

SIMPÓSIO DE ECOLOGIA, GENÉTICA E EVOLUÇÃO DE *DROSOPHILA* 08 A 11 de novembro, Brasília – DF, Brasil



Resumos

Abstracts

IX SEGED



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Introdução

O Simpósio de Ecologia, Genética e Evolução de *Drosophila* (SEGED) é um evento bianual que reúne drosofilistas do Brasil e do exterior desde 1999, e conta sempre com uma grande participação de estudantes. Em decorrência do constante diálogo entre os diversos laboratórios, os encontros têm sido muito produtivos para a discussão de problemas e consolidação de colaborações. Tendo em vista que as moscas do gênero *Drosophila* são excelentes modelos para estudos em diversas áreas (provavelmente os organismos eucariotos mais investigados pela Ciência), essas parcerias podem contribuir também para o desenvolvimento de áreas aplicadas, como a Biologia da Conservação e o controle biológico da dengue.

A primeira edição do SEGED ocorreu em 1999, na UFRJ, e fez tanto sucesso que a partir de então os eventos passaram a ser organizados bianualmente, sem interrupção. O número de estudantes e profissionais (brasileiros e estrangeiros) interessados em participar destas reuniões científicas tem aumentado a cada biênio, assim como as inovações no evento. Em 2013, por exemplo, foi incorporada ao simpósio a premiação dos melhores trabalhos dos estudantes, o que será mantido em 2015. As edições do evento que sucederam à primeira foram: (2001) na UNESP, São José do Rio Preto/SP, (2003) na UFRGS, Porto Alegre/RS, (2005) na UNICAMP, Campinas/SP, (2007) na USP, Ribeirão Preto/SP, (2009) na UFSC, Florianópolis/SC, (2011) no Museu Paraense Emílio Goeldi, Belém/PA, e (2013) na UFPE, em Porto de Galinhas/PE. A presente edição (2015) é a primeira realizada na Região Centro-Oeste.

Dentre as contribuições do evento, destacam-se:

 interação e convívio de grande parte da comunidade científica do país que trabalha e/ou interessa-se pelo estudo dos drosofilídeos, propiciando a consolidação de colaborações já existentes e o estabelecimento de novas;

ii. oportunidade para que os estudantes de graduação e pós-graduação apresentem e discutam os resultados de suas pesquisas com profissionais mais experientes;

iii. oportunidade para que os congressistas conheçam e debatam as últimas novidades nas grandes áreas contempladas por estudos com Drosophila;

iv. Apresentação de jovens talentos que atuam ou atuaram no exterior;

 v. estímulo a estudantes de graduação e pós-graduação mediante premiação dos melhores trabalhos nas áreas de Ecologia, Genética e Evolução, após avaliação por um qualificado Comitê Científico, representando diferentes Instituições do Brasil;
 vi. ampliação da inserção dos Programas Pós-Graduação vinculados ao evento no cenário internacional.



A Universidade de Brasília

Em 2015, o SEGED será realizado no Auditório 1 do Instituto de Ciências Biológicas da Universidade de Brasília. O texto a seguir relata sucintamente a história dessa instituição.

A criação da Fundação Universidade de Brasília foi proposta à Câmara dos Deputados no primeiro ato presidencial após a inauguração de Brasília, em 21/04/1960. O ano e meio que separou a proposição do ato oficial da inauguração da Universidade de Brasília (Lei n.º 3.998, de 15/12/1961) foi período que configurou a plenitude inovadora do processo de construção e implementação do Projeto Político Pedagógico. O antropólogo Darcy Ribeiro, idealizador, fundador e primeiro reitor da UnB, sonhava com uma instituição voltada para as transformações, diferente do modelo tradicional criado na década de 1930. No Brasil, foi a primeira a ser dividida em institutos centrais e faculdades. Nessa perspectiva foram criados os cursos-tronco, nos quais os alunos tinham a formação básica e, depois de dois anos, seguiam para os institutos e faculdades.

Desde então, a UnB cresceu muito. Em 2002, a UnB tinha 21.734 alunos regulares registrados nos cursos de graduação, quase 53 vezes o número de alunos do primeiro vestibular, em 1961. Foram criados quinze cursos noturnos, e na pós-graduação, o aumento no número de alunos também foi significativo. Atualmente, a instituição conta com 2.594 docentes efetivos que respondem por 117 cursos de graduação, 97 de mestrado e 70 de doutorado. Em 2013 a UnB atendeu mais que 45.000 alunos.

O *campus* Universitário Darcy Ribeiro - nome dado durante homenagem feita em fevereiro de 1995, dois anos antes da morte do idealizador da UnB -, tem 3,95 milhões de metros quadrados (395 ha), equivalente a 33 estádios do Maracanã, o maior estádio do mundo. Desses, mais de 500 mil metros quadrados são de área construída. O *Campus* Darcy Ribeiro oferece agências bancárias, agência dos Correios, posto de gasolina, lojas de conveniência, barbearia, sapataria, papelarias, fotocopiadoras, livraria, restaurantes e lanchonetes, dentre outros serviços.







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Ecologia

Ecology



Urbanization effects in the assemblages of drosophilids (Diptera, Insecta) in the Atlantic Forest in northeastern Brazil

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THE REPLACEMENT of forests by urban landscapes is an event taking place quickly around the world, challenging conservation biology to understand how it affects the richness and abundance of species. The drosophilids are excellent models to assess this issue. These are small insects, easy to catch, abundant in nature and sensitive to small environmental changes, which is reflected in the size of natural populations and in the diversity of species occupying a particular habitat. Here we investigated the abundance and richness of drosophilids in two areas with different levels of conservation in the Atlantic Forest dominium, in Pernambuco sub region. This area is one of the most deforested of the Brazilian Atlantic Forest, the least studied and least protected by conservation actions. Drosophilids were collected once a month for a year, starting in May 2012, in Dois Irmãos Park (DOI), a conservation unit in the municipality of Recife, state of Pernambuco and in a urbanized environment, the Rural Federal University of Pernambuco (RUR), that surrounds this preservation area. Drosophilids were captured using traps containing banana bait. In each collection effort, 10 traps were spaced 30 m away from one another along a transect, and were left for three consecutive days. In total, 52,516 drosophilids distributed in 44 species were collected. RUR showed higher species richness, 41, compared to 29 species observed in the preserved environment, DOI. It was observed greater abundance of native species in DOI, emphasizing the presence of Drosophila willistoni and D. paulistorum. The exotic species, D. malerkotliana and Zaprionus indianus, were the most abundant in RUR. The highest abundance of exotic species in more urbanized areas confirms what was found in other studies with drosophilids. Our results reveal that these organisms are excellent indicators to assess the conservation status of certain environments in the Atlantic Forest of Northeast Brazil.

> Financial aid: FACEPE/CAPES/CNPq



High richness of drosophilids (Insecta, Diptera) in a Brejo de Altitude area in Pernambuco, Brazil

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THE BRAZILIAN ATLANTIC Forest is divided into eight sub-regions, among which is the Brejos de Altitude. This sub region is located north of the São Francisco river, which extends from the state of Alagoas to Rio Grande do Norte. The Brejos de Altitude occurs at altitudes above 500 meters and consists of "islands" of forest surrounded by semi-arid vegetation (caatinga). Many studies have pointed to the high species richness in the Brejos de Altitude indicating these areas as priorities for biodiversity conservation. However, this is one of the most deforested sub regions of the Brazilian Atlantic Forest, mainly by anthropogenic activities related to agribusiness. This study contributes to the knowledge of the richness and abundance of Drosophilidae family in an area of Brejos de Altitude (Mata da Colônia) in the municipality of Bonito, Pernambuco. Monthly samples were taken for a year, starting in February 2010. For each sample were used 10 traps made from plastic bottles using banana bait. The traps were distributed randomly in the study area and apart for at least 30 meters. In total were sampled 27,654 drosophilids belonging to more than 50 species, including four genres: Drosophila, Zaprionus, Scaptodrosophila and Zygothrica. Drosophila willistoni was the most abundant species in all months of sampling, representing more than 65% of drosophilids. The second most abundant species was D. simulans, which represented approximately 7% of the total sample. The native species were always more abundant in relation to exotic in every month of sampling. Data from this study are compared with those obtained for other surveys to coastal areas of Atlantic Forest in northeastern Brazil.

> Financial support: FACEPE/CAPES/CNPq



Temporal variation of drosophilids (Diptera; Drosophilidae) in response to rainfall in a rainforest remnant in the amazon estuary

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ONE OF TODAY'S most pertinent issues is trying to understand how climatic conditions affect ecosystem dynamics. In terms of biodiversity, the challenge is to estimate the degree of impact in order to find a suitable response to environmental changes. Drosophilids are insects which are highly sensitive to changes in environmental variables, providing rapid responses to questions concerning population growth and are a favored choice as bioindicators of recent environmental changes in relation to biodiversity. In this study, the influence of climatic variability on the tropic community of frugivorous drosophilids was tested, evaluating the temporal dynamics of the species and the overall structure of the community. Fortnightly collections were made in order to meet these requirements over a period of four years in a rainforest remnant located in the metropolitan region of the city of Belém, on the Amazon estuary. Drosophilid data was obtained using the trap method, with climatic elements were obtained using data provided by the meteorological office located in Belém. Species-specific responses concerning climatic factors were observed, especially in relation to quantities of rainfall. Time series models demonstrated the relationship between the abundance and richness of the drosophilids in relation to accumulated rainfall, as well as the previous episodes in terms of the number of individual specimens and species present within the community. The observed standards contributed to better understanding of the way that the drosophilid community functions in the Amazon region, providing us with an insight into how the rainfall can affect the structure of this community.

Keywords: Population fluctuation; Rainfall; Time series



The assemblage of Drosophilidae of remnants of the Coastal Forests of Bahia in Pratigi protection area

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THE HIGH INTEREST in using Drosophila for diversity monitoring reinforces the importance in filling geographical gaps in the knowledge of the diversity of this group. So, the aim of the present study is to perform a biodiversity inventory of Drosophilidae in an important area of forest remnants of the ecoregion of Coastal Forests of Bahia, situated in Juliana Farm, Igrapiúna, in Ecopole II of Pratigi protection area. The forest remnants are located in an agroforestry production area. Samples were taken with banana-baited traps in 10 transects of 300m. Each transect was spaced at least 500m from each other and samples were taken in five points per transect, spaced 60m from each other, in August 2014 and March 2015. Specimens were identified by external morphology. A total of 5,030 individuals were analyzed, distributed at least in 33 morphospecies of the genera Drosophila, Scaptodrosophila, Zaprionus, Neotanygastrella and Zygothrica. Drosophila was the most abundant genus, especially the willistoni (49,7%) and saltans (29,8%) species groups, besides melanogaster, annulimana, cardini, coffeata, immigrans, repleta and tripunctata groups and D. impudica. A comparison of the transects sampled in both seasons showed that the abundance of exotic species was higher in March (17%) than in August (0,3%). A total of 113 specimens were sampled as adults or preadults in flowers and fungi, distributed in the genera Zygothrica (76%), Drosophila (13%), Hirtodrosophila (6%) and Scaptodrosophila (5%). Future analysis of male terminalia of the specimens and of records in the literature possibly will confirm the first records for Bahia of D. coffeata species group, D. elliptica subgroup and of several species of Zygothrica and Neotanygastrella. So, the present study contributes to fill a geographical gap of knowledge of Drosophilidae diversity and obtain some preliminary data to allow future studies on spatio-temporal dynamics, monitoring and testing other ecological and evolutionary hypothesis.

Keywords: Atlantic Forest, diversity inventory, community ecology Financial aid: PPBio Mata Atlântica, OCT – Organização de Conservação da Terra



First registry of *Drosophila carcinophila* at South America, Brazil

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THIS WORK DESCRIBES the occurrence of *Drosophila carcinophila* in Southern Brazilian coastal strand of vegetation, on Rio Vermelho's neighborhood, Florianópolis, Santa Catarina state. The samples were gathered monthly, using six PET bottles traps with two liters of capacity, each one filled with chicken liver and bovine meat, with 25 g of both mixed in. The traps were placed on field for four days per month, along 12 months, always using the same bait. These collects were made from November 2012 until October 2013. The six traps were hung at strand vegetation by twines, varying between 60 to 130 cm height. All the 7671 individuals collected were kept in ethanol 70°, and its identification reached the taxonomic level of family. For drosophila was found, but repleta group members, and an invasive species recently discovered in Santa Catarina state: *D. suzukii*. This record leads to evidences that *D. carcinophila* spreads throughout entire Brazilian territory, coming from its original distribution site, the Caribbean islands, since the 1960s when it was collected by William Heed.

Keywords: strand vegetation, Drosophilidae, Brazil, South America



The use of above-species taxonomic categories of Drosophilidae for monitoring areas

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MONITORING IS THE use of biological indicator taxa, in a time series, to inform about the environment and its biodiversity based on the analysis of patterns already known. Therefore, it can identify potential deviations from the norm that might have been generated by various effects. Considering that environmental changes are accelerating, we need to speed the analysis of biodiversity for its conservation. An alternative to reduce the cost and time required for processing the sampled material is to identify the organisms at higher taxonomic categories, a concept known as "taxonomic sufficiency." In this paper we investigate if the identification of drosophilids to the category of "group" and genus could be used similarly to the use of its species, since the availability of qualified people for identification in long-term projects is often restrictive. Twelve collections were performed bi-monthly between 2013 and 2015, in forest and savanna areas of the IBGE Ecological Reserve (DF) in Cerrado biome. The sampling effort was 90 traps per catch. We captured 74.362 individuals representing 50 species and four genera: Rhinoleucophenga, Scaptodrosophila, Zaprionus and Drosophila. Due to the high richness of Drosophila, it is traditionally divided into "groups" of species, usually monophyletic. Thirteen of these groups have been identified. All comparisons made between species versus groups/genus matrices, using the Mantel test with 9999 permutations (Bray-Curtis method), were significant (<0.001, with r> 0.91 for all analyzes performed). Thus, the use of above-species categories may be an alternative route of identification in long-term projects. As drosophilids are susceptible to the same threats that affect other taxa, the monitoring of areas, including protected ones, must be continuously encouraged. In this sense, it would be of great value an identification process that generates similar results to the traditional ones, while demanding less time and resources.



Monitoring drosophilids in the Brazilian Savanna: two years of standardized collections

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VARIATIONS IN SPACE and time, as observed in climatic conditions and resource availability, are important factors that modify drosophilid communities. In the Brazilian Savanna, particularly, drosophilid populations suffer severe bottlenecks during the dry season, from May to September, and recover again in the rainy season. This study investigated the temporal variation of drosophilids in this highly seasonal biome over two years, using a well-established experimental design, aiming at improving the understanding of the species-time relationship and its use in conservation studies. Twelve collections were made bimonthly, between October 2013 and August 2015, using 90 traps with fermented banana baits distributed in forest (45) and savanna (45) areas of the IBGE Ecological Reserve (DF). Among the 74.362 specimens captured, we recognized 50 species, whose distribution differed markedly throughout time. Approximately 90% of all individuals came from of the six collections made in the rainy season (November to April), and the highest richness was recorded in February and April, for both forests (40 species; 83% of the total number of species sampled) and savannas (33 species; 89%). In addition, in February and April we recorded the highest abundance of neotropical species and the lowest βdiversity in forest areas. These results contributed to understand the dynamic species-time relationship of these insect assemblages, and allow us to suggest that, for monitoring purposes, the best sampling period is the final half of the rainy season.



Monitoring *Drosophila suzukii* in the Cerrado: is this species establishing?

