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Ecological Observations and Germination of *Sterculia chicha* Seeds Colonized by *Anastrepha bezzii*

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ABSTRACT: *Anastrepha bezzii* (Diptera: Tephritidae) larvae are common inside fruits of *Sterculia chicha* (Malvaceae) in Minas Gerais State, Brazil. The seed dormancy and attack by *A. bezzii* of the native species *S. chicha* can be detrimental to seed germination and thus the natural spread of this plant. The objectives of the current study were to compare ecological observations of *A. bezzii* and *S. chicha* in the field and laboratory, and to assess the germination of *S. chicha* colonized by *A. bezzii*. A total of five seeds that were either intact or punctured by *A. bezzii* larvae were removed per *S. chicha* fruit. The seeds were subjected to a germination test performed using one of four treatments: intact seeds without scarification (T1); intact seeds scarified for 3 min with sandpaper number 40 on one side of each seed (T2); drilled seeds without scarification (T3); and drilled seeds scarified for 3 min with sandpaper number 40 on one side of each seed (T4). The germination of *S. chicha* seeds was highest in the T2 group (72.6%), intermediate in the T1 (51.7%) and T4 groups (33.1%), and lowest in the T3 group (11.2%). The attack by *A. bezzii* of the fruits and seeds of *S. chicha* can be detrimental to seed germination and thus the further establishment of this native species in the field. The scarification of intact seeds of *S. chicha* for 3 min with sandpaper number 40 on one side of each seed can overcome the dormancy for its production in nurseries.

Key words: Damaged seeds, Malvaceae, Phytophagous insects, Seed germination, Tephritidae

INTRODUCTION

Sterculia chicha A. St. Hil. (Malvaceae) of the subfamily Sterculioideae, is a native to Brazil that can reach 30 m in height. The species occurs from central Brazil to the southeast of the country, in the states of Bahia, Espírito Santo, Goiás, Minas Gerais, Rio de Janeiro and São Paulo (Taroda and Gibbs, 1982). It is cultivated as an ornamental plant for shade and is suitable for use in reclamation projects. *Sterculia* species have a pan-tropical occurrence, with approximately 300 species distributed throughout tropical forests areas of Southeast Asia and South America (Eiras *et al.*, 2007). *Sterculia chicha* blooms from January to March and fruits between July and October, but with variation among regions (Taroda and Gibbs, 1982). Despite its importance and the widespread nature of its occurrence, there are few studies of the biology, physiology and ecology of this important genus (Eiras *et al.*, 2007). *Sterculia* seeds are consumed by both wildlife and humans, albeit raw, cooked or roasted by the latter. The seeds are tasty and rich in nutrients, although they have low sodium content, thus promoting the proper functioning of the cardiovascular system (Carvalho *et al.*, 2008; Fernandes and Silva, 2011). Honeybees, flies and wasps are common visitors to the flowers of this species. The seeds are surrounded by a protective gum that is found in the stem; the gum is used in the

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manufacture of cosmetics and in the food industry, mainly as an emulsifying agent (Zampa *et al.*, 2007; Eiras *et al.*, 2010). *Sterculia* trees are formed of a light wood that has only a low durability when exposed to extremes of weather. The wood is used for internal features, such as liners and boards, in the manufacture of matchsticks and has potential for cellulose production. When opened, its fruits are often used as ashtrays.

