adults were used as controls. Resistant insects had previously been obtained in our laboratory; sensitive and resistant adults were analyzed in this study. Whole adults were homogenated at 4$^{o}$C and the protein contents were quantified. The monooxygenase activity of all the systems was then measured. Significant quantitative differences in the monooxygenase activity were observed when sensitive and resistant insects were compared. Also, the sensitive insects that were treated with malathion for 5 and 16 hours showed significant quantitative differences in the monooxygenase activity, as compared to the non-treated sensitive insects. Ongoing experiments in our laboratory aim to indentify the genes that are involved in malathion resistance in *O. fasciatus*.
Supported by: FAPERJ-PESAGRO, INCT-CNPq/FAPERJ (INCT Molecular Entomology), CAPES, CNPq and PIBIC/CNPq.

## VE04 - OCCURRENCE, CULTURE, MORPHOLOGICAL CHARACTERIZATION AND PHYLOGENY OF *TRYPANOSOMA* (*MEGATRYPANUM*) *MELOPHAGIUM* FROM SHEEP KEDS IN CROATIA

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*Trypanosoma* (*Megatrypanum*) *melophagium* is a nonpathogenic parasite of sheep transmitted by the sheep ked *Melophagus ovinus* (Hippoboscidae), an obligatory bloodsucking ectoparasite restricted to sheep. *T. melophagium* is a *T. theileri*-related trypanosome also restricted to sheep. Sheep keds were detected in fleece of all sheep not treated against ectoparasites from six organic farms in Croatia. A large number of keds were collected, dissected and their guts examined for the presence of trypanosomes by microscopy. Keds were positive for trypanosomes morphologically compatible to *T. melophagium,* showing epimastigotes in the lumen or attached to the gut epithelium, and rounded metacyclic trypomastigotes. Positive guts generated a culture of trypanosome with large epimastigotes, which was identified by ITS1rDNA-PCR as a *T. theileri*-trypanosome. Sequence of SSUrDNA was employed to infer the positioning of this new trypanosome in relation to other *T. theileri*-trypanosome in the phylogenetic tree of *Trypanosoma*. Although differing from all other *T. theileri*-trypanosomes, the isolate from sheep keds nested within *T. theileri* clade, closer to isolates from antelope and cervid than to a cattle isolate of *T. theileri* from Croatia. Together, host-origin, morphological and molecular data corroborate classification of the trypanosome from sheep keds as *T. melophagium*. The very low parasitemia in animals infected with *T. melophagium* prevented the observation of the scanty parasite on smears of blood sheep. We have also performed a survey by IFA for antibodies against *T. melophagium* in sheep infested with trypanosome-positive keds and in ked-free sheep. Results were all negative, corroborating previous reports about lack of antibodies against *T. theileri*-trypanosomes in infected bovids. This is the first description of *T. melophagium* in sheep keds from Croatia, and the first time that an isolate from this species was established in culture and included in phylogenetic studies.
Supported by CNPq-Brasil

**VE05 - The role of lipophosphoglycan in the interaction of *Leishmania chagasi* with *Lutzomyia longipalpis*, the vector of visceral leishmaniasis.**

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*Leishmania* promastigotes synthesize an abundance of membrane-anchored glycoconjugates such as the lipophosphoglycan (LPG), the major surface glycoconjugate of *Leishmania*. It is involved in the vector's specificity for parasite development, as demonstrated in Old World sandfly vectors. However, there is no study relating the role of the LPG in the interaction of *Leishmania chagasi* with its sandfly vector *Lu. longipalpis*. Here, *LPG1* deficient mutants of *L. chagasi*, which synthesize a truncated LPG core, were produced and tested in *Lu. longipalpis*. Adult female sandflies were fed through a chick skin membrane device containing mouse blood with *L. chagasi* BH46 wild type (*Lc*wt) and *LPG1*$^{-}$ knockout (*Lclpg1*KO) ($4x10^7$ parasites/mL). Blood-engorged females were separated and sacrificed in 12h intervals. *Lc*wt and *Lclpg1*KO produced infections in 100% of the sandflies in 12h. At 48h, infections were absent in 27% of the insects infected with *Lclpg1*KO and few viable promastigotes were found in the positive sandflies. After 60h, no positive insects were found in the *Lclpg1*KO group. In contrast, 100% of the insects infected with *Lc*wt were positive in all times. To determine the role of midgut proteases in the early *Lclpg1*KO parasite mortality, a trypsin inhibitor was added to the bloodmeal. The addition of trypsin inhibitor increased the parasites survival in *Lc*wt and *Lclpg1*KO. Supporting the view that the blood-fed midgut is a hostile environment for the parasites and that the LPG coat is protective during this early stage of infection, the trypsin inhibition promoted the survival of *Lclpg1*KO group for 48h. However, the *Lclpg1*KO parasites were excreted with the bloodmeal after 48h. These data suggest that the presence of LPG is essential for the parasite survival in the early blood-fed midgut and for the posterior development in the midgut even in the *Lu. longipalpis*.
Supported by PAPES IV, AMSURD (Pôle Amériques) and CPqRR/FIOCRUZ.