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ARGUABLY, THE MOST successful species in adapting to new environments and changing them drastically is *Homo sapiens*. The presence of invasive species, in turn, has increased along with human trade between continents. However, not all introduced species will become invasive, only those that establish and extend their geographical distribution in the new area. Drosophila suzukii is most likely native from Southeast Asia, and has been found in Europe and the US in 2008. In March 2013, it was recorded in southern Brazil. In this study, we evaluated the variation in the abundance of this species in the IBGE Ecological Reserve (DF), where we have been collecting drosophilids since 1998. Over the past two years, the sampling effort was standardized to 90 traps laid every two months. The first record of D. suzukii in the Cerrado biome was in December 2013, when we caught three individuals. In 2014, we collected six individuals of this species. Remarkably, in the first four collections of 2015, we have already captured 37 flies of D. suzukii, and about one-third of them came from forests. In the previous collections, all specimens were collected in savanna vegetation. This pattern of co-occurrence in savanna and forest formations is consistent with two other species of the group D. melanogaster, to which D. suzukii belongs: D. simulans and D. malerkotliana. Among the drosophilids collected from October 2013 to August 2015 in this Ecological Reserve, D. simulans (n = 6598) and D. malerkotliana (n = 3364) were found in savanna (30% and 55%, respectively) and forest vegetation (70% and 45%). Our data suggest that D. suzukii is establishing in the Brazilian Cerrado, and the impact it might have on resident populations in this biome is still unknown. This species is globally classified as a pest. Therefore, its monitoring is of the utmost importance for management and control actions in this new region of invasion.



Preliminary survey of *Drosophila* (Diptera, Drosophilidae) in an Atlantic Forest fragment near sandbanks in the Santa Catarina coast

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THE DROSOPHILA FAUNA in the Neotropical region is highly diverse and it is widely used in works about ecology, genetics and evolution. The goal of this work was to perform a preliminary survey of the occurrence of Drosophila species in one Atlantic forest fragment located near to sandbanks in the Santa Catarina coast, in Bombinhas city. The flies were collected using open traps on the ground, containing fermented banana. The specimens were captured using entomological nets and brought to the laboratory, where they were identified by means of identification keys and, when needed, examining the aedeagus. A total of 400 individuals were captured, resulting in 12 different species. Three belonged to the repleta group: D. coroica (3), D. carolinae (7) and D. mercatorum (10); three to the melanogaster group: D. melanogaster (11), D. simulans (29) e D. montium (5); two to the cardini group: D. polymorpha (93) e D. neocardini (49); one to the saltans group: D. sturtevanti (8); one to the guaramunu group: D. griseolineata (6); one to the immigrans group: D. immigrans (6); and one to the tripunctata group: D. mediopuctata (2). The Shannon diversity index was 2.145, and the Pielou's equity index was 0.792. The *repleta* group was as one of the most diverse groups in terms of the number of species (3). However, they had lower abundance compared to the other species. The higher abundance was observed to the cardini group species, with D. polymorpha (93) being the most abundant. This group is widely distributed due to its high ecological versatility, and D. polymorpha is a species that is adapted to dry environments, having high incidence in cerrado, sandbanks and inland forests. The higher abundance in this survey must be due to the proximity of the collection area to the coast sandbanks. The species diversity found in this preliminary survey stimulates further works on the biodiversity of the region sampled, and demonstrate the importance of the preservation of the local fauna.

Keywords: *Drosophila*, Biodiversity, fauna

Financial aid: CAPES and UNICENTRO



Larval nutritional requirements and female oviposition preference reflect in figs colonization for pest *Zaprionus indianus*

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SPECIES COEXIST using the same nutritional resource by partitioning it either in space or time, but few studies explore how species-specific nutritional requirements allow partitioning. Zaprionus indianus and Drosophila simulans co-exist in figs by invading the fruit at different stages of maturation; Z. indianus colonizes ripe figs, whereas D. simulans oviposits in decaying fruit. This fact provided the Z. indianus become an important pest in this monoculture in Brazil. Larvae of drosophilids feed on yeast growing on the fruit, which serves as their primary protein source. Because yeast populations increase as fruit decays, we find that ripe fruit has lower protein content than rotting fruit. Therefore, we hypothesized that Z. indianus and D. simulans larvae differ in their dietary requirements for protein. We used nutritional geometry to assess the effects of protein and carbohydrate concentration in the larval diet on life history characters in both species. Survival, development time, and ovariole number respond differently to the composition of the larval diet, with Z. indianus generally performing better across a wider range of protein concentrations. Correspondingly, we found that Z. indianus females preferred to lay eggs on low protein foods, while D. simulans females chose higher protein foods for oviposition when competing with Z. indianus. We propose the different nutritional requirements and oviposition preference of these two species allows them to temporally partition their habitat.

Keywords: Larval diet; Life-history traits; Macronutrient requirements; Nutritional geometry; Oviposition preference; Stage of ripeness/decay; Temporal partitioning Financial aid: Portugal. Fundação para a Ciência e a Tecnologia (Portugal) – M.J.A.Carvalho (SFRH/BPD/75993/2011), N.E.Martins (SFRH/BPD/62964/2009), M.J.A.Carvalho and C.K.Mirth (EXPL/BEX-BID/0497/2013). Brasil. C.Matavelli - CNPq (proc. 437 140903/2010-0) and PDSE/Capes (BEX 12281/12-3)



Parasitoid wasps collected by *Drosophila* traps in Brazilian Savanna

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A PARASITOID INSECT is an organism whose larva develops within a single arthropod host and always induces its death. Like other insects, larval and pupal stages of Drosophila species are attacked by many parasitoids. Most of them belong to the families Braconidae, Figitidae, Pteromalidae and Diapriidae. To find their hosts, first these parasitoids look for Drosophila habitat clues, like rotten odor, and then for host clues, like the odor of adults and larvae. Although some parasitoids of drosophilids have already been recorded in Brazil, little is known about them and their interactions. To investigate parasitoid wasps in the Brazilian Savanna, we used drosophilid traps with fermented banana in bimonthly collections, from October 2013 to August 2014, in the Ecological Reserve of IBGE (RECOR). The parasitoids were then classified to the species level, whenever possible. We found 124 parasitoid wasps, and 118 were identified to the genus level. The highest abundance of parasitoids occurred on February (nearly 51% of all individuals collected). In contrast, in October we did not collect any wasp in our traps. Figitidae was the main family in our samples, accounting for 90% of the wasps representing five genera: Aganaspis, Dicerataspis, Leptopilina, Nordlandiella and Tropideucoila, but we have also recorded the families Braconidae (genus Aphaereta) and Platygastridae (genus Leptacis). Up to now, three species were identified in samples: Tropideucoila weldi, Leptopilina boulardi and Dicerataspis grenadensis. We do not know exactly why these wasps entered in the traps, but they were possibly attracted by habitat and Drosophila clues. The traps attracted many drosophilids, and it is known that these genera can parasitize Drosophila or at least, Diptera. Leptopilina boulardi, in particular, is a well-studied parasitoid specialized in frugivorous Drosophila. This study indicates possible parasitoid wasps of drosophilids in the Brazilian Savanna, which may be interacting and causing selective pressures in *Drosophila* populations. Hence, more studies are required to verify host-parasitoid associations and their interactions.



High abundance of exotic drosophilids (Insecta, Diptera) in two contrasting forest ecosystems of the Cerrado biome in Paracatu, Minas Gerais, Brazil

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THE CERRADO BIOME is the most biodiverse savanna in the world mainly because of the great variety of species observed in its gallery forests. These forests occupy only 5% of the Cerrado and occur naturally as patches, but the high destruction rates of these environments have led to a further isolation of the remnants. The combined effect of habitat destruction with the typical seasonality of the Cerrado may negatively affect the biological properties of gallery forests with consequences to their biodiversity. Thus, we made collections of drosophilids in the Cerrado of Paracatu/MG to understand local patterns of biodiversity. We performed four collections between October and December 2014 in two contrasting forest ecosystems: one was a quite degraded remnant (RF) located in the center of Paracatu (17°13'S, 46°53'W) and the other a relatively preserved natural fragment (PF) situated about 10 km from the center of this town (17°15'S, 46°52'W). We sampled each forest by the exposure of five retention traps containing fermented banana. All the collected insects were sorted and identified. Furthermore, we quantified the total amount of exotic and neotropical species. Overall, we catch 4020 drosophilids representatives of 21 species of the genus Drosophila, Rhinoleucophenga, Scaptodrosophila and Zaprionus. All collections performed in RF produced results (3433 drosophilids belonging to 17 species), while in PF only two did (587 drosophilids belonging to 15 species). In both forests, the species richness of exotics virtually did not change (RF: 6; PF: 5), however they accounted for about 90% of the total abundance in RF and about 60% in PF. Although the general data show a low drosophilid species richness (only 11% of the drosophilid checklist of the Cerrado) and a dominance of exotics, such data are consistent with the diversity patterns observed for the transition between the dry and rainy seasons of the Cerrado, where environmental conditions are still stressful for the establishment of the most of drosophilids associated with gallery forests. Finally, a better description of the local biodiversity patterns may be achieved through additional taxonomic surveys.

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The importance of the Caatinga protected areas to maintain the diversity of drosophilids

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BRAZIL IS A MEGADIVERSE country, with a wide variety of fauna and flora. The Brazilian government protects natural areas through Conservation Units, and the Caatinga is the biome with the smallest protected area, despite covering 11% of the national territory. The objective of this study was to assess the potential of conservation units in the Caatinga to protect biodiversity by comparing abundance, richness and diversity of native and exotic drosophilid species inside and outside these areas. Three conservation units were evaluated in two years: Estação Ecológica Raso da Catarina (Bahia state), Parque Nacional do Catimbau (Pernambuco) and Parque Nacional Serra da Capivara (Piauí). Around 23.000 drosophilids, belonging to 32 different species were recorded in 12 samples (six inside and six outside of conservation units) reflecting the most updated list of drosophilids for the Caatinga. Twenty six species (20 natives and six exotics) were recorded inside the conservation units, while 29 species (21 natives and eight exotics) were captured outside. Samples collected inside and outside conservation units were highly similar in terms of native species composition, since over 70% of these species were observed simultaneously in these environments. The species belongs to four genera: Drosophila (the most diverse with 24 species), Rhinoleucophenga (with six species) and Scaptodrosophila and Zaprionus (one species each). Interestingly, 50% of the species of Rhinoleucophenga genus were captured exclusively inside conservation units and for three of them (R. capixabensis, R. *joaquina*, and *R. punctulata*) this is the first record in the Caatinga. At all, native species were significantly more abundant inside conservation units, while exotic ones were more abundant outside. Some species still undescribed, occurred exclusively within protected areas, stressing the importance of conservation efforts to maintain the biodiversity of native species.

Keywords: Abundância, Drosophila, região Nordeste, semiárido Financial support: FAPERGS and FACEPE



Higher diversity on samples of Drosophilidae taken with banana baited traps on Atlantic Forests then on Amazon Forests

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DROSOPHILIDAE COMMUNITIES ARE used for studying ecological processes, as well as biological indicators. For this, it is necessary to obtain knowledge about the differences among the communities dwelling on different vegetation types. In spite of increasing publications describing local and regional faunas, studies comparing different biomes are still scarce. Drosophilidae samples obtained with fermented banana baited traps, settled close to the forest floor, were analyzed to test if the communities from Atlantic Forest and Amazon Forest consistently differ on composition, abundance patterns or diversity. When traps were sampled in consecutive days, the bait was renewed between turns. For quantification of sampling effort each sampling day on one trap was considered a sample unit. All male specimens were identified by terminalia analyses, and only data from males were used. From Atlantic Forest we used data from 12 sampling campaigns, taken from 3 localities in São Paulo State (Southeastern Brazil), resulting on 944 samples. From Amazon Forest, data from 65 sampling campaigns, from 15 localities from Pará state (North Brazil), comprising 3.960 sample unities, was used. At Atlantic Forest it was obtained 29.289 males, classified among 125 morfospecies, 71 of them identified as described species. The Amazon Forest samples include 34.170 specimens, from 107 morfospecies, including 63 identified species. Ordination analyses revealed consistent differentiation between biomes, both on composition and abundance patterns. Rarefaction analyses revealed a consistent pattern of higher diversity on Atlantic Forest. The generality of those findings needs to be confirmed on studies covering the amplitude of geographical distribution of the studied biomes. Nevertheless, the patterns described are opportunities for comparison with other taxa, as well as for addressing hypothesis about the causes for the difference on numbers of detected species described here.

> Financial aid: FAPESP, PPBio, Norte Energia S.A., PIBIC/UFPA



Fifth Six Identified Drosophilidae Species Collected from Banana Baited Traps at Altamira City (PA) Neighborhood, at Xingu River Basin

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THE XINGU RIVER basin is a sampling gap from many taxa, including Drosophilidae. This work presents the Drosophilidae species list currently registered on the monitoring program of the Belo Monte Dam building project, at this area. The sampling plan is based on 8 RAPELD compound plots disposed along the Xingu River margins, at the neighborhood of Altamira city, inside a polygon measuring about 100 km on its major length. This region comprises a heterogeneous landscape, covered by anthropic fields and forest fragments, including seasonally flooded areas. Each compound plot is a rectangle measuring 10 x 1km, with 6 to 10 subplots. A subplot is a 250m length trail. Each subplots receives 4 traps, baited with banana fermented with Sacharomyces cerevisea (1%) for 36 hours, on each campaign. Each trap is used on two consecutive turns on each campaign, with the bait renewed between turns. Consecutive turns on the same trap were polled to make a sampling unit. All male specimens were identified by terminalia analyses, and only data from males was used. Two campaigns have being executed per year, since February from 2012, resulting on 7 sampling events. The actual dataset has 1.760 sampling units and 28.378 identified males. Ninety five morfospecies were detected and 56 of them were identified as described species. This species number is higher than the published lists for 22 of Brazilian states. Nevertheless, there are reasons to consider that it is far from complete: 1) 39 species remains to be identified or described; 2) The sampling method is selective, restricted to species attracted to banana baits; 3) Rarefaction analyses reveals that many species are expected to be detected on consecutive campaigns. The high number of species detected probably results from the intensive sampling effort, to the sensibility due to identification by male terminalia and to the heterogeneity of sampling areas.

Financial aid: Norte Energia S.A., Programa PIBIC/UFPA



Choice test between three different fruits by the fruit-fly Zaprionus indianus Gupta, 1970 (Diptera: Drosophilidae)

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ZAPRIONUS INDIANUS IS a kind of fly belonging to Drosophilidae family that was first found in Brazil in 1999. This fly, supposedly from Africa, was accidentally introduced in this country, becoming Ficus carica L pest, species of purple-fig widely grown in Valinhos city, SP. Since pest-species introduction, there was a significant prejudice in purple-fig production due to the fly lays its eggs in fig ostiole, making it unfit for human consumption. This led to considerable economic losses, since Brazil is one of the ten largest fig producers in the world. For that reason, it is of great interest to study the reproductive dynamics of that species with the aim of developing efficient control methods. From this premise, tests involving choice of oviposition between three different fruits have been performed. We used mango, papaya and guava, matched in pairs, in three tests, five replicas each. Those fruits were chosen due to they are often grown under the same properties or surrounding properties of fig growers, and they can be used as substrate for oviposition in purple-fig off season. Each replica had 30 adult flies, 10 males and 20 females. The fruits were exposed couple-matched, separated by a distance of 30cm. Replicas remained in climatic chambers (Eletrolab, EL 202) for a period of 10 hours, with controlled temperature and light. Then, the eggs in each fruit were counted and it was applied Z-test (Zar, 1999) to assess if there was a statistically significant preference for a particular fruit. Statistically, there was significant preference for guava compared with papaya (p value approximately zero) and also with mango $(p=1,42x10^{-5})$. In the test with just papaya and mango, significant preference of females was for mango (p = 9,96x10-5). Considering those information, it was possible to better understand the species feeding preference and thus enable more-efficient management techniques in population control of this fly. Further the control of this species in purple-fig, our findings suggest attention of producers to possible infestation of guava by this pest species, especially in fig-purple off season.