Tephritidae insects occur throughout temperate and tropical areas worldwide, where they damage the seeds of native and cultivated plants (Honek and Martinkova, 2005; Koprdova and Martinkova, 2006; Knodel *et al.*, 2011). Species of this family are distributed in approximately 500 genera, five of which have economic importance (Pearson and Callaway, 2008). In Brazil, *Anastrepha* Schiner, 1868 (Diptera: Tephritidae) is an important genus as it includes pest species that damage fruits and seeds, thus impacting on the export of fresh fruits and affecting the germination of native plants (Uramoto *et al.*, 2008; Silva *et al.*, 2010; Ronchi-Teles *et al.*, 2011). The sampled number of fruit flies and the species diversity can differ among even neighboring orchards owing to ecological differences among orchards and the surrounding areas (Aluja *et al.*, 1996, 1997; Uchôa and Nicácio, 2010). Species of *Anastrepha* have adapted to their hosts as a result of coevolution, although with different degrees of specialization (Ovruski *et al.*, 2007; Boykin *et al.*, 2010). *Anastrepha bezzii* Lima, 1934 occurs in Mexico (Chiapas) to Venezuela, following the Andes to Brazil (Goiás, Mato Grosso do Sul, Minas Gerais, and Rio de Janeiro States). It is known to occur in *S. chicha* fruits from Viçosa, Minas Gerais State, Brazil. However, it was thought to be rare in Brazil as only three specimens were known: the holotype male, collected in Manguinhos, Rio de Janeiro State, Brazil in 1902 and deposited in the Instituto Oswaldo Cruz; a female collected in Campinas, São Paulo State, Brazil, which was apparently lost, and another female that was collected in Viçosa, Minas Gerais State, Brazil in 1938 and deposited in the Museu de Zoologia of Universidade de São Paulo (USP) in Piracicaba, São Paulo State, Brazil. This female was redescribed, which, in addition to confirming the occurrence of *A. bezzii* in Brazil and on *S. chicha*, also showed that it occurs in other countries, including Costa Rica, Panama, Mexico, Peru and Venezuela, on *Terminalia catappa* L. (Combretaceae) (native to India and New Guinea) and *Sterculia apetala* (Jacq.) Karst (Malvaceae) (native to America) (Fig. 1) (Zucchi, 1984).

The objectives of the current study were to report ecological observations on the interaction between *S. chicha* and *A. bezzii* in the field and laboratory and to assess the seed germination of this plant colonized by larvae of this fly. Germination test of *S. chicha* seeds colonized by *A. bezzii* larvae is important to recommend a method of breaking dormancy for its production in nurseries.

MATERIALS & METHODS

Fifty fruits that were ripe and open were collected from the ground in May, 2011 after falling from a mature tree of *S. chicha* cultivated in the herbarium of Universidade Federal de Viçosa (UFV) in Viçosa, Minas Gerais State, Brazil (20º45'S, 42º51'W,

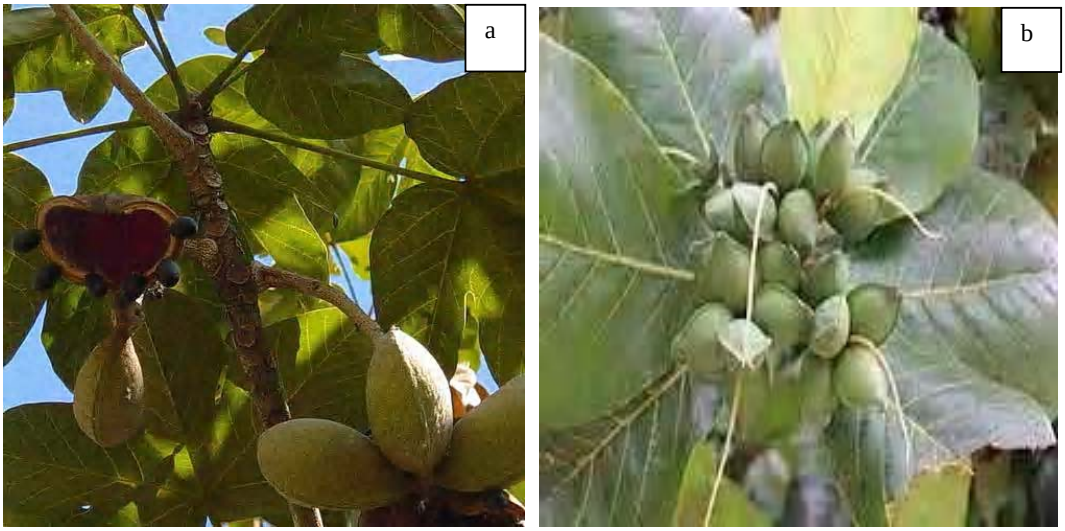

Fig. 1. Host plants of *Anastrepha bezzii* (Diptera: Tephritidae) according to Zucchi (1984): *Sterculia apetala* (Malvaceae) (a) **and *Terminalia catappa* (Combretaceae)** (b) **seeds**