**VE06 - MORPHOLOGICAL ASPECTS OF THE SENSORIAL ORGANS OF THE ANTENNAE OF*Lutzomyia ovallesi* AND *Lutzomyia migonei* (DIPTERA: PSYCHODIDAE) FEMALES, VECTORS OF CUTANEOUS LEISHMANIASIS IN SOUTH AMERICA.**

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*Lutzomyia migonei* and *Lutzomyia ovallesi* are considered anthropophylic species that lives around the houses, found in South America, especially in Venezuela, and are incriminated vector species of *Leishmania braziliensis* and *Leishmania mexicana*. The sensilla are sensorial organs of different forms and functions, usually found in different places in the body of the insects, as well as, the antennae. The sensilla present in the antennae are responsible for the recognizing stimulus and are considered involved in the feeding process, aggregation, mating and are capable of noticing of odors, humidity and temperature. As both of species are morphologically very similar and sympatric in certain areas, in this study, we compared the types of sensilas found in female antennae organs, in order to detect within and between-species differences that woud help in their identification, using the Scanning Eletron Microscope (SEM) and *Laser* Confocal Microscope (LCM). We used *L. migonei* and *L. ovallesi* from a closed laboratory colony maintened in the Experimental Parasitology Laboratory, University of the Andes, Merida and Venezuela. The antennae were dissected, fixed in glutaraldehyde and processed for scanning eletron microscope. The chaetic sensilla type were fixed in formaldehyde to be processed for nuclei visualization with DAPI staining and lectin bindings (CON A, RCA, PNA, BS1, WGA, HPA). The SEM revealed that both species have a par of thin antennae composed of sixteen segments. Four sensillae types were observed: chaetic, squame, coeloconic (grooved and "praying-hands"), small and large trichoid (both fine- and blunt-tipped). The first segment, the scape, has a triangular format and the presence of the *smaller trichoid and the blunt-tip trichoid* sensillae was identificaded. The second segment, the pedicel, is of a sphere-like shape and the sensilla present were the smaller trichoid and pointed-tip trichoid. Posterior to the pedicel, it was possible to visualize fourteen extremely thin flagellomeres covered with microtrichias. The first flagellomeres in both species is diferent from the others due to its superior size that is extremely long. In *L. ovallesi*, the *pointed-tip trichoid* sensillae was found, in all the flagellomeres, but in *L. migonei* this type of sensillae was only found from the fifth flagellomere to the fourteenth, being absent in the first four flagellomeres. The others flagellomeres were extremely similar in both species. The DAPI staining marked nonspecifically the nucleus of chaetic sensilla. The lectin bindings confirmed the existence of distinct cell populations in chaetic sensilla as visualized by Concanavalin A. The WGA lectin binding enhanced the epithelium and the some others lectins as the HPA were negatives. Our morphological observations can be used as additional taxonomic characters to help on their identification.

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## VE07 - ROLE OF SNARE PROTEIN IN THE SECRETIOPN CASCADE OF TICK SALIVA