Keywords: fruit-fly; oviposition behavior; choice test; guava Financial aid: CNPq



Relative humidity influencing on emergence of Zaprionus indianus Gupta, 1970 (Diptera: Drosophilidae)

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THE AFRICAN FLY Zaprionus indianus was accidentally introduced in Brazil and became a pest of purple-fig because females lay their eggs in the fig ostiole, preventing the human consumption. Many studies have been done to better understand biological aspects of this species. Taking into account the flexibility of this fly in occupying several niches with different environmental conditions, it is important to learn more about the interference of abiotic factors, such as relative humidity (R.H), on its life-cycle. Besides, it is interesting to know if its fitness traits are related to different responses to stressful conditions. Thus, 300 one-day pupae were collected and each one was weighed and had its length and width measured. After, they were individualized and divided into three treatments: control (R.H. 55±5%), high relative humidity (R.H. 80±5%), and low relative humidity (R.H. 30±5%) groups. Each vial with respective pupae was observed every day and were also checked the sex of emerged adults and the period of pupation for each specimen. In control group, 74% emerged, 86% in high R.H. group and only 45% emerged in low R.H. group. Chi-square test, used to compare proportions, showed that low R.H. group was significant different in relation to the two others. Using Kruskal-Wallis test, no difference was detected in period of pupation between groups and between sexes. About biometric parameters, emergence was independent of pupae trait values in all treatments. The role of abiotic factors, such as relative humidity, in life history of fruit flies was already reported in previous studies, so this present project confirmed the influence of humidity in the biology of one more fruit fly species. According to the literature, it is common the occurrence of differences in period of pupation between sexes in many drosophilids, because females emerge earlier than males. The opposite is also found in other groups. The results related to biometric parameters corroborate those of other authors. Considering that, in this experiment, larvae were feed with the same food, the variation on biometric parameters found in pupae may be due to the quantity of food that each larva ingested and assimilated. However, further studies are necessary to better understand the effects of environmental conditions on biological aspects of this pest.



New surveys in the study of chromosomal polymorphism of Drosophila willistoni populations collected in Bahia and Pernambuco states, Brazil

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DROSOPHILA WILLISTONI STANDS out for presenting a high level of chromosomal reorganization, easily viewed through the presence of paracentric inversions in their polytene chromosome. The characterization of this genetic pattern among geographic populations has been studied for several decades. Rohde and Valente (2012) made an extensive photographic record 50 different rearrangements among 30 locals, and set their breakpoints on the fotomapa the species. Among the rearrangements described by the authors, there is XL-E, present in the left arm of chromosome X and different from fotomapa arrangement of the species D. willistoni (XL-A) for only an inversion (inv. E). In this study we analyzed the pattern of chromosomal banding and frequency of chromosomal inversions in D. willistoni of Biological Reserve Saltinho (SAL), in Pernambuco and a private forest in the municipality of Jiquiricá (JEQ) in Bahia, two Atlantic Forest fragments of Northeastern Brazil. The findings showed low frequency of chromosomal inversions in both populations studied. SAL and JEQ were different in relation to the XL chromosome, with the presence of XL-E rearrangement only in JEQ. Previous reports done by our research group indicate that populations of Pernambuco are monomorphic for XL rearrangements, whose order is identical to the banding pattern of chromosome of photomap of the species (XL-A), considered the most ancient. However, the identification of the XL-E variant in JEQ suggests that this local is inserted in a border area of genetic variability for the species, when compared to other Northeastern states such as Pernambuco. This genetic variation observed in the XL of D. willistoni meets with the biogeographical and ecological diversity hypothesis of the Atlantic Forest, situated north and south of the São Francisco River, which is the key to understanding the evolution of Amazonian and Atlantic biota. In the area of Atlantic Forest at north of São Francisco would have occurred biotic exchanges between these two major regions of South American forests, during the Cenozoic. The chromosomal results observed to date support this ecological differentiation and point to the need to expand the samples in the state of Bahia.

Keywords: Diversidade; Nordeste do Brasil; Floresta Atlântica; inversões cromossômicas Financial support: CNPq, FACEPE, PROPESQ-UFPE



Natural diet causes pronounced wing shape asymmetry compared to seasonal diet in *Drosophila mojavensis* (Diptera: Drosophilidae)

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FLIES OF THE DROSOPHILA mojavensis sonorensis subspecies feed and lay eggs on necrotic tissues of the columnar cactus Stenocereus thurberi. However, when Opuntia fruits (tunas) are seasonably available they are used by this subspecies as an alternative host. Knowing that tunas have much more sugar than the columnar cactus, a question arises from this observation: do this seasonal host change generates an environmental stress on the larval development? The level of asymmetry is one way to evaluate the effect of this seasonal change of the diet over this subspecies. Thus, the goal of this work was to verify the relationship between larval development and wing asymmetry of females from one D. m. sonorensis isofemale line collected in Las Bocas, state of Sonora, Mexico. Right and left wings of 200 females, half developed in S. thurberi seminatural medium and half developed in tuna semi-natural medium, were removed, rinsed in 70% alcohol, placed on slides and photographed. Twelve type I landmarks were marked using the TPSDig 1.40 software and the asymmetry analysis was performed in the MorphoJ 1.06 and Past 3.08 softwares, considering cactus and tuna separately and also together, with diet as a additional factor. For size, no significant difference between sides (p>0.05) was observed in all comparisons. Significant differences (p<0.001) for size were found for individuals and individual/side interaction, in all comparisons. For shape, significant differences (p<0.001) were detected in all analyses and all comparisons, including the difference between sides, with tuna apparently affecting less the asymmetry than cactus (evidenced by the lower F values). These results demonstrated that both diets generated shape asymmetry in the females analyzed, and the natural host seems to be slightly more stressful during larval development than tuna, despite tuna being a seasonal alternative host with much higher sugar content.

Keywords: *Drosophila mojavensis* sonorensis, Adaptation, Asymmetry, Host Seasonality Financial aid: CAPES, FINEP and UNICENTRO



Preliminary study on the use of fruits as breeding sites by drosophilids (Insecta, Diptera) in urban environments in Northeastern Brazil

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ONE OF THE MOST important issues for the understanding of how a community is structured in nature is the knowledge of the ecological niche occupied by their species. This is the starting point for the elucidation of how diversity is maintained and how species coexist. In the present study we evaluated different trophic niches occupied by flies of the family Drosophilidae through sampling fruit rotting in six urban areas located in the states of Pernambuco and Paraíba, Brazil. During the years 2014 and 2015 were collected fruit of Malpighia emarginata, Eugenia uniflora (cherry), Spondias mombin and Psidium guajava from which emerged 641 flies. In order of abundance were collected the following species Drosophila malerkotliana, Zaprionus indianus, D. ananassae, D. melanogaster, D. simulans, D. willistoni, D. nebulosa and D. fumipennis. The first five are exotic species and accounted for over 90% of the sample. Exotic species emerged of all investigated fruits, showing the great ecological versatility of these species. In all trophic resources investigated D. malerkotliana was the most abundant species. This finding corroborates that seen in other studies in urban areas in the state of Pernambuco with drosophilids capture in traps containing baited traps. Among the native species of the Neotropical region, only D. nebulosa emerged of all trophic resources investigated. Drosophila willistoni used all resources except Psidium guajava as breeding sites. Drosophila fumipennis just emerged of Spondias *mombin* suggesting that this is a more specialist drosophilid in relation to fruit investigated.



Temporal behavior of drosophilids in a grassland area in the South of Brazil

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THE PAMPAS BIOME is a Subtropical grassland region presenting striking climatic characteristics, with negative records of temperature in the winter up to 40°C during the summer. Samples of drosophilids were taken in natural and urban areas during 12 climatic periods. The presence and relative abundance of exotic and Neotropical species in the composition of Drosophilidae assemblages were temporally and spatially analyzed through nMDS and PERMANOVA tests. The assemblages were identified temporally, as well as the relative roles of temperature, rainfall and type of environment to the structuration of the assemblages. The following hypothesis was tested: (i) periods of high and low temperatures are important to the maintenance of local diversity of Drosophilidae in pampas. During stressing climatic conditions the assemblages were negatively affected, except the assemblage from the inner of forest. Temperature was the component more related to the assemblage variation followed by the kind of environment. The reasons for this influence include local environmental variants, such as temperature and resources availability, but possibly also evolutionary characteristics of each species. Thus the conservation of the pampas biodiversity depends on the conservation of natural environments and the ability of species to develop survival strategies to face the changes in the global climate conditions.

Keywords: Pampa; Drosophilidae; temperature

Financial aid: CNPq, PRONEX-FAPERGS (10/0028-7), CAPES



An interactive key for identification of common drosophilid species in the Brazilian Savanna

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THE FAMILY DROSOPHILIDAE currently includes more than 4.000 species distributed in 75 genera. These flies reveal unique morphological and ecological diversity among Muscomorphans, but at the same time there are groups of species that are pretty similar. The study of the neotropical communities, in particular, faces a big difficulty: the lack of identification keys for these species. In this study, we developed an interactive key for identification of the most common drosophilid species from the Brazilian Savanna, aiming to help the identification process and reduce the time consumed for this task. The key was developed based on external and internal morphological characteristics of the flies: head, thorax, abdomen and male terminalia. We then developed a software and a web application using programming languages and technologies like Java, Javascript, HTML and CSS. Therefore, the application design is user-friendly, with an autoexplained interface. The user has only to choose some characters, among many, to proceed with the identification of the focal specimen. As more characters are chosen, more precise will be the identification. Then, based on the user entries, the software will properly evaluate and return the result. Therefore, the software design excludes unnecessary steps of identification turns, making this task faster than using a common key. Currently, the key supports eight species: Drosophila atrata, D. cardini, D. nebulosa, D. mercatorum, D. polymorpha, D. prosaltans, D. sturtevanti and D. willistoni. Nevertheless, the software/application will be in constant upgrading, allowing more than 100 species and 200 characters in the database. In sum, this highly portable software, able to be used on most of computers, will certainly provide new scenery for flies' identification, by making this task easier for people with less identification experience.



Drosophilidae communities at Araucaria Moist Forest: changes in composition and diversity through time

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THE ECOREGION OF ARAUCARIA Moist Forest is located in southern Brazil and in the Argentinean province of Misiones. The diversity of Drosophilidae is still poorly known in this ecoregion, which is one of the most endangered of the Atlantic Forest biome. The Iguacu National Park, in the Brazilian state of Paraná, along with natural areas in Argentina, is one of the largest continuous forest remnants of the biome. Samples were taken in the Araucaria Moist Forest on Iguaçu National Park, during two seasons: summer of 2013 and autumn of 2014. Twenty bananabaited traps were placed along a 2000 m transect in Manoel Gomes Trail, each of them 100 m apart. The traps were settled from the edge to the center of the forest, decreasing in altitude (from 627 to 508 meters). Six environmental components were measured: altitude, temperature, humidity, rainfall, canopy cover and understory density. Specimens sampled were identified by general morphology. Overall, 951 drosophilids were collected, 197 of them in the summer and 754 in the autumn sample. In summer, 10 morphospecies were found, distributed in two genera: Drosophila and Hirtodrosophila. The Drosophila groups found were annulimana, canalinea, cardini, guarani, repleta, tripunctata and willistoni. All the taxa found were endemic to Neotropics. The most abundant were: D. sgr. willistoni (66,4%), D. polymorpha (13,1%), D. gr. repleta (9,1%) and D. ornatifrons (7,1%). On the other hand, 20 morphospecies were found in autumn, distributed in three genera: Drosophila, Zaprionus and Zygothrica. Two exotic species were found, but at very low abundances: Zaprionus indianus (0,1%) and Drosophila malerkotliana (0,1%). The most common taxa were: D. gr. tripunctata (45,8%), D. sgr. willistoni (17,7%), D. polymorpha (15,7%) and D. gr. guarani (12,8%). Correlation tests showed that richness was higher in the points of the forest of lower altitudes and higher understory density. In addition, the abundance, richness and Shannon diversity index were significantly higher in the autumn when compared to the summer samples.



First occurrence register of *Drosophila boraceia* (Diptera, drosophilidae) in Santa Catarina State, Brazil

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DROSOPHILA BORACEIA AT first inspection looks like a species belonging to the fasciola subgroup, of repleta group, because of the *fasciola*-like pattern of fused spots on the thorax, quite distinct structures of the male terminalia. However, in some structures of the male's terminalia, D. boraceia seems more similar to one ungrouped and also spot-thoraxed species, D. atalaia. D. boraceia belongs to the subgenus Drosophila, but can't be assigned to any recognize groups of genus Drosophila. Was proposed another new group, named peruensis group, which included D. atalaia, D.pauliceia, D.boraceia, D. peruensis, with diagnostic characters including: small flies, with most setae and setulae of the thorax and head arising from dark brown spots, which may be somewhat fused; wings with both main cross veins slightly or strongly darker than longitudinal ones, except in *D. atalaia*; surstylus mostly microtrichose; hypandrium somewhat square-shaped; aedeagus stout, bearing a pair of ventral, subapical spurs; paraphysis somewhat square-shaped. Oviscapt bearing both peg-shaped and trichoid-shaped ovisensilla, the distalmost discal one being longer than proximal ones, except in D. atalaia. Mostly inhabit the wet Atlantic Rainforest biome. The specimen of D. boraceia was collected in Atlantic Rainforest at Parque Estadual da Serra do Tabuleiro (27º48'20'' S, 48º33'50'' W) along with: D. willistoni, D. capricorni, D. mediopunctata, D. mediopicta, D. bandeirantorum, D. nappae, D. paraguaiensis, D. guaru, D. polymorpha, D. annulimana, D. ararama, Scaptodrosophila latifasciaeformis. Here we are widening the geographical distribution of this species to a more austral point in the Neotropical Brazilian territory.

Keywords: Drosophilidae, Atlantic Rainforest, austral distribution, Serra do Tabuleiro



Morphological characterization of six common *Drosophila* species in the Brazilian Savanna

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THE DROSOPHILIDS FROM the Brazilian Savanna have been studied since 1998, by the Laboratório de Biologia Evolutiva of the Universidade de Brasília (LBE), and there are currently 123 species recorded in this biome. In all, 110 of these species are of Neotropical origin, and much of this fauna is difficult to recognize due to the lack of identification keys. In this study, we characterize six neotropical species common in this biome: Drosophila willistoni, D. cardini, D. stutervanti, D. mecatorum, D. nebulosa, D. polymorpha, D. prosaltans. We analyzed specimens maintained in the collections of the LBE, collected at different localities in the Brazilian Savanna. Through a stereomicroscope integrated with a camera, we generate sets of pictures for each specimen, by focusing on a given structure at different depths. Next, we digitally stacked the sets of photomicrographs to create an all-in-focus composite (open source software CombineZP at http://www.hadleyweb.pwp.blueyonder.co.uk/CZP/Installation.htm). The images were then edited in the software Adobe Photoshop CS6 and Adobe Photoshop Lightroom 5, to improve the accuracy of the resolution. For each species, we prepared an illustrated sheet showing the major diagnostic characteristics: costal index, number of branches in arista, mesonotum and abdomen pattern, body color and setae (vibrissae, orbital, pleural, acrostichal, dorso-central, prescutellar and scutellar). The images, combined with morphological data, could be a practical way to identify these species, helping not only to identify drosophilids in the Brazilian Savanna, but also in other Brazilian biomes. The idea of this project is to build a taxonomic net that helps researchers to understand, identify and compare different species in a fast and online way. Therefore, we suggest that this study could be joined with similar initiatives in progress in other regions of Brazil, aiming a cohesive compendium of Neotropical drosophilids identification.