651m) (Fig. 2). The fruits were placed in plastic cups and taken to the Laboratório de Controle Biológico de Insetos (LCBI). Tephritidae larvae were removed from the inside of the *S. chicha* fruits and/or seeds using a brush and placed in plastic cups filled with fine sand moistened with water and containing some *S. chicha* seeds. The cups were kept in an acclimatized room at 25 ± 1°C, under a 12-h photoperiod and 70 ± 10% relative humidity (RH) until the adults emerged.

Five seeds that were either intact or punctured by Tephritidae larvae were removed per fruit of *S. chicha*; seeds that were cracked and/or broken were discarded. The material was treated with the fungicide Derosal 500 SC® (Manufacturer: Bayer S/A, Municipality: Curitiba, State: Paraná, Country: Brazil) at a concentration of 3 ml/L water and subjected to germination tests using one of four treatments. Four repetitions were used per treatment, each one with 25 seeds: (T1) intact seeds without scarification; (T2) intact seeds with scarification for 3 min with sandpaper number 40 on one side of each seed; (T3) drilled seeds without scarification; and (T4) drilled seeds with scarification for 3 min with sandpaper number 40 on one side of each seed, using methodology proposed for *Sterculia foetida* L. (Malvaceae) (Santos *et al.*, 2004). This scarification process breaks the seed coat, making it permeable, allowing imbibition. The mechanical scarification was tested to determine a simple, effective and inexpensive method and that could be used by nurseries, without damage to them and to the environment.

Sterculia chicha seeds were soaked in distilled water for 24 h and placed on paper rolls inside plastic boxes. These boxes were kept in an incubator at 25 ± 1°C and under constant light. The paper rolls were moistened with distilled water throughout the course of the experiment. Germination was evaluated 35 days after the beginning of the experiment (Santos *et al.*, 2004). Data were submitted to variance analysis (ANOVA) and means were compared by Tukey test at 5% probability, using the software SAEG (2007) (UFV). Germination data were transformed in Arcsin "%/100. Female Tephritidae were kept for 72 h inside rearing cages until they acquired the characteristic color of the adult body and wings. Specimens were then killed and sent to the Instituto Biológico in Campinas, São Paulo State, Brazil and to Universidade Federal Rural do Semi-Árido in Mossoró, Rio Grande do Norte State, Brazil, where they were identified as *A. bezzii* and deposited in the appropriate collections (Fig. 3).

General ecological observations (i.e. characteristics of damage, presence of other insects, the occurrence of oviposition and pupation) were made of the interaction between seeds of *S. chicha* and *A. bezzii* larvae. These were recorded after daily morning observations in the field during April and June 2011. Some larvae of *A. bezzii* were placed in rearing cages and kept at 25 ± 1°C, a 12-h photoperiod and 70 ± 10% RH in the LCBI at UFV. These larvae were also observed. Fruits and seeds of *S. chicha* damaged by *A. bezzii* were stored in a cold chamber and were deposited in the collection.

RESULTS & DISCUSSION

The germination of *S. chicha* seeds was significantly higher in intact seeds subject to treatment T2 (72.6 ± 4.9%), intermediate in intact seeds subject to treatment T1 (51.7 ± 5.8%) and drilled seeds subject to treatment T4 (33.1 ± 6.1%), and lowest in drilled

Fig. 2. Mature tree of *Sterculia chicha* (Malvaceae) (a) **and its seeds damaged by *Anastrepha bezzii* (Diptera: Tephritidae) larvae, besides others insects and write fungi** (b) **in Viçosa, Minas Gerais State, Brazil**