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Soluble *N*-ethylmaleimide-sensitive fusion protein attachment protein receptors (SNAREs) have been identified as the key components of the protein complexes that facilitate the vesicle traffic. Members of this family are expressed in all eukaryotic cells including neuronal and non-neuronal cells, of which Ykt6 (from *Saccharomyces cerevisiae*, v-SNARE) is proved to be a multifunctional protein in the membrane fusion. In the present study, a tick homologue of Ykt6, termed HlYkt6 was isolated from the ixodid tick *Haemaphysalis longicornis*. The predicted 199-residue protein HlYkt6 contains a longin domain (LD) that only resides in v-SNAREs and a v-SNARE motif. The putative amino acid sequence of HlYkt6 shows higher similarity to the mammal (*Mus musculus*, 64%) than to the yeast (42.8%). RT-PCR and Western blot analysis indicated that the gene and encoded protein was expressed ubiquitously in different tissues of the tick. Silencing of HlYkt6 gene resulted in a significant decrease of the engorged body weight (82.9 ± 26.8 mg vs. 232.17 ± 59.1 mg in PBS-injected control group and 178.7 ± 57.0 mg in GFP dsRNA-injected control group) and high mortality of replete ticks (100% in tested group vs. 4.8 % and 20.4% in control groups). Disruption of HlYkt6 mRNA led to the suppression of the saliva secretion. Moreover, the secreted liquid in HlYkt6 dsRNA-treated ticks showed lower anti-coagulant activity (APTT time: 25.25 ± 1.50s) than that of the control groups (39.25 ± 0.50s in PBS-treated control group and 40.0 ± 1.41s in GFP dsRNA-treated control group, $P<0.001$ by student's *t*-test). The result suggests an important role of HlYkt6 protein in the secretion cascade of tick saliva, and disruption of HlYkt6 mRNA probably blocks the exocytosis, hence lead to the failure of saliva secretion, which further effects tick successful blood feeding and survival.
Supported by BRAIN and JSPS.

## VE08 - Cysteine peptidase inhibitors impairs *Trypanosoma cruzi* adhesion to *Rhodnius prolixus* explanted midguts

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Cruzipain is the major lysosomal cysteine peptidase of *Trypanosoma cruzi*, which is the causative agent of Chagas' disease. This enzyme is expressed at variable levels in all developmental forms and strains of the parasite. Cruzipain is required for parasite infectivity and intracellular growth in mammalian cells, however, its role in parasite interaction with the vector has been overlooked. Here, we have analyzed the effects of the pre-treatment of *T. cruzi* with a panel of different cysteine peptidase inhibitors or anti-cruzipain antibodies on the parasite adhesion to *Rhodnius prolixus* posterior midgut. The parasites were treated for 1 hour with iodoacetamide, leupeptin, antipain or E-64 at 10 µM or cystatin at 1 µg/ml and allowed to bind to *R. prolixus* explanted guts for 15 minutes. Afterwards, the number of parasites/midgut epithelial cells was estimated by randomly counting at least 100 epithelial cells in quadruplicate. The interaction rate of the parasites treated with the cysteine peptidase inhibitors was on average 70% lower in comparison to the untreated parasites. In addition, the treatment of *T. cruzi* cells with anti-cruzipain antibodies (1:1000) induced a significant reduction (64%) in the adhesion to the insect posterior midgut. Collectively, our results suggest a possible involvement of cysteine peptidases in the interaction between *T. cruzi* and epithelial cells from the invertebrate host. Supported by: MCT/CNPq, FAPERJ and FIOCRUZ.

## VE09 - ECO-BIOLOGICAL CHARACTERISTICS OF *RHODNIUS PALLESCENS* IN SANTA FE DISTRICT, PANAMA

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*Rhodnius pallescens* is considered the main vector of Chagas disease in Panama and the only triatomine bug that transmits *Trypanosoma rangeli*. Nevertheless, updated information on its biogeographical traits is scarce/limited.
We report here a study designed to extend the known geographical distribution of this triatomine, its eco-biological characteristics and epidemiological importance.
The study area was located in the Province of Veraguas, District of Santa Fe in Eastern Panama. Specifically, the bugs were collected on peridomestic palm trees (*Attalea butyracea)* from the community of La Culaca (494477.15 mE 941285.93 mN). This region is mainly a humid forest with an altitude ranging from 400 to 800 m. Triatomines were collected systematically by direct searches on the palm crow. The host feeding sources were evaluated by a Dot blot assay and the trypanosome infection index by a PCR analysis.
To our knowledge, this is the firth report of *R. pallescens* on this mountainous region of the country. A total of 633 *R. pallescens* (nymphs and adults) were collected during this study. Preliminary observations show that the morphology of *R. pallescens* from La Culaca differ significantly from reference specimens of lower altitudes found in Central Panama. The bugs collected from La Culaca presented an overall darker coloration and larger sizes. It was found that 55% of the examined bugs fed on *Didelphis marsupialis,* the finding of positive bugs for others feeding sources was scarce (2-4%). A high infection index with *T. rangeli* (68.7%) was detected; this percentage is higher compared with those found in bugs collected in Central Panama (40%). The infection index with *Trypanosoma cruzi* was of 62.7% and with mixed infections of 44.2%. These results will improve our understanding about the morphological variability presented by *R. pallescens* and will also give support for the establishment of a national entomological surveillance program.
Supported by ICGES.