Drosophilidae demographic structure from Pampa to Atlantic Forest biomes in Rio Grande do Sul, Brazil

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ALTHOUGH MANY SURVEYS have been carried out in Rio Grande do Sul, the state's northern region remains with few studies. Our study aims to compare the data obtained previously on drosophilids assemblages of Rio Grande do Sul in Pampa Biome, and in a transition region between the Pampa and Atlantic Forest with our sample in a Atlantic Forest fragment, the Parque Estadual do Turvo (PET) to highlight the distribution of Drosophilidae species. Flies were collected using 20 standard traps along one transect. Specimens were separated using morphological characters. In the transition area were reported 5,007 individuals of 6 genus, Drosophila, Zaprionus, Zygothrica, Amiota, Leucophenga and Rhinoneucophenga and 15 Drosophila groups, with Drosophila melanogaster group being the most abundant, followed by D. guarani, D. tripunctata, D. cardini and D.repleta (44.86%, 16.54%, 10.99%, 9.99% e 6.78%) respectively). D. willistoni, D. immigrans, D. pallidipennis, D. busckii groups and Zaprionus genus did not reached 3% each. D. bromeliae and D.anulimana groups, and Zygothrica, Amiota, Leucophenga and Rhinoleucophenga genus did not reached 1% each. On Pampa were sampled 762 individuals from 5 genus (Drosophila, Amiota, Zaprionus, Zygothrica, Rhinoleucophenga) and 11 Drosophila groups, D. repleta and D. melanogaster groups (50.26% and 40.49% respectively) were the most abundant. D. willistoni group with Zaprionus and Rhinoleucophenga genus did not reached 3% each one. D. tripunctata, saltans, cardini, guarani groups, as well as Amiota and Zygothrica genus did not reached 1% each one. In our collect, in the PET, 1,210 individuals were caught only one genus with 9 different species groups. The groups D. willistoni, D. tripunctata, D. saltans and D. guarani (45.46%, 27.85%, 16.53% e 5.78%, respectively) were the most representatives, D. repleta, D. cardini, D. melanogaster, D. annulimana and D. immigrans groups together did not reached 5%. As expected the transition region presented most richness in relation to PET and the Pampa Biome. Analyzing the species composition, the PET seems to be the more preserved environment, once we could find only autoctones species groups. Although these are preliminary data, they are a brief drafting of whole biodiversity of the region that can be used to subsidies to conservation planning.

Keywords: Pampa, Atlantic Forest, Assemblage Diversity, Demographic Structure, Conservation



Differences on *Drosophila* species group attracted by fallen fruits and banana baits

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IN MARCH 2015, we found fallen fruits of Annona sp. (Annonaceae) in the Parque Nacional do Itatiaia, RJ, Brazil (PNI). Then we collected Drosophila specimens by sweeping entomological nets over the fruits and compared them to the flies attracted to banana baits, set up at about two meters, during four days. After that, we brought the collected flies to laboratory to be identified by their external morphology. We classified Drosophila specimens into five groups: tripunctata (TRP), calloptera (CAL), cardini (CAR), guarani-annulimana (GUA) and Sophophora (SPH). We also brought the fruits to our lab and confirmed the occurrence of Annona sp. in PNI searching for gatherings via speciesLink. Besides, we also isolated yeast colonies from Annona sp. fruits. First, we homogenized 25g from different parts of the fruit on 225ml of 0.1% protonated water using Stomacher 400 blender at high speed for two minutes. Then, we streaked colonies in YPD medium (1.0% yeast extract; 2.0% glucose and peptone; pH 6.5; with 1.0% Cloramphenicol) and incubated them at 30°C for 48 hours. We identified the yeast species by submitting sequences of D1-D2 domain of the large-subunit rRNA gene to GenBank database and comparing them to entries for yeast. We collected 3509 specimens over the fruit (SPH = 90.6%; GUA = 3.9%; TRP = 3.4%; CAR = 1.9%; CAL = 0.2%) and 6850 specimens over the baits (SPH = 87.3%; TRP = 8.5%; GUA = 1.8%; CAR = 2.0%; CAL = 0.4%). We collected on the first day 56 tripunctata specimens over fruit - 13.2% of the sample. However, after the second day, tripunctata percentage collected over the fruit plunged to less than 4.0%, while the number of these specimens collected over banana baits were at least four times higher. In several studies, species from tripunctata group were mostly attracted to baits inoculated with Saccharomyces cerevisiae. The only yeast we isolated from Annona sp. fruit was Kazachstania exigua. We interpreted this pattern as a clear preference of flies from tripunctata species for S. cerevisiae. We intend to investigate the preference behavior of different Drosophila species to yeast, as well as the influence of different substrates in baits attractiveness.

Keywords: Behavior, Food preferences, Choices, Itatiaia, Yeast Financial aid: CAPES, CNPq, FAEPEX-UNICAMP, FAPESP, FAPEMIG



Temporal and micro spatial dynamic of the Drosophilidae assemblage in local scale in a Restinga forest in the southern Brazil

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ASSEMBLAGES HAVE DIFFERENT responses face ecological parameters and are influence by temperature, humid, rainfall and habitat size, for example. In this sense, we verify the temporal and micro spatial influence in a Drosophilidae assemblage in a 23ha Restinga forest, in the southern Brazil. The individuals were monthly sampled from Feb 2013 to Jan 2014 using, in each month, 12 traps baited with banana and yeast. The traps were spaced 60m from one each other and were distributed in a trail, with four traps close to the edge and eight in the interior of the forest. A Canonical Correspondence Analysis (CCA) tested the temporal variation and the influence of the maximum temperature, minimum temperature, rainfall and humidity in the assemblage. The influence of the position of the traps were analyzed in each month by Mantel tests and, to performed it, the traps were UTM georeferenced and the distances between all the traps were calculated by Euclidian distance analyses. It was sampled 25,093 drosophilids from 46 species. The CCA stablished a gradient determined by the maximum and minimum temperature, were the samples of Mar, Apr, May, Nov, Dec and Jan, with higher temperature, had the dominance of D. simulans, D. willistoni and D. polymorpha, and the samples of Feb, Jun, Oct, Jul, Aug and Sep had higher abundance of D. mediopunctata, D. paraguayensis and D. griseolineata. The Mantel test suggests that the trap position do not influence the assemblage, even when it was used the dissimilarity index of Jaccard or Morisita-Horn (0.4<p<0.8 for all comparisons). Our results suggest that the assemblage changes in a time scale more than a micro spatial scale.



The decaying stage of *Psidium guajava* (guava) fruits as a determining of the guild of Drosophilidae (Insecta, Diptera)

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SPECIES CAN SHARE trophic resources and should be susceptible to competitive exclusion, depending on their success in exploiting it. Most species of Drosophilidae oviposit in decayed fruit, and a way to avoid competition is to use the resource at different stages of decomposition. Describing when the colonization occurs in the available resources helps us to understand the species interactions. This study describes the guild of Drosophilidae associated to guava in ripe fruits on the plant and fruits decaying on the ground. The guavas were collected in a site of the UFPel (31°48'S; 52°43'W). A total of 1,539 individuals emerged from the fruits; 843 from fruits on the plant and 696 from fruits on the ground. Of the 23 fruits collected on the plant, 70% were colonized and of the 25 from the ground, 88% were colonized. The species recorded were Drosophila suzukii, D. willistoni, D. simulans, D. melanogaster, Zaprionus indianus, D. griseolineata, D. ananassae, D. paraguayensis and D. polymorpha. The richness were superior in guavas on the ground (all nine species), while from the fruits on the plant emerged only the four former species. Drosophila suzukii reached 89% of the guild and was dominant in fruits from both sampling, however, its incidence were superior in fruits on the plant, probably because it is the only Drosophilidae in Brazil that can pierce the fruit. It is assumed that other species that colonized the fruits on the plant have taken advantage of fruits already drilled or open otherwise. The exotic species presents high capacity of use of resources in different stages than the native D. *willistoni*. The species that use the fruits on the plant must have competitive advantage, because the colonization happens earlier than in other species. Further, the colonization of the fruits for these species should change the resource physical and chemically.



Evaluation of net sweeping as a sampling method of drosophilids in the National Park of Iguaçu, Paraná, Brasil

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THE METHODS OF sampling Drosophilidae generally are based on the use of baited traps, mostly banana. Although they are very effective, they presuppose that the flies are attracted by this type of food. Those species not attracted to the bait cannot be detected even when present in the site studied. The present study proposes to evaluate the method of net sweeping for sampling Drosophilidae as a complementary method. An entomological net of 14 centimeters of diameter was used, applying it in the forest vegetation along a distance of 100 m for each sample. The procedure was repeated 20 times in a transect of 2,000 m in Manoel Gomes trail, in the National Park of Iguaçu, Céu Azul, Paraná, Brazil. Sample of adults (with net) and preadults (with emergence) on resources (fungi and fruits) found on the same transect were also collected. A total of 705 individuals were analyzed, belonging to the genera Zygothrica, Hirtodrosophila, Diathoneura and Drosophila (in this last case the groups tripunctata, willistoni, calloptera, guarani, repleta, canalinea and cardini were identified). The 17,7% of the individuals were sampled with net sweeping in the forest, 9,9% as preadults in fungi and 62,1% in fruits, and 6,6% and 3,7% as adults on fungi and fruits, respectively. The richness was higher with sampling with net sweeping: nine of the eleven identified groups were found with this method, while three and two groups were found breeding on mushrooms and fruits, respectively, and five and six were found as adults in fruits and fungi, respectively. The results suggest that, concerning to richness, the net sweeping is more effective than the sampling directly on breeding or feeding sites. Although the method can be influenced by factors such as the time of sampling, future comparisons with samples of baited traps will evaluate the effectiveness of net sweeping as a potential additional method of sampling, without the bias produced by the bait of the traps.

Keywords: Atlantic Forest, diversity inventory, trophic resources

Financial aid: Fundação Araucária, PPBio Mata Atlântica



Seasonal variations in abundance and richness of drosophilids (Insecta, Diptera) in Atlantic Forest environments in Northeastern Brazil

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SEASONALITY IS AN IMPORTANT aspect related to the insect survival strategy, it is of fundamental importance for the understanding of population dynamics and community structure. Seasonal patterns may affect relative abundance of individuals, reproductive activity and the availability of food resources. In the present study we evaluated the seasonal patterns in abundance and richness of native and exotic species of drosophilids in Brazilian Atlantic forest fragments in the north of the São Francisco River, in the Pernambuco sub region. In this area well defined seasonal variations occur between wettest and driest periods. Three forest fragments were studied: the Ecological Station Tapacurá (TAP), the Experimental Station Itapirema (ITA) and the Charles Darwin Ecological Refuge (DAR). In each area two samples were taken at the highest rainfall period and two in the drier period. Ten traps were used with banana bait in each collection, spaced 30 meters from one another. In total, 40,911 drosophilids were collected belonging to 36 species. In DAR were observed 27 species (22 collected in the dry season and 20 in the rainy), followed by TAP with 25 (18 in the dry season and 25 in the wet) and ITA with 22 (17 in the dry season and 20 in the rainy). In all forest fragments there was a greater abundance of drosophilids in the dry season, in ITA the abundance of drosophilids in this season was approximately 90% compared to the rainy season, in DAR was over 83% and in TAP was almost 70%. The abundance of exotic species was higher in the dry season in the three areas surveyed. Except for TAP, the abundance of native species was also higher in the dry season; however, this difference was not as pronounced as that observed for the exotic between stations. During the rainy season the native species were always more abundant than the exotic. Drosophila malerkotliana was the most abundant exotic species in all locations and D. willistoni and D. sturtevanti stood out among the native.

> Financial aid: FACEPE/CAPES/CNPq



Drosophilidae mycophagous in the Brazilian Amazon and their hosts

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UNDERSTAND THE INTERACTION processes is not an easy task, given the complexity of associated relations. These are not just controlled by variations in relation to age and size of organisms, by its position in the environment or determining environmental factors, but are also conditioned by the genetic variation within populations, which controls large part of dynamics of interactions between species. To understand the processes of interaction, primarily the interagents communities must be characterized and tested the nature of the relationship for signs of evolutionary processes and co-evolution between them, then test appropriate methods for pattern detection in species sets, thus identifying processes capable of generating certain standards. Throughout the evolution, species can suffer different types of interactions, such as parasitism in the strict sense to symbiotic relationships. And in this context are found the interactions between Drosophilidae and its resources. This study aimed to investigate the Drosophilidae community associated with fungi in Caxiuanã National Forest (FLONA), State of Para in the Brazilian Amazon. A total of 161 samples of fungi was collected, distributed in 64 species and morphospecies, belonging to nine orders. These samples were found to Drosophilidae 5124 individuals, distributed in 55 species and morphospecies. Among the drosophilids the Zygothrica genus has presented the greatest richness, with 26 species, followed by *Hirtodrosophila* genus with 19. With respect to fungi, the Polyporaceae family was the one that endured richer Drosophilidae, with 35 species. The distribution of abundance of drosophilids Guild was adjusted by the model geometric series, which describes a community with predominance of dominant species. The richness estimators approximated have estimated richness 80 species of mycophagous drosophilids for FLONA. Richness Drosophilidae is positively related to the fungus wealth, though not with its abundance. The guild Drosophilidae to FLONA Caxiuanã is characterized by species group Zygothrica genus, lineage grouping species with mycophagous habits for the Neotropical region.

Keywords: Mycophagos, Zygothrica, Hirtodrosophila, FLONA of Caxiuanã



Structure Drosophilidae-Fungi Interaction Network in the Amazon

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THE INTERACTIONS OF network structure has important implications for the coexistence of species and the stability of communities and their understanding helps in understanding the coevolutionary processes. The analysis of ecological networks seeks more than understand the structure of interaction between two distinct assemblages. They let describing the role of each organism in the community, and are important tools for understanding the patterns and processes that maintain diversity in natural communities. This study aimed to investigate the structure of Drosophilidae-Fungi interaction networked communities in the Amazon region. Were built quantitative and qualitative interaction matrices and created probability matrices to determine if the network structure is determined by the occurrence, abundance, phenology of species and possible combinations. To this were calculated metrics and each containing factor combinations. The results showed that the abundance of interactions was best predicted interactions between pairs. The structure of Drosophilidae-Fungi network was determined by models of abundance and phenology. The results corroborate other works on mutualistic networks, which had abundance as a predictor of interaction in networks. The interactions between macroscopic fungi and Drosophilidae can be structured by other biologic factors not included in the study, but the abundance is a neutral factor that helps to structure the interaction as well as the phenology. The pattern of interaction Drosophilidae-Fungi network is the nested type. This is the standard usually found in mutualistic networks, indicating that this may be the predominant type of relationship between these organism.