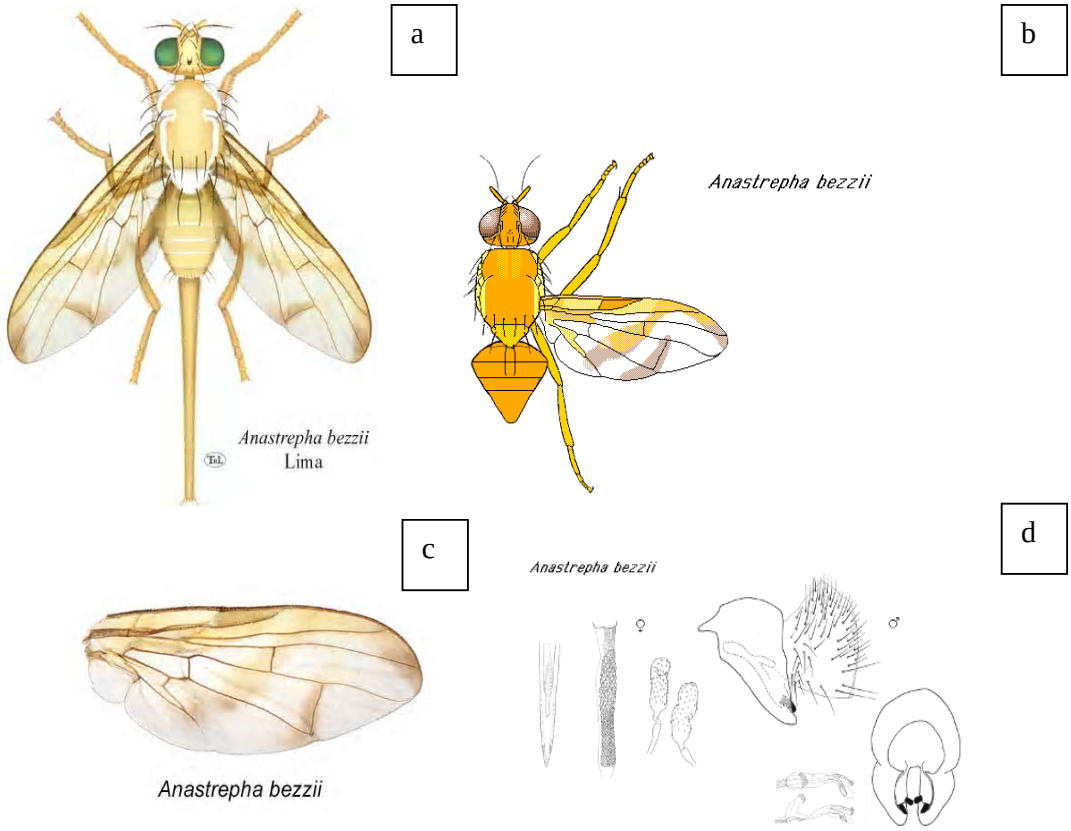

Fig. 3. Habitus female (dorsal), wing (a)**; habitus male (dorsal), wing** (b)**, wing** (c) **and female terminalia, male terminalia** (d)

seeds subject to treatment T3 (11.2 ± 9.8%) ($P < 0.05$; $F = 12.03$; *Variation coefficient*= 19.8%). The fact that the highest rate of germination occurred in intact seeds of *S. chicha* suggests that *A. bezzii* larvae reduce the germination rate of the seeds of this plant. The percentage of *S. chicha* seeds damaged by *A. bezzii* larvae was lower than in an experiment performed in 1993 in Viçosa, Minas Gerais State, Brazil because the females may be deposited more eggs in fewer seeds in the current study. In addition, larvae emerged from eggs that were not contained within the fallen fruits collected (Santos *et al.*, 1993). Larvae of *A. bezzii* consume most of the endosperm of *S. chicha* seeds, rendering them unable to germinate. The interior of damaged fruits showed the remains of consumed seeds and the light-brown feces of the larvae. Seeds that had been attacked were loose inside the fruits, which made them easy to distinguish.