## VE10 - SEASONALITY AND BEHAVIOR OF TABANIDS (DIPTERA: TABANIDAE) IN PLANALTO CATARINENSE, SANTA CATARINA – BRAZIL

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Tabanids are considered an important mechanical transmitters of several pathogenic agents to livestock's in South America, among them, the *Trypanosoma vivax*, *Trypanosoma evansi* and *Anaplasma* spp. The objective of this study is to determine the composition and the behavior of tabanids in Planalto Catarinense that could be an important place in a future epidemic outbreak. The seasonality of tabanids was analyzed during the period of March 2007 to February 2009. One black mixed breed horse was used as bait for the collections that occurred once a month during two hours in the afternoon. 915 samples were collected and corresponded to six genera. *Catachlorops* spp. was the most predominant (62%), following by *Chrysops* spp., *Tabanus* spp. (13%), *Dichelacera* spp. (6,7%), *Fidena* spp. (4,7%) and *Acanthocera* spp. (0,7%). The tabanids appeared in periods of heat, mainly in the summer, also occurring in spring, when the relative humidity had a mean of 78,32%. We observed that these insects disappeared during May to August (autumn and winter), when the relative humidity had a mean of 81,94%. Most of insects were collected in thorax and abdomen (356), following by superior and inferior limbs (253) and in smaller amount in head and neck (168). The genera *Catachlorops* obtained larger incidence in thorax/abdomen and limbs, with 315 and 183 samples respectively, while the genera *Chrysops* had occurrence only in neck and head with 98 samples, not observed in other body areas. This is the first work of tabanids description in Santa Catarina, Brazil.

**VE11 - DISTRIBUTION OF NERVE FIBERS IN THE SALIVARY GLANDS OF *Rhodnius prolixus***

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*Rhodnius prolixus* is a hematophagous insect vector of *Trypanosoma cruzi* and *Trypanosoma rangeli*. The hematophagy is possible, besides other factors, because of saliva secreted during the bite that antagonizes hemostatic, inflammatory and immunological systems imposed by the host. The salivary gland is surrounded by two muscles layers, which contracts and ejects saliva during the bite. With the purpose of characterize the gland innervation, a study was carried out with 5$^{th}$ instar nymphs of *R. prolixus* (unfed and during feeding). The insects were provided by the Laboratory of Triatomines (CPqRR - FIOCRUZ). Glands were processed as routine for Laser Scanning Microscopy (LSM) and Scanning Electron Microscopy (SEM). For LSM, the glands were incubated with the antibodies anti-serotonin, anti-tyrosine and anti-dopamine; control group was not incubated with the primary antibody. The SEM indicated that at least two nervous arrive in the salivary gland: one by the accessory lobule and another by the posterior and/or anterior region of principal lobule. They ramify and reach the muscle fibers. In unfed insects, only the activity of serotonin was observed along the entire duct system. In insects during feeding, the whole salivary gland was surrounded by a meshwork of serotonergic branched fibers. Tyrosine innervation was found in the muscle layers in the middle of the gland. The duct, during feeding, showed serotonergic, dopaminergic and tyrosine activity. Further studies are being done in order to better understand the morphological characteristics of the salivary complex and to correlate them with the organ physiology.
Financial support: CPqRR/FIOCRUZ

**VE13 - The role of Triatominae saliva and lysophosphatidylcholine as modulators of intracellular signaling in murine macrophages**