Keywords: Complex networks, Abundance, Probabilistic models



First registry of Paracacoxenus guttatus at neotropical region

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AMONGST DIPTERA, DROSOPHILIDAE is one of the largest families including almost 3000 species distributed on two sister-group subfamilies: Steganinae and Drosophilinae, being Steganinae more basal group of these two due their highest number of plesiomorphic characteristics in comparison with Drosophilinae. The description of Cacoxenus subgenre, a later described subgenre Paracacoxenus, and currently fusion state of these two groups came by a long and complex historic that include numerous replacements and synonymous. Today, both genres were taken as synonymous and currently placed as sister-group of Amiota and Gitona. Paracacoxenus guttatus distribution follows the presence of blister rust fungus, without exclusivity among fungal species and *P. guttatus*. Those fungus are commonly found in association with guava trees near our gathering spots. In one of periodical gathering at Ribeirão da Ilha, at Parque Estadual da Serra do Tabuleiro in Florianopolis, Santa Catarina state, we found only three male of *P. guttatus* beside some *Megaselia spp.*, a known genre considered by other authors associated to rust fungus, and some commonly found species of Drosophila sp. in the collecting point: D. polymorpha, D. cardinoides, D. capricorni, D. willistoni and a bunch of species of tripunctata goup. This is the first neotropical record of *Paracacoxenus guttatus* and such generates several discussions about its new distribution and their consequences to natives populations in the Atlantic Rainforest area.

Keywords: *Paracacoxenus*, Atlantic Rainforest, neotropic



Twenty-three confirmations and eleven new registers of Drosophilidae in Minas Gerais: just starting to fill the gap

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DROSOPHILIDAE HAS A historical significance as a model organism in the development of genetics and ecology. It is a highly diversified group, especially in the tropics, providing great opportunities to study its ecological and evolutionary dynamics in such environments. However, the sampling efforts were concentrated in some Brazilian states, resulting in gaps in knowledge and underestimates of the diversity of these insects. With this work, which is currently in progress in our lab, we aim to expand our understanding of the diversity of Drosophilid in Minas Gerais state. We sampled the flies between January and September of 2015 using baits and sampling adults attracted to natural substrates, and identified them by the morphology of the male genitalia. We concentrated our sample efforts in Vicosa but collected flies from Araponga, Catas Altas, Juiz de Fora, Lavras Novas, Rio Pomba and Tiradentes. All the studied specimens were preserved in ethanol, and their genitalia, if removed, were stored in microtubes with glycerin. Until now, we identified 23 Drosophilidae species, from which 11 are new records for Minas Gerais: 10 Neotropical species: Drosophila arauna, D. guaru, D. mediosignata, D. neosaltans, D. quadrum, D. roehrae, D. trapeza, Zygotrhica dispar, Z. orbitalis, Z. vittinubila; and one is an invasive species with economic significance, D. suzukii. Our results raise to 72 the number of recorded species in MG. This work is the first step to establishing a research line regarding the ecology and evolution of tropical Drosophilidae in our research group.

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Genética

Genetics



Application of the comet assay genotoxic test in *Drosophila melanogaster* larvae exposed to 4-nitro-pyrimidine compound

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DROSOPHILA MELANOGASTER IS widely studied in genetics, but recently has emerged as an alternative organism for studies in toxicology and mutagenicity, substitute for the use of mammals in toxic genetic tests. Due to its easy cultivation and manipulation the species has been used in the comet assay methodology that seeks to recognize simple or double breaks in DNA after exposure of individuals to the compounds of interest. Rings of pyrimidine derivatives have shown important biological properties, including anti-inflammatory activity. The aim of this study was to evaluate the possible genotoxic effect of 4-nitro-pyrimidine compound in somatic cells of D. melanogaster by comet assay. Larvae from Oregon-R strain were submitted to the compound at concentrations 0.39, 0.78, 1.56 and 3.12 mg/mL. It was also prepared a negative group, treated just with the solvent, and one positive control group subject to cyclophosphamide (1mg/mL). ANOVA and Bonferroni tests were applied to the results. After collecting the hemolymph of 180 larvae per treatment, the hemocytes were superimposed on slides covered with agarose for cell lysis and subjected to electrophoresis. During the microscopic analysis the damages were classified into five categories, with 0 being intact nucleoids and the others ranging from minimal damage to most (1-4). The statistical results indicate no significant differences between the treatments and to the negative control group, what means that the different concentrations of 4-nitro-pyrimidine compound is not genotoxic in the experimental conditions applied. The results point to use co compound as a promising drug safe for human health.

Keywords: comet assay, genotoxicity, pyrimidine

Financial aid: FACEPE, CNPq and PROPESQ-UFPE



Compound 3-nitrophenyl-pyrimidin is not genotoxic in somatic cells of *Drosophila melanogaster* evaluated by SMART test

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DROSOPHILA MELANOGASTER IS an organism which allows the application of the genotoxic SMART methodology (Somatic Mutation and Recombination Test). This test is based on the identification of phenotypes (flr^3 and/or mwh) mutants which represent the occurrence of phenotypic expression of the mutation (chromosomal or genic) and/or recombination caused by exposure of larvae to compounds of interest. The application of the SMART test reaches groups of cells, the larval imaginal discs, which proliferate separately during the metamorphosis and will originate the adult cells and body structures, such as wings. The purpose of this study was to investigate the genotoxic effect of pirimidinic 3-nitro-phenyl-pyrimidine compound, a pyrimidine base analog recognized as an anti-inflammatory substance. Drosophila melanogaster larvae resulting from the standard cross (flr^3 females with malesmwh) were subjected to different concentrations of the 3-nitrophenil-pirimidine: 0.39 mg/mL, 0.78 mg/mL, 1.56 mg/mL and 3.12 mg/mL. It was also prepared a negative group, treated just with the solvent. After treatment, the cells of the wings of the adults were examined with optical microscopy at 400x. The results of microscopic analysis and quantification of phenotypic alterations present in trichomesof the wings of the trans-heterozygous adults treated with 3-nitro-phenyl-pyrimidine were not statistically different from those shown in the negative control. Therefore it can be said that, under the used experimental conditions, 3-nitro-phenyl-pyrimidine has no mutagenic or direct recombinogenic action. This result supports the idea already presented in the literature that the 4aminopirimidynic compounds have low toxicity and are promising anti-inflammatory prototype molecules, showing no effects related to the induction of point mutation and / or somatic recombination.

Keywords: somatic mutation, human health, pyrimidine compound model organism Financial aid: FACEPE, CNPq andPROPESQ-UFPE



Transferability of SRR primers developed for *D. mediopunctata* to the species *D. sturtevanti*

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DROSOPHILA STURTEVANTI BELONGS to the sturtevanti subgroup of the saltans group. It presents a wide geographic distribution, occurring from Mexico to southern Brazil, including the Caribbean islands. Due to the high cost of developing microsatellite markers (SSR) for each species, the transferability of SSR primers between related species have been tested and used in population and evolutionary studies. The aim of this work was to analyze the transferability of SSR primers, originally described for D. mediopunctata to D. sturtevanti. Twenty primers that were successfully amplified using a sample composed of a pool of individuals from an isofemale line of D. sturtevanti, were selected. The amplifications were performed under the same conditions and reagent concentrations reported in the literature, using individuals of D. sturtevanti collected in Matão/SP, Nova Granada/SP and João Pessoa/PB. Only two loci amplified in 15 individuals out of 45 tested, even after applying protocol modifications to increase the transferability success. The optimal annealing temperatures for these two loci were tested. However, no pattern was obtained. Despite the original amplification of the 20 loci using a pool sample of one isofemale line of *D. sturtevanti*, our work showed that their transferability in individual samples of natural populations is rather difficult. Drosophila mediopunctata and D. sturtevanti belong to different subgenera, being therefore phylogenetically distant. Thus, throughout both species evolutionary processes, mutations could have become fixed in the flank region of the SSR loci, resulting in null alleles, which could be responsible for the low transferability (only 10%). Therefore, the tested markers were not suitable for genetic variability analyses of D. sturtevanti natural populations, requiring more tests with other primers or the use of strategies for obtaining and synthesizing specific primers for this species.

Keywords: *saltans* group, molecular markers, genetic variability, microsatellite DNA, transferability Financial aid: FAPESP 2014/14059-0; CAPES



Somatic mobilization of transposons: activators factors and controls mechanisms

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TRANSPOSITION ELEMENTS (TEs) correspond to considerable fractions of most genomes, as 80% of maize genome, 45% of the human genome, and for Drosophila species with genome sequenced its extend from 2.5 to 25%. The mobilization of TEs into genomes can promote mutations, changing the coding region of genes, inserting new regulatory or processing elements of genes. Thus, although today there is a consensus that the TEs are a valuable source of genetic diversity for evolution, we also know that many transposition control mechanisms are present in organisms to minimize the deleterious effects that the transposition in excess can promote. Until recently little attention has been given to somatic mobilization of TEs. Recent studies has indicated that the somatic mobilization may be involved in phenomena such as cancer, degenerative diseases, aging and also neuronal plasticity. The mariner element, first discovered in Drosophila, is able to promote somatic mobilization. We have used a system of D. simulans, consisting of non-autonomous mariner element and autonomous elements, called white-peach system, being an appropriate model to measure somatic transposition rate, quantifying the number of red "spots" in a white eyes mutant that was caused by the insertion of a mariner element in the *white* gene. The somatic mobilization of element inserted into white gene promotes the reversion of mutation, restoring the normal eye pigmentation (red). Using this system we have shown that somatic mobilization of the *mariner* element is induced by thermal stress, increased transcription of the element, as measured by qPCR. The *mariner* element has a promoter similar to those of Hsp genes (heat shock proteins). We have also demonstrated, by "temperature shift" assay, that occurr somatic mobilization throughout all Drosophila development, and that it's cumulative. Other abiotic factor we have tested was the ultraviolet radiation, which was not able to promote transposition. Substances that increase the reactive oxygen species (ROS) decreases the somatic transposition. In a "white-peach" system of D. melanogaster we found evidences that in this species the mariner element is inactivated by heterochromatinization of genomic region in that the mariner element is present. Our data suggest that various abiotic stressors factors can cause dissimilar effects on somatic mobilization of the *mariner* element, as well as different species, even closely related species such as D. melanogaster and D. simulans.



Non-genotoxic effects of the lichenic compound usnic acid in somatic cells of *Drosophila melanogaster* evaluated by SMART test and Comet Assay

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LICHENS ARE STRUCTURES resulting from the symbiotic association between fungi and algae and are used in medicine for centuries for therapeutic purposes, by producing a series of secondary metabolites. Among these metabolites can highlight the usnic acid, which has great importance in medicine due to its anti-inflammatory, antitumor, antimicrobial and tuberculostatic properties. Despite its qualities for use in human health, few studies have investigated its potential mutagenic and/or genotoxic risks. This study evaluated the toxic-genetic effect of usnic acid in somatic cells of *Drosophila melanogaster* (Diptera, Drosophilidae) through two methodologies: the Mutation Testing and Somatic Recombination (SMART) in wing cells and the Comet Assay with larval hemocytes. The acid was isolated from lichen Cladonia substellata, using the hot exhaust system in Soxhlet apparatus and ether solvents, acetone and chloroform. The purity of the compound was assessed from the chromatography technique in thin layer. After performed crossings between two lines of D. melanogaster genetically established for the SMART test (mwh and flr^3) the larvae descendants were subjected to different concentrations of acid usnic. In the Comet Assay were used larvae descendants of D. melanogaster Oregon-R strain. Seven concentrations were tested in both assays: 0.16, 0.32, 0.64, 1.26, 2.53, 5.05 and 10.11 mg/mL. Positive control (mitomycin 1mg/mL to SMART and cyclophosphamide at 1mg/mL in comet assay) and a negative control (with the mixture of solvents, distilled water, Tween 80 and ethanol, in both tests) were also established. The SMART test resulted in more than 70% of survival individuals (no toxic effect) with no significant levels of genotoxicity in somatic cells. Similarly there was no genotoxic effect evaluated by Comet Assay in all concentration and experimental conditions applied. The results point to the use of usnic acid as a promising drug, safe for human health and the environment.

Keywords: usnic acid; mutagenicity; SMART; Comet assay Financial aid: FACEPE, CNPq, CAPES, PROPESQ-UFPE



Absence of the mutagenic effect of the lichenic salazinic acid in *Drosophila melanogaster*, evaluated by the wing SMART test

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NATURAL PRODUCTS ARE used by humans for centuries. The search for relief and cure of diseases by eating grass and leaves may have been one of the first forms of use of such substances. Among the large number of organisms used in this area, the lichens have been gaining prominence since its metabolites are widely described by its potential as antihypertensive agents, antitumor, antiviral, anti-inflammatory and antiparasitic. Among these molecules the salazinic acid is particularly important, which has antimicrobial properties, anti-inflammatory and antioxidant. In order to ensure a better use of salazinic acid this study evaluated its possible mutagenic effect through the Somatic Mutation and Recombination Test (SMART) in somatic cells of Drosophila melanogaster. The acid was isolated from Ramalina complanata by the hot exhaust method, in a Soxhlet apparatus using solvents: ether, acetone and chloroform. The compound was purified from successive recrystallizations with *n*-hexane, purity was determined on thin-layer chromatography. After being made crosses between two strains of D. melanogaster genetically established for SMART test (*mwh* and flr^3) the descendants organisms were subjected to different concentrations of salazinic acid (0.32, 0.64, 1.26, 2.53 5.05 and 10.11 mg/mL) using as positive control mitomycin (1mg /mL) and as negative control (distilled water, ethanol and tween 80). SMART results showed toxicity only at concentrations of 5.05 and 10.11 mg/mL, with death of more than 70% of treated individuals. In the other concentrations (0.32, 0.64, 1.26, 2.53 mg/mL) there was no genotoxic effects. The data indicate that this phenolic compound has no genotoxic effects under the test conditions.

Keywords: salazínic acid; mutagenicity; SMART

Financial aid: FACEPE, CNPq, CAPES, PROPESQ-UFPE



Illustrated Manual of *Drosophila* species from São Paulo State - Volume I

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DROSOPHILA (DIPTERA, DROSOPHILIDAE), a model organism biological in studies with emphasis in genetics, evolution and ecology, lacks an updated manual for beginners researchers containing general information on species morphology, manipulation in laboratory and taxonomy. There is a classic work dating from 1950 and others from this decade, but they are of limited access and incomplete in relation to the currently known species. Our research group began preparing an illustrated manual containing description of collection methods, maintenance of the species in laboratory, diagnostic morphological characteristics of the species as well as an identificationkey for the main species from the State of São Paulo. It should be emphasized that of over 130 known species in Brazil, 95 have been recorded in this State. Photographic plates of 33 species and an identification key involving the different groups of the Drosophila genus and the species Zaprionus indianus will also be included. We intend to have this material printed and available online in order to be used in support to studies of drosophilidae biodiversity in the Neotropical region. The flies were collected by traps containing bait prepared with macerated banana and biological yeast (Saccharomyces cerevisiae) that were left in the wood for three days. The identification of the captured drosophilids was performed using an identification key through observations of the external morphology. For cryptic species, we also included an analysis of the aedeagus, that is part of the male terminalia. At this time, we will present the material on melanogaster group (D.melanogaster; D.simulans, D. malerkotliana and D. anannassae).