The high rate of germination of intact S. chicha seeds that had been scarified confirms that such treatment is suitable for use in the management of the waterproofed seeds of the Malvaceae in the field (Baskin and Baskin, 1997). It is likely that scarification broke the dormancy of the seed coat, as has been reported for seeds of S. foetida subjected to the treatment (Santos et al., 2004). Mechanical scarification increases the permeability of the seed coat to water, allowing soaking and accelerating the germination process, although it can cause the breakage and cracking of seeds (Frank and Baseggio, 1998). The rate of germination of the native plants Erytrina velutina Willd. (Fabaceae) and Leucaena diversifolia (Schltoll.) Benth. (Fabaceae) increased after their seeds were scarified (Silva and Matos, 1993; Bertalot and Nakagawa, 1998).

Other phytophagous insects were found inside the fruits of *S. chicha*. Including larvae of a species of Scolytidae, which fed on the exocarp, and nymphs and adults of *Dysdercus* sp. (Hemiptera: Pyrrhocoridae), which sucked the exudate after dehiscence of the fruits. In addition, hyphae of white fungi were observed colonizing the *S. chicha* seeds. Similar observations were made in a study of *S. chicha* fruits carried out in 1993 (Santos *et al.*, 1993). In other studies, seeds of the native plant *Sclerolobium* sp. (Fabaceae) collected in two localities of Viçosa, Minas Gerais State, Brazil

were found to be damaged by *Sennius amazonicus* Pic., 1929 (Coleoptera: Bruchidae) and two species from the genera *Sennius* and *Amblycerus* (Coleoptera: Bruchidae) (Santos *et al.*, 1997); in addition, seeds of *Melanoxylon brauna* Schott (Fabaceae) were found to be infested by two species of beetle, *Sennius cupreatus* and *Sennius spodiogaster* Kingsolver, 1987 (Coleoptera: Bruchidae) (Santos *et al.*, 1991).

Larvae of *A. bezzii* were found inside both the green and the ripe fruits of *S. chicha*, suggesting that females of this insect learn to oviposit inside fruits containing seeds. The insects occurred also inside fruits, in the space among seeds. Different larval instars of *A. bezzii* were found, suggesting that the females oviposit at different times. More larvae were concentrated inside the fruits, where they were attached to the internal capsule and left to pupate. These larvae remained inside the seeds awaiting fruit dehiscence, following which they pupated once they reached the soil. Some larvae left the seeds before the fruits had opened, becoming attached to the mucilage (sticky exudation) found inside the fruits, where they tended to perish. Larvae that were inside fruits that had not opened also perished, probably because they were unable to penetrate the exocarp. The larvae are immobile before pupation and they were white or slightly beige in color, turning, over time to yellow then black with orange stripes. At this stage in the laboratory, they showed a tendency to aggregate in the same place before pupating.

CONCLUSION

Anastrepha bezzii attacks fruits of *S. chicha* in Viçosa, Minas Gerais State, Brazil and reduces the germination of seeds of this plant, although scarification helps to break dormancy and aids further establishment of this plant. Ecological observations on the interaction between *S. chicha* and *A. bezzii* in the field and laboratory were also described. These results suggest that scarification of intact seeds of *S. chicha* for 3 min with sandpaper number 40 on one side of each seed can overcome the dormancy for its production in nurseries. The next step in investigating this interaction is to quantify the observations and to study a method of control for *A. bezzii*.

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REFERENCES

Aluja, M., Celedonio-Urtado, H., Liedo, P., Cabrera, M., Castillo, F., Guillen, J. and Rios, E. (1996). Seasonal population fluctuations and ecological implications for management of *Anastrepha* fruit flies (Diptera: Tephritidae) in commercial mango orchards in southern Mexico. J. Econ. Entomol., **89 (3)**, 654-667.

Aluja, M., Jimenez, A., Camino, M., Pinero, J., Aldana, L., Castrejon, V. and Valdes, M. E. (1997). Habitat manipulation to reduce papaya fruit fly (Diptera: Tephritidae) damage: Orchard design, use of trap crops and border trapping. J. Econ. Entomol., **90 (6)**, 1567-1576.