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*Rhodnius prolixus* is a vector of *Trypanosoma cruzi* in South America. The parasite is transmitted by vector feces deposited on human skin during blood feeding. One of the routes of host cell invasion occurs through the wound produced by the insect bite. Parasite thus faces a cell environment within the wound previously stimulated by saliva. <i>R. prolixus<i> saliva and feces stores the bioactive lipid lysophosphatidylcholine (LPC) (Golodne et al, 2003). LPC is a powerful modulator of cell signaling in mammalian cells. Our group has recently shown that vector-derived LPC is an enhancer of *T. cruzi* infection through an yet completely understood immunosuppressant mechanism (Mesquita et al, IAI, 2008). In the present work we tested the role of LPC on intracellular signaling in murine peritoneal macrophages. Saliva and LPC were able to reduce both lipopolysaccharide (LPS) and *T. cruzi*-induced NO production in macrophages in a dose dependent fashion. The expression of inducible nitric oxide synthase (iNOS) gene in LPS stimulated murine macrophages was blocked by saliva and LPC. Moreover, protein kinase-directed antibodies identified the activation of GSK-3 and Akt in saliva-treated macrophages. Macrophages treated with LPC or saliva showed a diferent morphology than controls cells. The above set of results shows that previous exposition to saliva manipulates the intracellular signaling system of host macrophages. The identification of macrophage proteins regulated by such intracellular signaling systems is under way in our laboratory and may conduct to novel strategies in the future to block Chagas disease transmission. Supported by CNPq, FAPERJ, IFS, OMS.

## VE14 - POLYMERASE CHAIN REACTION FOR DETECTION OF *TRYPANOSOMA CRUZI* IN NATURAL INFECTED TRIATOMINES FROM SEMIARID POTIGUAR

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The Polymerase Chain Reaction (PCR) has been applied for detection of *Trypanosoma cruzi* using whole blood and sera of chagasic patients, or in feces of triatomine bugs with high sensitivity. In this study *T. cruzi* was detected by parasitological techniques, using intestinal contents of triatomines captured at intradomicile, peridomicile and sylvatic environments of three municipalities of the State of Rio Grande Norte. All captured triatomine bugs had their species identified and samples of their intestinal contents were obtained by abdominal dissection, and evaluated individually under a microscope at 400× magnification for the presence of trypanosomes. The species of triatomines captured were: *Triatoma brasiliensis*, *T. pseudomaculata* and *Panstrongylus lutzi*. *T. cruzi* natural infection was higher on *P. lutzi*, detected by direct exam in 28.3%, xenoculture 22.2% and PCR 67.9%. While in *T. brasiliensis* the *T. cruzi* was detected by direct examination, xenoculture and PCR, respectively 7.5%, 11.3% and 23.4%. The rate of natural infection was lower in *T. pseudomaculata* with 1.1% of positivity by direct exam, while PCR was 28.6% of examined triatomines. PCR was more sensitivity to detect *T. cruzi* than others techniques and demonstrated great potential for application in molecular epidemiology to monitor infection status in field studies. *T. brasiliensis*, *T. pseudomaculata* and *P. lutzi* are endemic species in the semiarid zone of the northeast of Brazil. *T. brasiliensis* is considered the most important vector in this region with wide geographical distribution and occupies a great variety of ecotopes. The identification of *P. lutzi* with high *T. cruzi* natural infection index in sylvatic environmental emphasizes the transmission linking among sylvatic and domestic cycles and reinforces the need for constant epidemiological surveillance to prevent the spread of the parasite.
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## VE15 - *ANOPHELES SPP.* (DIPTERA:CULICIDAE) VARIATIONS AND DYNAMICS IN A RURAL SETTLEMENT AT ACRELÂNDIA, ACRE, BRASIL.

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*Anopheles* spp. include species of major epidemiological importance, because of their role as the main malaria vectors. Studying mosquito variability, their behavior and the various regions they infest are the main objectives of medical entomology. This study consists in monitoring mosquito variability during one year, in two different regions of a rural settlement, the “Ramal do Granada” located in Acrelândia, Acre. The collections are performed during three days, two days for three-hour captures and one day for a twelve-hour capture. Mosqutoes were collected by human landing inside the houses and at the peridomiciles. Each collection is made between intervals of two months, following the seasonal variation of rainfall, environmental factor closely connected to the presence of the anopheles. Populational variations and density has been analyzed, together with molecular analysis of *Anopheles darlingi* by mtDNA sequencing. The number of collected mosquitoes was greater in all collections nearby the settlement limits, representing newer settlements. The twelve-hour captures have shown the presence of only one density peak occuring between 7:00PM to 9:00 PM. Several anopheline species have been identified: *A. darlingi, A. deaneorum, A. triannulatus, A. rangeli, A. albitarsis, A.brasiliensis, A. oswaldoi,* with predominance of *A. darlingi*, that presented low intraspecific variation. The data presented shows differences inside the settlement that correlate with malaria transmission and could increase the tools to improve entomological surveillance.
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