Keywords: systematics, Drosophilidae, neotropical biodiversity, identification of species Financial aid: FAPESP (2014/14059-0)



Using YGS to uncover the origin of D.pseudoobscura Y chromosome

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THE Y CHROMOSOME is involved in both sex determination and male fertility, and is of great biological importance. The canonical theory states that it evolved through the acquisition of a male determining factor (M) by an autosomal chromosome and subsequent loss of recombination, which leads to a degenerated, highly repetitive Y chromosome with low gene content. However, in Drosophila, the Y chromosome is gaining more genes than losing, contrasting the expected path for a canonical Y, and there is as species, D. pseduoobscura, whose Y chromosome is not homologous to the D. melanogaster Y. While there are previous studies that discuss these facts, only a few species have their Y-linked gene content fully described. That is mainly due to Y chromosome sequences usually appearing fragmented and unmapped, due to the repetitive nature of the chromosome. In order to overcome the difficulties of studying the Y chromosome and to better understand it's evolution the YGS method was created, which compares assembled mixed sex genomes with female short reads and identifies Y-linked sequences with remarkable precision. In the common ancestor of the D. pseudoobscura and D. affinis species group all Ylinked genes went through a translocation to the dot chromosome, however D. pseudoobscura still has a Y chromosome essential to male fertility, with unknown gene content and origin. In order to understand the origin and evolution of the D. pseudoobscura Y we used the YGS method coupled with mRNA sequencing to describe the Y-linked gene content of *D. pseudoobscura*. We were able to identify more than 60 potential gene candidates, within several previously unmapped scaffolds. One of these is the gene CG6661, a single copy gene present in the Y chromosome, an aldehyde dehydrogenase with testis expression. Determining if genes like CG6661 are recent acquisitions of the D. pseudoobscura Y or were already present before the translocation will improve our understanding of this Y chromosome.

Keywords: Y chromosome, *D.pseudoobscura*, genomics

Financial aid: CNPq and FAPERJ



Low mutation rate in mitochondrial genes COI and COII in Neotropical populations of *Drosophila nebulosa*

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STUDY ASPECTS OF divergence in Drosophila nebulosa populations were the aim of this study. This species belongs to the group *willistoni* (*bocainensis* subgroup) of *Drosophila*, and is widely distributed in the Neotropics, from the United States and Mexico, to southern Brazil and Argentina. In northeastern Brazil has been quite sampled in many different environments, from the semiarid region, the Caatinga, even in the Mangroves, Atlantic Forest and Upland Forest. To better understand the genetic relationships among populations of D. nebulosa from different locations of Neotropical region, were analyzed partial DNA sequences of four nuclear genes, Plexin B (Pb), Ankyrin (Ank), Host Cell Factor (Hcf) and Zinc Finger Homedomain-2 (Zfh-2), and two mitochondrial, Cytochrome oxidase subunit I (COI) e Cytochrome oxidase subunit II (COII). After sequencing of the six partial genes from 15 different populations, the results were surprising in shown a very low rate mutation for both mitochondrial genes and high and variable rates of mutation for each the nuclear genes. These nuclear genes are located in the fusioned F+E chromosome in D. nebulosa, expected to have low variability because the ancestral condition of F (a dot chromosome). Along 695 bp of COI and 665 bp of COII just two and three haplotypes were recognized, respectively. Nuclear genes Ank (786 bp) and Hcf (516 bp) presented eight different haplotypes, Pb (753 bp) 11 haplotypes and Zfh-2 (831 bp) presented 21 haplotypes. Zfh-2 gene showed the high variability and was the most diverse between the genes analyzed, with almost one different haplotype present in each population, related to 22 SNPs. In the opposite, COI and COII were much conserved in D. nebulosa species even when we compare geographically distant populations such as from Mexico, Peru, Colombia, Costa Rica and North and South of Brazil. Study processes related to evolutionary divergence within the same species is extremely interesting and this type of study can elucidate issues associated with speciation process, geographical species expansion and fitness effect acting within populations.

Keywords: Diversidade molecular, *D. nebulosa*, genes nucleares, genes mitocondriais Financial aid: FAPERGS and FACEPE



A family of euchromatic tandem repeats with potential regulatory roles in *Drosophila virilis*

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BESIDES THE TYPICAL clustering of satellite DNAs at heterochromatic regions of chromosomes, arrays of complex tandem repeats (TRs) have also been found spread throughout the euchromatin in some species. Herein we studied a family with 172 bp long TRs recently identified in the sequenced genome of D. virilis. After similarity-based clustering of D. virilis sequencing reads, the 172-bp TRs appeared as the 3rd most abundant repetitive family of the genome, spanning 0.85 % of its estimated size. We extracted over four thousand TRs from euchromatic scaffolds of the D. virilis genome assembly. These repeats are organized in arrays up to 14,704 bp long (over 85 repeats) with a mean array size of 3,192 bp. The arrays were found in scaffolds mapped to the chromosomes 2, 3, 4 and 5. The chromosomes X and 6 seem to be devoid of 172-bp TRs. We analyzed 344 array flanking regions of 500 bp each. Only 7,2% of these showed sequence similarity to transposable elements (TEs), suggesting that the 172-bp TRs are not part of any known TE. We also found that small RNA transcripts from the 172-bp TRs are predominantly produced in embryos and gonads of D. virilis, with an average 27-fold transcript enrichment in gonads over carcasses, and an average 18-fold enrichment in embryos over carcasses. This result is consistent with piRNA (PIWI-interacting RNAs) production. It has been shown in Drosophila and other organisms that TEs and satellite DNA insertions may promote local regulatory effects on gene expression mediated by small interfering RNAs (RNAi). The abundance of euchromatic 172-bp TRs, the fact that these arrays may often be close to genes and the presence of small RNA transcripts derived from these repeats offer an interesting model to study the regulatory effect of euchromatic TRs.

Keywords: euchromatic repeats, satellite DNA, RNAi, gene regulation

Financial aid: CAPES, FAPEMIG



Regulation of transposable elements and genes in hybrids between *Drosophila mojavensis* and *D. arizonae*

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CROSSES BETWEEN CLOSE species can lead to genomic disorders, often considered as the cause of hybrid incompatibility. In order to understand the first steps in hybrid incompatibility, we performed reciprocal crosses between two species of *Drosophila* (D. mojavensis and D. arizonae) that diverged less than 1 MYA. We sequenced ovarian mRNA from the parental strains and from hybrids obtained from reciprocal crosses, as well as ovarian piRNA from the reciprocal crosses. Our results show that the parental lines differ in their genes expression (~20% genes differentially expressed) but more importantly in the TEs expression (~40% TE differentially expressed). On the contrary, reciprocal hybrids presented mean levels of genes expression when compared to the parental lines. TEs were also mainly regulated in hybrids with only few exceptions that were either higher expressed in hybrids than in the parental lines. The presence of piRNA with a pingpong signature is in agreement with this observation. Our results show that no global activation of TE is observed when crossing closed related Drosophila species, but instead, some specific elements are totally uncontrolled.

Financial aid: FAPESP, CNPq



Quantitative composition of protein, lipids, minerals and humidity in different *Drosophila* culture medium

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ONE THE MOST important aspect of success of Drosophila culture in laboratory is the quality of the culture medium. Different concentrations of important nutrients can affect not only the viability of the stocks (mate preference, egg-laving and longevity) but also the quality of polytene chromosome preparations of larvae and other important characters under selection. If specific sources of consumed calories are taken into account, the dietary composition can plays a critical role in determining physiology and lifespan in *Drosophila*. Using *Drosophila melanogaster*, some authors are developing studies about the diet composition and its relation with behavior, physiology, and lifespan, regarding yeast-rich and excess dietary sugar, for example. So, the diet composition, alone and in combination with overall caloric intake, is capable to modulate lifespan, consumption, and fat deposition in flies. To better understand the dietary composition of food where species of drosophilids are maintained, we began the analyses of proteins, lipids (fat), minerals and humidity of five different recipes of medium, one cornmeal-agar-yeast currently used in our laboratory, three other variations with different ingredients (molasses or melado, banana in natura and potato dehydrated), and one standard culture medium, with no agar or yeast, established by Margues et al. and used since 1966 in some drosophilid laboratories. In the medium currently used in our the laboratory the composition of proteins was 2,01 %, lipids 0,44 %, minerals 0,30 % and humidity 81,95 %, all statistically different from Margues et al. because have more lipids, minerals and humidity, and less proteins. This and other results are better discussed in this work.

Keywords: Meio de cultivo; proteínas; lipídeos; nutrição Financial aid: CNPq, FACEPE, PROPESQ-UFPE



Identification and syntenic relationships of the Cid gene in three species from the *Drosophila* subgenus

José Ricardo Teixeira, Gustavo C.S. Kuhn

CENH3 IS A CENTROMERE-SPECIFIC histone H3 variant (Cid in Drosophila) that acts as an epigenetic marker for centromere identity and is required for kinetochore assembly. Unlike H3, CenH3 exhibits a high degree of variability. The most variable region of the protein is the Nterminal tail, which varies considerably in sequence and length, even amongst closely related species. Such feature allows its discrimination from H3. A single-copy gene codes for CenH3 in most eukaryotes studied so far. In Drosophila, Cid is a well-characterized gene but most studies are concentrated in few species from the *melanogaster* subgroup (subgenus Sophophora), where it was found located in the chromosome arm 2R (Muller element C) between the BBC and CBC genes. In this work, we aimed to investigate Cid evolution at a longer time frame. Thus, we selected three species (D. mojavensis, D, buzzatii and D. virilis) with sequenced genomes from the subgenus Sophophora, distant from the melanogaster subgroup more than 40 mya. Blast searches allowed identification of Cid, later confirmed by sequence alignments and phylogenetic trees (containing both H3 and Cid sequences). As expected, Cid is located between the BBC and CBC genes in the chromosome 5 (Muller element C) of D. mojavensis and D. virilis. However, in D. mojavensis Cid is located within an inverted block comprising at least 60 kb, suggesting chromosomal rearrangement. No other copy of Cid was found in these two species. Unexpectedly, in D. buzzatii Cid is located between two different genes in the chromosome 3 (Muller element B). The analysis of PacBio long reads from D. buzzatii confirmed this finding. The data suggest inter-chromosomal segmental duplication of Cid, although we did not detect a copy of Cid on its original location in the genome assembly. We will now conduct further sequence analysis and perform experimental studies (PCR and FISH on chromosomes) in order to better understand this unusual feature of Cid in D. buzzatii.

Financial aid: FAPEMIG



Monitoring the genetic Drosophilidae diversity in Atlantic Forest Biome on northern Rio Grande do Sul, Brazil

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IN THE LAST three decades some faunal surveys of Drosophilidae have been done in several environments in the Neotropical region, especially in Brazil but approximately 30 to 50% of the drosophilids in Brazil have not yet been described. This discouraging scenario is exacerbated by the degradation of some biomes which causes a profound loss of species diversity, as well as the loss of information about the present structure of their communities. In the territory of Rio Grande do Sul, remaing Atlantic Forest corresponds to only 7.5% of original area with a high degree of fragmentation. Historical and contemporary fragmentation events have influenced species diversity and distribution in this biome. Flies of the family Drosophilidae show up as excellent models for study. These are small insects, easy to catch, with well-known taxonomy and respond to environmental fluctuations. The objective of the present study is to evaluate genetic structure in populations of the Drosophila sturtevanti collected in three collection points in sites that are remnants of Atlantic Forest in the northern state of Rio Grande do Sul, Brazil, in order to contribute to the understanding of the processes that affect the patterns of genetic variability in this species. The populations were sampled in sites with different level of environmental changes: Universidade Federal de Santa Maria campus in Palmeira das Missões-RS (UFSM), Parque Estadual Papagaio Charão in Sarandi-RS (PEPC) and Parque Estadual do Turvo in Derrubadas-RS (PET). Individuals were identified based on the external morphology and male genitalia. Total DNA was extracted from three individuals of each survey site performing to the protocol cationic detergent CTAB (cetyl trimethylammonium bromide). After extraction, a partial fragment of COI gene was amplified and sequenced using primers LCO and HCO, decribed for DNA barcoding analyzes - to Cytochrome Oxidase I genes. Our collect effort recoverd 6652 flies from UFSM, 252 from PEPC e 1210 from PET. Fortunatelluy we found flies from the saltans group (D. sturtevanti) in the three collect points. PCR amplicons recovered from samples showed size of 710 bp approximately, as expected, allowing us to follow to the sequencing step. From now, we will perform the amplicons automatic sequencing in order to accomplish the structural and evolutionary analysis of COI gene in the three populations. This methodology will evaluate the effects of habitat fragmentation on the genetic diversity of natural populations of this species.

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Chromosomal inversion polymorphism in *Drosophila neocardini* population from the state park of Serra do Tabuleiro, SC – Brazil

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THE SPECIES *DROSOPHILA neocardini* (Streisinger, 1946) belongs to the subgenus Drosophila, group and subgroup cardini. Its distribution comprises the Neotropical region, and it is most frequently found in humid areas. In order to study the chromosomal polymorphism of this species, adult samplings of *Drosophila* populations were obtained from the state park of Serra do Tabuleiro. The collection was performed during the period of February and March, 2015. The flies were obtained by placing traps containing fermented banana bait. *Drosophila neocardini* were identified based on their particular morphological features and through dissection of male genitalia. 16 isofemale lines were maintained in instant medium for oviposition. Slides with salivary glands were prepared, and samples were analyzed by observing the banding patterns of polytene chromosomes. At least 5 nuclei from each dissected larvae were analyzed. The results point to a moderated polymorphism of paracentric inversion in the population studied. Also, a new paracentric inversion in the chromosome X of *D. neocardini* was described as well as its implication in the chromosomal evolution of the species.



Population genetics of *Drosophila polymorpha* from Santa Catarina State using a fragment of *period* gene as molecular marker

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DROSOPHILA POLYMORPHA IS a species of the subgenus Drosophila, cardini group. There is no work on the genetic structure of their populations in any region of its distribution area. This study will contribute to a better understanding of the genetic structure and demography of D. polymorpha populations from Atlantic Forest regions of the Santa Catarina. Samples were collected, using traps containing fermented fruits, in three protected areas, Canela Preta State Biological Reserve, Serra do Tabuleiro State Park and Aguaí State Biological Reserve, localized respectively in the northern, central and southern of Santa Catarina. Species were separated using morphological characters and the identity of D. polymorpha individuals was confirmed using COI gene. A fragment of Period gene was amplified by PCR using primers designed based on an alignment of sequences from another Drosophila species. PCR products of twelve individuals of each population were sequenced in both directions and the identity of amplicon was confirmed using the blast tool. A fragment of 660bp of the Period gene was used for analysis of polymorphisms intra and inter populations and to infer the demography of populations. These analyses were conducted using DNASP 5.10.01 and Proseq 2.91 programs. A haplotype network was constructed using the program TCS 1.21. The three populations have low genetic diversity (nucleotide diversity in Aguai= 0.0029 is the highest value). Fst values ranged from -0.0169 to 0.0255 (latter value between Aguai and Canela Preta), indicating a lack of genetic structuring of populations. Thirteen Period haplotypes were sampled, considering all populations, with an overall haplotype diversity of 0.660. The haplotype network shows a star-shaped, with a considerable number of rare haplotypes separated by single nucleotide differences, and a lack of clustering of haplotypes by population. Different neutrality tests, as Tajima's D, Fu and Li's (D* and F*), Fu's Fs and R2 present significant deviations from neutrality, especially when considering the populations together. When considering each population separately, Tabuleiro present more tests with significant deviations from neutrality. The results found in different tests, indicate a probable population expansion some time in the relatively recent past.