Baskin, J. M. and Baskin, C. C. (1997). Methods of breaking seed dormancy in endangered species *Iliamna corei* (Sherff) Sherff (Malvaceae), with special attention to heating. Nat. Areas J., **17 (4)**, 313-323.

Bertalot, M. J. A. and Nakagawa, J. (1998). Overcoming dormancy of *Leucaena diversifolia* (Schlecht.) Bentham Seeds K 156. Rev. bras. sementes, **20 (1)**, 39-42.

Boykin, L. M., Shatters, R. G., Hall, D. G., Dean, D. and Beerli, P. (2010). Genetic variation of *Anastrepha suspensa* (Diptera: Tephritidae) in Florida and the Caribbean using microsatellite DNA markers. J. Econ. Entomol., **103 (6)**, 2214-2222.

Carroll, L. E., White, I. M., Freidberg, A., Norrbom, A. L., Dallwitz, M. J. and Thompson, F. C. (2006). Pest fruit flies of the world. Version: 8th available in http://delta-intkey.com/ffa/www/ana_bezz.htm

Carvalho, M. G., Costa, J. M. C., Souza, V. A. B. and Maia, G. A. (2008). Evaluation of some physical and nutritional parameters of almonds of chicha, sapucaia and castanha-do-gurgucia. Rev. Ciênc. Agron., **39 (4)**, 517-523.

Eiras, C., Santos, A. C., Zampa, M. F., Brito, A. C. F., Constantino, C. J. L., Zucolotto, V. and Santos, J. R. (2010). Natural polysaccharides as active biomaterials in nanostructured films for sensing. J. Biomater. Sci.-Polym. Ed., **21 (11)**, 1533-1543.

Eiras, C., Passos, I. N. G., Brito, A. C. F., Santos, J. R., Zucolotto, V., Oliveira, O. N., Kitagawa, I. L. and Constantino, C. J. L. (2007). Electroactive nanocomposites made of poly(O-methoxyaniline) and natural polysaccharides. Quim. Nova, **30 (5)**, 1158-1162.

Franke, L. B. and Baseggio, J. (1998). Dormancy overcoming in *Desmodium incanum* DC. and *Lathyrus nervosus* Lam. seeds. Rev. bras. sementes, **20 (2)**, 182-186.

Honek, A. and Martinkova, Z. (2005). Pre-dispersal predation of *Taraxacum officinale* (dandelion) seed. J. Ecol., **93 (2)**, 335-344.

Knodel, J. J., Ganehiarachchi, G. A. S. M., Beauzay, P. B., Chirumamilla, A. and Charlet, L. D. (2011). Impact of planting dates on a seed maggot, *Neotephritis finalis* (Diptera: Tephritidae), and sunflower bud moth (Lepidoptera: Tortricidae) damage in cultivated sunflower. J. Econ. Entomol., **104 (4)**, 1236-1244.

Koprdova, S. and Martinkova, Z. (2006). Pre-dispersal predation and seed damage of *Centaurea scabiosa* L. (Asteraceae). J. Plant Dis. Prot., **20 (1)**, 305-308.

Lima, A. C. (1934). Fruit flies of the genus *Anastrepha* Schiner, 1868 (Diptera: Tephritidae). Mem. Inst. Oswaldo Cruz, **28 (4)**, 487-575.

Norrbom, A. L. (1991). The species of *Anastrepha* (Diptera: Tephritidae) with a *Grandis*-type wing pattern. Proc. Entomol. Soc. Wash., **93 (1)**, 101-124.

Ovruski, S. M., Wharton, R. A., Rull, J. and Guillien, L. (2007). *Aganaspis alujai* (Hymenoptera: Figitidae: Eucoilinae), a new species attacking *Rhagoletis* (Diptera: Tephritidae) in the neotropical region. Fla. Entomol., **90 (4)**, 626-634.

Pearson, D. E. and Callaway, R. M. (2008). Weed-biocontrol insects reduce native-plant recruitment through second-order apparent competition. Ecol. Appl., **18 (6)**, 1489-1500.