Keywords: *Drosophila polymorpha*; population genetics; *period* gene; Santa Catarina Financial aid: FAPESC, Universidade Federal de Santa Catarina



DINE-TR1 in D. virilis and D. americana: widespread genomic distribution, association with satellite DNA emergence and piRNAs

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DROSOPHILA INTERSPERSED ELEMENTS (DINEs) are a profuse but poorly understood group of Helitrons present in many Drosophila species. The overall structure of DINEs includes two conserved blocks 5' and 3' (A and B, respectively) that may or may not contain a few central tandem repeats (CTR) between them. We identified a subset of DINE elements containing CTRs of approximately 150 bp, termed DINE-TR1, in the genome of several *Drosophila* species. Here, we focused on DINE-TR1 in two closely related species with sequenced genomes from the virilis group, D. americana and D. virilis. A combination of bioinformatics and fluorescent in situ hybridization (FISH) to DNA fibers (fiber-FISH) revealed that the CTRs within DINE-TR1 have expanded into satellite DNA-like arrays in both species. FISH on D. virilis and D. americana chromosomes showed that DINE-TR1 is highly abundant in pericentromeric heterochromatin boundaries, telomeric regions, and in a large portion of the Y chromosome. It is also present in the centromeric region of one autosome from D. virilis but not in D. americana, suggesting recent expansion to centromere in D. virilis. In silico searches revealed that DINE-TR1 is often found in tandem arrays with up to 11 copies. This is the largest Helitron head-to-tail array detected to date. Finally, we found that DINE-TR1 is abundant at piRNA clusters throughout the genome, and small DINE-TR1-derived RNA transcripts (~25 nt) are predominantly expressed in male and female gonads. These features, together with its profusion in pericentromeric regions, indicate the involvement of this element in the piRNA pathway. We suggest the potential for piRNA-mediated regulatory roles of DINE-TR1 at local and genome-wide scales.

Keywords: *Drosophila virilis*, *Drosophila americana*, transposable element, genome evolution, Helitrons Financial aid: FAPEMIG



Genetic diversity of native and exotic drosophilids of the Brazilian Atlantic Forest

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GENETIC DIVERSITY IS necessary in order that species evolve and adapt to environmental changes. Insects, among which are the drosophilids, are an abundant group in tropical forests consisting of native and exotic species. We assess the genetic diversity of Drosophila sturtevanti, a native species of Neotropical region and D. malerkotliana, an exotic species. Three Atlantic forest fragments located north of the São Francisco River in Pernambuco sub region were studied: the Ecological Station Tapacurá (TAP), the Experimental Station Itapirema (ITA) and the Charles Darwin Ecological Refuge (DAR). Genetic diversity was assessed by the dominant marker Inter Single Sequence Repeats (ISSR) with three primers (ISSR1 ((CA)₆AT), ISSR2 ((CA)₆GC) e ISSR3 ((CA)₆AG). Forty-two individuals of D. sturtevanti were studied (13 TAP 18 and ITA 11 DAR) and 44 of D. malerkotliana (13 TAP 19 and ITA 12 DAR). It was observed higher genetic diversity for native species, which presented 54 amplicons (20, 15 and 19 for ISSRs 1, 2, 3, respectively), of which 52 were polymorphic. Drosophila malerkotliana presented 49 amplicons (17, 14 e 18 for ISSRs 1, 2, 3, respectively), of which 48 were polymorphic. Drosophila sturtevanti presented higher polymorphism for the three ISSRs in TAP (ISSR1 13, ISSR2, 12 and ISSR3 14 polymorphic fragments) compared to D. malerkotliana (ISSR1 and ISSR2 10 and ISSR3 12 polymorphic fragments). The same pattern was observed in ITA (ISSR1 15, ISSR2 ISSR3 12 and 16 for *D. sturtevanti* and ISSR1 10, ISSR2 9 and ISSR3 14 for *D. malerkotliana*) and DAR (D. sturtevanti ISSR1 15, ISSR2 9 and ISSR318 and D. malerkotliana ISSR1 13, ISSR2 8 and ISSR3 17). These results show more pronounced genetic diversity for native species in all forest fragments and suggest the greater adaptability of D. sturtevanti compared to D. malerkotliana against possible environmental changes.

Financial aid: FACEPE, CAPES, CNPq



Haplotype and nucleotide diversity of COI and COII in Drosophila sturtevanti of the forest fragments in the northwest region of São Paulo

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DROSOPHILA STURTEVANTI BELONGS to saltans group, sturtevanti subgroup, being the most geographically wide species within the group. To several markers, the species populations presents high genetic variability, being an excellent model to population structure studies and to phylogeographic and phylogenetics analyses. The aim of this work was to analyse the haplotype and nucleotide diversity of samples collected in two Atlantic Forest fragments of northwest region of São Paulo, located at Matão (21.37°S 48.32°W) and Nova Granada (20.53°S 49.32°W). The genomic DNA was extracted from the flies, followed by the amplification of mitochondrial genes Cytochrome Oxidase I (COI) and Cytochrome Oxidase II (COII) through Polymerase Chain Reaction (PCR) of 15 individuals of D. sturtevanti from each locality. The resulting sequences, to each gene, were compared with those available in the database using the BLAST tool of GenBank, to verify the sequences identity. After editing and alignment with the aid of BioEdit software, the sequences were analyzed using the software DNASp, to calculate the following statistics parameters: number of haplotypes observed, base pairs, polymorphic sites, haplotype diversity (Hd), and nucleotide diversity (Pi). The DNA fragments presented 235bp and 314bp, to COI and COII, respectively, being observed 7 haplotypes to COI and 5 to COII, and a single haplotype [H1] shared between the two areas for the COI gene. Polymorphic sites were observed, 51 in COI and 96 in COII. It showed high haplotype diversity (0,964 - COI; 0,786 -COII) and nucleotide (0,095 - COI; 0,023 - COII) when compared with other population of this species from Panamá and others *Drosophila* species analyzed to these genes. These results may indicate a high population size for Drosophila sturtevanti and some degree of population structure for this region.

Keywords: Cytochrome Oxidase I, Cytochrome Oxidase II, mitochondial markers, *saltans* group Financial aid: FAPESP 2014/14059-0; Bolsista CAPES



Applying the model organism *Drosophila melanogaster* to detect genotoxicity associated to air pollution

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IN ORDER TO BIOMONITORING environmental pollutants, various methodologies have been applied in recent decades using model organisms as biomarkers of genotoxic mutagenic and/or recombinogenic effects. Aim of this study was to test the sensitivity of the model organism Drosophila melanogaster exposed to different levels of air pollution associated with urbanization, and its genotoxic effect, assessed by the comet assay. The study site was the Vitória de Santo Antão city, in Pernambuco state of Brazil. The municipality has been a constant target large industrial investment that brings not only economic benefits but also increased air pollution due to the release of gases from factories, industries and vehicles, which can be potentially toxic, causing serious problems to the health of population and other bodies. In this work were studied larvae of *Drosophila melanogaster* exposed for six days to air pollutants in an urban and rural area of the municipality. The results were compared to a negative control group, exposed in the same period to a preserved area called Catimbau National Park (Buíque, Pernambuco), distant 242 km from Vitória de Santo Antão. The genetic damage of the three treatment groups (rural, urban and national park) were classified into four categories, from zero (no damage) to four (highly damage), according to the standards of comets formed. The results were analyzed using ANOVA and Bonferroni statistical test that showed significant increase of genetic damage in organisms submitted to the urban environment, compared to the rural and the national park. The results pointed towards that more studies can be conducted on air pollution in urban environments with the sensitive model organism D. melanogaster.

Keywords: Ensaio cometa, genotoxicidade, centro urbano

Financial aid: FACEPE, CNPq and PROPESQ-UFPE



Description of the gene content of the Y chromosome of three Drosophila species of repleta group

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IT IS ESTIMATED that around one fifth of *Drosophila* genome is in the Y chromosome. Despite this fact, until today, the Y chromosome of only two species of this genus is known in depth, D. melanogaster and D. virilis. Much of the problem happens due to intrinsic Y features, such as being heterochromatic and rich in repetitive regions. Y chromosome genes may affect some aspects such as sex ratio and behavior but is mainly associated with male reproductive functions. In that regard knowing the gene content of these chromosomes may be relevant to several areas of biology that use Drosophila as a model. Recent studies show that the gene content of Y chromosome varies in the different Drosophila groups however it is still unknown how the Y differs in close related species. New studies using such species could possibly reveal significant differences even in the Y gene composition among them. A good model for this proposal is the sibling species of repleta group: D. mojavensis, D. arizonae, and D. navojoa. Previous studies have indirectly suggested that these three species have different gene composition in the Y. This present study aims to describe the content of coding genes in the Y chromosome of them. We will use the YGS (Y chromosome Genome Scan) method, which consists of comparing short-read sequences of females with the male assembled genome, in order to identify candidate sequences. In relation to D. mojavensis, one of the 12 genomic species described, it was used the genome published in 2007. In this case, over 100 candidates were identified, and now we are in the process of annotating its genes. In relation to D. arizonae and D. navojoa, the genomes were assembled by Dra Therese Markow group (University of California, San Diego) who also collaborated with this project. D. arizonae has already been analyzed by the YGS and sequences candidates are being prepared for annotation. Finally, the genome of D. navojoa is being prepared for the YGS.

Keywords: Y chromosome, D. mojavensis, D. arizonae, D. navojoa, Sibling species Financial aid: CNPq and FAPERJ





Evolução

Evolution



Phylogenetic relationships of three understudied *Drosophila* groups

Eduardo G. Dupim, Suzana C. Vaz, Antônio Bernardo de Carvalho

THE GENUS DROSOPHILA contains >2.000 described species, subdivided into several subgenus and tens of taxonomy groups. Over the last 30 years many molecular studies dealing with parts or entire Drosophila phylogeny have been published, clarifying the relationships between the majors Drosophila clades and its closely related genera. However, the relationships of several minor groups remains completely unknown. Here, as part of a study about the Y chromosome evolution in 400 Drosophila species, we investigate the unknown phylogenetic relationships of the three Drosophila species: D. lamottei (which belongs of dentissima group, a minor group of subgenus Sophophora); D. carbonaria (the single member of the carbonaria group within subgenus Drosophila, related with virilis-repleta radiation); and D. ponera (an unclassified specie of the subgenus Drosophila, possibly related to the quinaria group). We constructed phylogenetic trees with the species of interest and related species of the same subgenus, using concatenated sequences of Amyrel and kl-5 genes (~3Kb), analyzed by maximum likelihood and Bayesian inference. Our results show that D. lamottei is closely related to the melanogaster group, whereas D. carbonaria is a sister-group of the melanica group, and D. ponera is related with Immigrans-tripunctata radiation. These phylogenetic relationships are in accordance with our previous results about the Y chromosome gene content of these species.



Productivity in *Drosophila* species (*saltans* subgroup, *saltans* group, *Sophophora* subgenus): effects of *Wolbachia* in normal and introgressed strains

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REPRODUCTIVE ISOLATION MECHANISMS prevent or decrease the exchange of genes between species or populations that are in process of speciation. These mechanisms comprehend a series of processes operating at different levels of reproduction, from pre to post-zygotic barriers. Lately, studies have indicated that the absence of progeny can also be promoted by interactions of symbionts and their hosts. The most known endosymbionts capable of interacting with insects, affecting their reproduction, are the proteobacteria of the genus Wolbachia. We investigated the Wolbachia effects on reproduction, analyzing the productivity (number of progeny) in crosses among four strains of Drosophila saltans and D. prosaltans and two introgressed strains started with F1 hybrids of both species. Intra and intercrosses of infected and uninfected strains were performed. The elimination of Wolbachia for producing uninfected strains was performed by treatment with tetracycline. According to the literature, the main mechanism resulting from the interaction symbiont-host is cytoplasmatic incompatibility (CI) and occurs in intercrosses between uninfected females and infected males. The results of our tests were variable. Several combinations expected to be sterile (when CI effect occurs) were the most or one of the most productive. In other cases, intercrosses of both infected parents, expected to be highly fertile, were sterile or very low fertile. Considering all the crosses analyzed, the present results indicated absence of CI. The high productivity found in crosses between infected females and uninfected males indicated that this combination is responsible for the maintenance and spreading of Wolbachia infection in the strains used. In addition, the elimination of the bacteria in male parents produced beneficial effects, annulling the cross sterility. Finally, we suggest that the differences found in the introgressed strain crosses might be caused by their chromosome constitution.

Keywords: cytoplasmic incompatibility, host-symbiont interaction, incipient isolation, introgression, reproductive fitness Financial aid: CNPq, Programa de Pósgraduação em Genética IBILCE-UNESP



First phylogenetic proposal to *Rhinoleucophenga* Hendel 1917 (Drosophilidae, Steganinae): exploring treatments of continuous characters and molecular data sets

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RHINOLEUCOPHENGA IS A NEW WORLD genus abundantly found in the Neotropical savannah and grassland areas. In order to propose a first phylogenetic hypothesis to this genus, ten species were redescribed and 17 new ones were described, increasing ca. 56% the number of species in the genus. Phylogenetic relationships are proposed based on morphological and molecular data sets obtained from 67 taxa: 50 as ingroup and 17 as outgroup. The phylogenetic analyses were performed based on 166 morphological characters: 42 transformational discrete characters, 20 neomorphic discrete characters, 93 body measures and 11 setae meristic counting (104 continuous characters). From the continuous characters also were performed 58 ratios. Continuous characters were treated as additive ones, rescaled from 0 to 1 in order to avoid an excessive weighing of character transformation, and analyzed without discretization; it is the first time that continuous characters are not discretized in Drosophilidae phylogenetic studies. Five analyses were performed with TNT Software; the matrixes were elaborated as follow: (A) 42 transformational, 20 neomorphic and 58 continuous rationed characters; (B) 42 transformational, 20 neomorphic and 104 continuous characters; (C) 42 transformational, 20 neomorphic and 58 continuous rationed characters log-transformed; (D) 42 transformational, 20 neomorphic and 104 continuous characters log-transformed; (E) 42 transformational and 20 neomorphic characters. All analyses were performed with and without implied weighting. Rhinoleucophenga was presented as a paraphyletic genus in all analyses. The continuous data set presented high phylogenetic signal, mainly continuous characters treated as ratios, being fundamental to improve the branches support obtained through only discrete data. Some species clades were always obtained, and named as follow: R. pallida; R. brasiliensis; R. punctulata; R. paullistorum; R. alada; R. brasilis; R. tangaraensis and R. montensis; R. matogrossensis and R. nigrescens. A fragment of the Cytochrome oxidase I (COI) gene is being used to suggest a phylogeny based on molecular data. After, combined analysis of morphological and molecular data will provide a more comprehensive evolutionary scenario to Rhinoleucophenga.

Keywords: Systematic; Pampas; Paraphyletic; *Leucophenga*; *Gitona* Financial aid: CNPq, PRONEX-FAPERGS (10/0028-7), CAPES


Evolution of a new gene on the Drosophila Y chromosome

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ALTHOUGH SEX CHROMOSOMES arise many times independently, their evolution is shaped by the lack of recombination, among other evolutionary forces. Genomic studies with Y chromosomes are difficult, due to their richness of repetitive DNA, but they are involved in major biological phenomena, such as male fertility and sex determination. Drosophila offers advantages to this research, because there are several sequenced genomes and the possibility of cultivation in the laboratory. In the sequenced Neotropical species D. willistoni our research group has previously found a new Y-chromosome gene (CG18155Y). The goal here is to find when in the phylogeny of fruit flies this gene moved to the Y in the lineage that originated D. willistoni and unveil secondary moves to the Y chromosome of other species. We tested the linkage of CG18155Y to the Y chromosome of species evolutionarily close and distant from willistoni group using PCR with degenerated primers. The PCR was run separately to males and females and the amplification only on males showed the Y-linkage of CG18155Y. We focused on the two main subgenera of the Drosophila genus, and tested 120 species up to this moment. In the sister groups, willistoni and saltans, this gene appear to be Y-linked, showing positive results in all species tested. Unexpectedly, this gene seems to have moved to the Y chromosome in the Zaprionus genus as well. Thus in three species the PCR worked only on males, what means the locus is Y-linked. However, in the most of tested species of other groups the locus is on autosome or X-linked. This recently discovered gene moved to the Y chromosome in the common ancestor of willistoni and saltans groups, and secondarily, to Zaprionus. The independent gain of CG18155Y in these two lineages is a recent event, less than 60 MY. These findings are fully compatible with recent ideas about the evolution of Drosophila Y chromosome, in which the rate of gene gains is higher than losses.