Ronchi-Teles, B., Dutra, V. S., Costa, A. T. P., Aguiar-Menezes, E. D., Mesquita, A. C. A. and Silva, J. G. (2011). Natural host plants and native parasitoids associated with *Anastrepha pulchra* and other *Anastrepha* species (Diptera: Tephritidae) in Central Amazon, Brazil. Fla. Entomol., **94 (2)**, 347-349.

SAEG, (2007). Sistema para Análises Estatísticas, Versão 9.1: Fundação Arthur Bernardes - UFV - Viçosa.

Santos, T. O., Morais, T. G. O. and Matos, V. P. (2004). Mechanical scarification on seeds of *Sterculia foetida* L. Rev. Arv., **28 (1)**, 1-6.

Santos, G. P., Anjos, N., Zanuncio, J. C. and Assis Júnior, S. L. (1993). Danos e aspectos biológicos de *Anastrepha bezzi* Lima, 1934 (Diptera: Tephritidae) em sementes de *Sterculia chicha* St. Hill. (Sterculiaceae). Rev. Bras. Entomol., **37 (1)**, 15-18.

Santos, G. P., Zanuncio, T. V., Assis Júnior, S. L. and Zanuncio, J. C. (1997). Damage by *Sennius amazonicus*, *Sennius* sp. and *Amblycerus* sp. (Coleoptera: Bruchidae) on *Sclerolobium* sp. (Leguminosae) seeds. Rev. Biol. Trop., **45 (2)**, 883-886.

Santos, G. P., Zanuncio, J. C., Anjos, N., Silva, J. C. and Alves, J. B. (1991). Danos causados por *Sennius cupreatus* e *S. spodiogaster* (Coleoptera: Bruchidae) em sementes de *Melanoxylon braunea*. Rev. Ceres, **38 (18)**, 315-322.

Silva, L. M. M. and Matos, V. P. (1993). Estudo sobre dormência de sementes de mulungu (*Erythrina velutina* Willd.): viabilidade e presença de inibidores. Ci. agríc., **2 (1)**, 29-40.

Silva, A. G. D. E. and Fernandes, K. F. (2011). Chemical composition and antinutrients of raw and roasted chicha almonds (*Sterculia striata* A. St. Hill & Naudin). Rev. Nutr., **24 (2)**, 305-314.

Silva, J. G., Dutra, V. S., Santos, M. S., Silva, N. M. O., Vidal, D. B., Nink, R. A., Guimarães, J. A. and Araujo, E. L. (2010). Diversity of *Anastrepha* spp. (Diptera: Tephritidae) and associated braconid parasitoids from native and exotic hosts in Southeastern Bahia, Brazil. Environ. Entomol., **39 (5)**, 1457-1465.

Taroda, N. and Gibbs, P. E. (1982). Floral biology and breeding system of *Sterculia chicha* St Hil (Sterculiaceae). New Phytol., **90 (4)**, 735-743.

Uchôa, M. A. and Nicácio, J. (2010). New records of Neotropical fruit flies (Tephritidae), lance flies (Lonchaeidae) (Diptera: Tephritoidea), and their host plants in the South Pantanal and adjacent areas, Brazil. Ann. Entomol. Soc. Am., **103 (5)**, 723-733.

Uramoto, K., Martins, D. S. and Zucchi, R. A. (2008). Fruit flies (Diptera, Tephritidae) and their associations with native host plants in a remnant area of the highly endangered Atlantic Rain Forest in the State of Espirito Santo, Brazil. Bull. Entomol. Res., **98 (5)**, 457-466.

Zampa, M. F., Brito, A. C. F., Kitagawa, I. L., Constantino, C. J. L., Oliveira, O. N., Cunha, H. N., Zucolotto, V., Santos, J. R. and Eiras, C. (2007). Natural gum-assisted phthalocyanine immobilization in electroactive nanocomposites: Physicochemical characterization and sensing applications. Biomacromolecules, **8 (11)**, 3408-3413.

Zucchi, R. A. (1984). Redescription of the female of *Anastrepha bezzi* Lima, 1934 (Diptera: Tephritidae). Rev. Bras. Entomol., **28 (4)**, 561-562.