Keywords: Y chromosome, gene gain, *Drosophila*, *Zaprionus*



A systematic survey of *Wolbachia* prevalences in Neotropical and invasive Drosophilids of Minas Gerais

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WOLBACHIA IS A HIGHLY diverse genus of intracellular, maternally inherited bacterial endosymbionts, found in many species of arthropods and nematodes. Recent estimates suggest that these bacteria infect at least 70% of insects, causing reproductive alterations, such as cytoplasmatic incompatibility, male-killing, parthenogenesis induction and feminization of genetic males. Such alterations in host reproduction might generate a selective advantage for infected females, hence triggering high levels of infection in host populations. In this work, we used PCR methodology to test for the presence of Wolbachia in the specimens of drosophilid sampled in Minas Gerais state. We thus compared the Wolbachia sequences with those available in GenBank. The flies were captured using banana baited traps and individual females were transferred to culture medium to establish isofemale lines, maintained in B.O.D chambers at 25oC and a 12/12h light/dark cycle. The identification of each drosophilid host was determined by using male terminalia morphology and COI sequence bar coding. Single female total DNA was extracted using Wizard Genomic DNA Purification Kit (Promega). For Wolbachia straintyping, we probed the total DNA with wps, 16S and ARM primers. The PCR bands of wsp were purified, sequenced and compared with wsp sequences already published. Until now, the PCR reactions generated positive results for D. sturtevanti, D. melanogaster and D. willistoni. As expected, the *wsp* sequences found in *D. melanogaster* and *D. willistoni* presented no differences with sequences of these same hosts available in GenBank. In addition, the *wsp* sequence found in D. sturtevanti from Minas Gerais was identical to the earlier reported Panama sample of the same host, which is identical to the sequence obtained from sand fleas. Sequence identities and lack of polymorphisms suggest that the Wolbachia infections of D. sturtevanti has probably spread quite recently in Central and South America.



Evolutionary aspects of gene expression during *Drosophila melanogaster* spermatogenesis

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GENES THAT APPEARED recently in the evolutionary history of a taxonomic group are considered new genes. They can arise by exon recombination, transposons, lateral gene transfer, gene fission/fusion, DNA based duplications, retroposition, de novo origination or combinations of those mechanisms [1]. Although some of those new genes are functional, most of them become pseudogenes. However, when they happen to be functional, new genes can quickly convert to essential genes or bear an important function at different phases of the development of individuals of different species [2]. In this way, new genes expressed during spermatogenesis the system of male gamete development - are probably related to fertility, mobility, form and function of the sperm cells. And, therefore, must be more expressed during the late phases of the gamete development, bearing in mind that expression relates to functionality. Hence, this project aims to test the hypothesis that there is a relation between a gene's age and its expression during spermatogenesis. To test this hypothesis, we searched for a correlation among genes evolutionary ages and it's differential expressions in spermatogenesis phases in Drosophila. According to this hypothesis, it's expected that new genes are more frequently expressed during post-meiosis, the latest phase of germline development. To answer those questions, bioinformatics and computational biology tools were used and statistical methods were applied to correlate already available data of gene age [3] and gene expression during spermatogenesis phases in Drosophila [4]. Our results in Drosophila have shown that the proportion of new genes expressed in late expermatogenesis (meiosis and post-meiosis) is significantly higher then in the beginning of the processes (mitosis). Also, we found that the expression level of new genes is higher than the expression of old genes during meiosis and post-meiosis, and the opposite pattern occurs during mitosis. These results implicates that new genes have an important role during the late spermatogenesis, which could be related to sperm fertility and speciation process.

Keywords: *Drosophila*, spermatogenesis, differential gene expression, gene age, bioinformatics



Satellite DNA landscape in the sequenced genome of Drosophila buzzatii

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SATELLITE DNAs (satDNAs) are among the major and most rapidly evolving components of eukaryotic genomes. They are composed of tandem repeats that form long arrays of several kb typically located in heterochromatic regions. The organization and evolution of satDNAs are still poorly understood compared to other repetitive DNAs. D. buzzatii (Dbu) is a cactophilic species from the South American buzzatii cluster (repleta group).. The recent availability of the sequenced genome of *Dbu* provides a great opportunity to revisit and expand our studies on satDNA organization and evolution in this species, now in the context of whole genomic sequences. We screened the *Dbu* sequenced genome for satDNAs using the *Repeat Explorer* software. We then conducted sequence and phylogenetic analysis and determined their location by FISH on mitotic and polytene chromosomes. We found only two satDNAs in Dbu that, together, represent 1.9% of the genome, the lowest amount of satDNA described in Drosophila so far. The estimated *Dbu* genome size is also small (~160Mb) compared to other *Drosophila* species. The low satDNA content may be the result of selective constraints for maintaining a relatively small genome size. The *pBuM* satDNA is the most abundant one, with 1.7% estimated genomic contribution. The pBuM repeats from chromosome Y have undergone a faster rate of homogenization for chromosome-specific variants compared to the autosomes, what provides an interesting model to understand the evolution of satDNA arrays located on a single nonrecombining chromosome. The second satDNA, named TR198, is composed by 198 pb repeats, representing 0.2% of the genome. TR198 is located in the telomeric and euchromatic portions of all chromosomes, except the "dots" and Y. We did not identify telomeric retrotransposons in Dbu. Moreover, we found TR198 arrays with 3-5 repeats near genes. It remains to be investigated whether TR198 plays a role in telomere maintenance and/or in the regulation of nearby genes.

Keywords: Satellite DNA; *Drosophila buzzatii;* centromere, telomere Financial aid: CAPES, FAPEMIG



Ultrastructural aspects of spermatozoa and their phylogenetic application in *Zaprionus* (Diptera, Drosophilidae)

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THE ZAPRIONUS GENUS consists of about 60 species of drosophilids that are native of the Afrotropical region. The phylogenetic position of Zaprionus within the Drosophilidae family is still discussed. In the present study, ultrastructural features of spermatozoa of six species of Zaprionus genus and the species Drosophila willistoni and Scaptodrosophila latifasciaeformis were analyzed. Data obtained from the literature were also included in the cluster analyses. The ultrastructure revealed that the species have the same flagellar ultrastructure. The axoneme has a '9+9+2' arrangement: two pairs of central microtubules, nine peripheral doublet microtubules and nine accessory microtubules. Two mitochondrial derivatives close to axoneme were present, one larger than the other. Except for Z. davidi and Z. tuberculatus, the analyzed species presented paracrystalline material in both mitochondrial derivatives Moreover, cross-sections of the testes showed 64 spermatozoa per bundle in all species as well as the occurrence of cystic spermiogenesis. In the cluster analysis, six Zaprionus species, being four analyzed in the present study and six species of Drosophila, being five taken from literature were grouped, indicating proximity between them in the analyzed characteristics, but without forming a legitimate clade. The species Z. david, Z. tuberculatus, S. latifaciaeformis and D. hydei presented incongruent positions in the cladogram, perhaps requiring the analysis of a larger number of species to obtain a stronger relationship. Despite, the results indicated that the sperm ultrastructure is an important tool, which can help in phylogenetic and taxonomic studies on insect groups.

Keywords: Drosophilidae, mitochondrial derivatives, spermatogenesis Financial aid: CAPES



Symbiotic strategies of *Wolbachia* upon *Drosophila willistoni*: ontogenetic dynamics of infection and host fitness markers

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WOLBACHIA PIPIENTIS IS A BACTERIUM widely distributed among arthropods, whose reproduction it manipulates in order to increase its own frequency. In symbiotic relationships, the infection density can influence the endosymbiont manipulation capacity and the transmission success to the offspring. We characterized *Wolbachia* strains infecting a *Drosophila willistoni* population in different infection densities. Thus, to explain why such scenario has been maintained, we hypothesized that there might be differences in the bacterial load during host development. In addition, we conjectured it could be possible to find variations on host fitness when comparing flies infected with different infection types. Therefore, we investigated the infection dynamics of Wolbachia during the ontogeny of D. willistoni infected by low- and high-titer wWil and low-titer wAu-like, and possible fitness consequences on the hosts. We analyzed two isofemale lines from each infection type (same Wolbachia strain and titer). We extracted DNA from embryos, 3rd instar larvae (males and females), pupae, virgin and 20 days-old flies (males and females). The DNA samples were analyzed and the number of wsp (Wolbachia Surface Protein) copies was estimated by comparing sample Ct values with a standard curve by qPCR. Furthermore, we assessed classic fitness markers (fecundity, viability and survival) from flies belonging to each infection type. Generally speaking, the Wolbachia ontogenetic dynamics in these flies seems to be densityrather than strain-dependent. Curiously, in the case of high-titer infections there is a bacterial load increase in embryos and 20-days old females, which are exactly the ontogenetic stages where transmission fidelity is more critical. Moreover, although viability and survival data did not show any differences when comparing flies infected with distinct strains and titers, females infected with low-titer wAu-like showed to be more fecund that the others. We thus propose a model where the two main infection types (high-titer *w*Wil and low-titer *w*Au-like) are maintained in D. willistoni populations by equilibrium between a higher fecundity of wAu-like-infected females and a higher transmission success in flies infected with high-titer wWil.

Keywords: symbiosis; qPCR; density; fitness

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The effect of Y-chromosome substitutions on hybrid male sterility in *Drosophila* revealed by studies with mixedgenotype lines

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DROSOPHILA Y CHROMOSOMES display population variation within and between species. Ychromosomes from disparate populations might disrupt harmonious interactions between conspecific genetic factors and perturb the regulatory network of a species, leading to reduced male fertility, complete sterility or inviability. We used males with introgression genotypes constructed from three sister species, D. simulans, D. mauritiana and D. sechellia, to quantitatively assay the effect that heterospecific Y chromosomes have on male fertility. Males from 18 mixed-genotype lines were tested for fertility (progeny size). These lines were defined by their genomic composition, formed as a combination of: (1) Y-chromosome origin (D.simulans or D. sechellia); (2) genomic background origin (three lines of D. simulans: A14, SR12-2-7, or G23); and (3) introgressed D. mauritiana segment on D. simulans 3rd chromosome (homozygotes of previously built introgression lines: 3.4/8.4, 21.12/26.14, and 38.9/38.6). We find that D. mauritiana segments differentially interact with the Y-chromosome of D. simulans (Ysim) and D. sechellia (Ysech), generating significant differences in progeny size. Most notable, defined segments of *D. mauritiana* are more detrimental in the presence of the Ysim than in the presence of the Ysech, in spite of most of the autosomal and X-linked genome being from D. simulans. More specifically, one segment reduces fertility in the Ysim background but the infertility is partially rescued by the Y chromosome of D. sechellia. This result illustrates the multifactorial nature of genomic interactions involving the Y chromosome and autosomes. Our study supports the hypothesis that the Y chromosome might have contributed significantly to the evolution of reproductive isolation and to the conditional manifestation of infertility in specific genotypic combinations.



Wing and aedeagus morphometric analysis in five populations of *Drosophila sturtevanti* from Atlantic Forest fragments

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MORPHOMETRIC ANALYSIS OF the wing and the aedeagus has been used to study evolution in the genus Drosophila. Due to high levels of phenotypic plasticity, the wing is an excellent marker for environmental variation, while the aedeagus present a fast and divergent evolution when compared with other morphological characteristics, and is a diagnostic feature for species identification. We studied the variation of wings and aedeagi aiming to detect phenotypic plasticity, to observe the existence of a latitudinal cline and to detect genetic differentiation among populations. In this paper, we analyzed Drosophila sturtevanti males (sturtevanti subgroup, saltans group) collected in five fragments of Atlantic Forest, two from São Paulo State (Nova Granada - 20.53°S 49.32°W; Matão - 21.37°S 48.32°W) and Santa Catarina State (Piraí -26.30°S 48.85°W, Ribeirão da Ilha - 27.47°S-48.33°W; Aguaí - 28.25°S 49.32°W). The Procrustes ANOVA indicated significant variation in shape and size of the wings (p = 0.0088 e p < 0.0001), evidencing a more rounded shape in the southeast populations and more elongated shape in the south populations. The populations were clustered into two groups according to the State and the Discriminant Analysis showing that only 10.38% of the data were misclassified (Mahalanobis distance: 2.5064 and p < 0.0001). The Fourier Descriptors and Principal Component Analyses of the aedeagi (using the 5 main variables together) accounted for 90.58% of the contour's variation. We tested the equality of the strains by Kruskal-Wallis tests and we obtained significant difference to PC1 (H = 18.03, p = 0.0012), PC3 (H = 22.67, p = 0.0001) and PC5 (H = 18.03, p = 0.0012) 0.0012). With regard to the aedeagus, the most significant differences were detected in Aguaí population compared with the others. It is possible that the observed variation may be explained by environmental factors or others effects, such as natural selection, genetic drift, altitude, but apparently the environmental temperature is the main factor responsible for such variations. Other analyses, such as involving the use of the molecular markers and/or the analysis of reproductive features in intercrosses among the populations are being performed for a more robust interpretation of the results.

Keywords: populational differentiation, phenotypic plasticity, saltans subgroup, fragmentation Financial aid: FAPESP (2014/14059-0) and CNPq (4775524/2012-0)



What is the evolutionary story of the non-LTR *BS* element in *Zaprionus* species?

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TRANSPOSABLE ELEMENTS (TEs), repetitive sequences that can change position in the genome, are transmitted across generations via vertical inheritance, and the accumulation of evolutionary changes can lead to significant degradation and their extinction the species. However, due to its mobility, the TEs can be reintroduced in a species genome after being lost by degradation. The BS element is a non-LTR retrotransposon scarcely studied in the genus Drosophila. Moreover, no study of this element has been published focusing species of the genus Zaprionus, which also belongs to the family Drosophilidae. We searched for the occurrence of BS element in all 20 available Drosophila genomes, between them 13 belonging to the melanogaster group of Drosophila: D. erecta*, D. melanogaster*, D. simulans*, D. sechellia*, D. ananassae*, D. biarmipes, D. bipectinata, D. elegans, D. eugracilis, D. ficusphila, D. kikkawai, D. rhopaloa e D. takahashii and performed the search using as a full featured query the BS element (5,126 bp) of D. melanogaster. BS was found only in 8 species (D. simulans, D. sechellia, D. melanogaster, D. yakuba, D. erecta, D. pseudoobscura, D. persimillis, D. mojavensis). The species marked with * were previously shown to harbor BS by our research group. Additionally, we investigated the occurrence of BS in species of the genus Zaprionus. Sequence of BS in six Zaprionus species (Z. indianus, Z. africanus, Z. sepsoides, Z. gabonicus, Z. davidi and Z. ornatus) were amplified, cloned and sequenced and the phylogenetic and evolutionary relationships were reconstructed by neighbor-joining (p-distance model) using Mega 6, and the package Network. The reference sequence of BS (D. melanogaster) was downloaded from Repbase database as well as the Helena sequence of Drosophila simulans used as outgroup. The BS relationships do not reflect the species phylogeny, since all sequences of Zaprionus form a monophyletic group, clustered within the clade formed species of the *melanogaster* subgroup, supported by high bootstrap value (99%). Furthermore, the analyses showed sequences of Zaprionus are closely related to sequences of simulans complex. This relationship suggests an ancestral HT event from the melanogaster sub group, probably from the simulans complex ancestor to the Zaprionus subgenus ancestor, during the period of time when these two species groups overlapped geographically and temporally in tropical Africa. In order this hypothesis, and to have a broader understanding of the BS evolution, additional searches are being carried out trying to found this TE in other species of Zaprionus and Drosophila.

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