1(3S,6E)-Nerolidol-mediated rendezvous of Cyclocephalini beetles,

2*Cyclocephala paraguayensis*, in bottle gourd flowers

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33 Abstract

34 Cyclocephalini beetles of genus *Cyclocephala* (Coleoptera: Melolonthidae:
35 Dynastinae) use flowers of some plants as food, shelter, and mating stands. However,
36 little is known about floral scent chemistry involved in this interaction. Here we show
37 that a sesquiterpene alcohol mediates attraction of *Cyclocephala paraguayensis* Arrow
38 on bottle gourd flowers, *Lagenaria siceraria* (Cucurbitaceae). Both males and females
39 started to aggregate on flowers at twilight; after that, mating began and remained for
40 the entire night. The major constituent of the airborne volatiles from *L. siceraria* was
41 fully characterized as (3S,6E)-nerolidol, which elicited electroantennographic
42 responses on male and female antennae. Field bioassays showed that traps baited
43 with the natural stereoisomer or a mix of nerolidol isomers captured significantly more
44 males and females of *C. paraguayensis* than control traps. Analysis from the gut
45 content of these Cyclocephalini beetles showed the presence of pollen, suggesting that
46 they also use bottle gourd flowers for their nourishment. Taken together, these results
47 suggest that (3S,6E)-nerolidol plays an essential role in the reproductive behavior of *C.*
48 *paraguayensis* by eliciting aggregation, mating, and feeding.

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Introduction

Floral scents have been reported as important assets for the visitation of beetles of the tribe Cyclocephalini (Melolonthidae: Dynastinae) [1–4]. The interaction of beetles and flowers may result in mutualistic pollination typically by the cantharophilous floral syndrome of basal angiosperms, whose flowers are usually bisexual, protogynous, very scented, and thermogenic [4,5]. The Cyclocephalini beetles are primarily attracted by strong odors released by flowers as chemical cues to indicate a rendezvous site for food, shelter, and mating stands [4,6,7].

The floral volatile organic compounds (VOCs) in the plant families of Araceae, Annonaceae, Arecaceae, and Magnoliaceae, have been demonstrated to be comprised of pyrazines, ketones, esters, and methoxystyrenes [8–13]. By contrast, VOCs produced by eudicot flowers that are visited by Cyclocephalini beetles of the genus *Cyclocephala* have never been identified.

Cyclocephala paraguayensis Arrow, is a Neotropical species that is frequently found on eudicot plant species [14,15]. Here, we observed this beetle visiting flowers of *Lagenaria siceraria* (Molina) Standle (Cucurbitaceae), which incidentally represents a new host flower record for the beetle family Melolonthidae. This plant is originated from Africa and nowadays is spread worldwide [16]; it produces white flowers with separate sexes in the same plant (monoecious) but can assume andromonoecious form, when hermaphrodite flowers are present [17]. Anthesis occurs throughout the night, at which time the lepidopteran pollinators, specially hawkmoths, visit the flowers [18].

We surmised that the volatile components of bottle gourd flowers mediate aggregative behavior of *C. paraguayensis* for feeding and mating. We studied the floral scent chemistry of *L. siceraria* flowers and showed that (3S,6E)-nerolidol plays an important role in the reproductive behavior of *C. paraguayensis* by eliciting aggregation, mating, and feeding.

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88 Results

89 Sexual and feeding behavior of insects

90 The behavior of *C. paraguayensis* visitation on *L. siceraria* flowers was
 91 observed under field conditions at night for four hours (7:00 p.m to 11:00 p.m.). Beetles
 92 activity started by the twilight (~7:30 p.m.), when males of *C. paraguayensis* were the
 93 first to land on *L. siceraria* flowers (n = 36; two-tailed Exact Binomial test; $P = 0.011$). At
 94 8:00 p.m., the number of visiting males and females was 18.50 ± 5.97 (mean $\pm$ SD),
 95 increasing until 11:00 p.m. to 26.75 ± 7.46 . Paired males and females of *C.*
 96 *paraguayensis* (2.25 ± 3.2) were present on bottle gourd flowers from 8:00 p.m. until
 97 the end of the observation period. Both sexes of *L. siceraria* flowers were visited.
 98 Seventy-five percent of bottle gourd flowers harbored more than one mating couple,
 99 thus, suggesting that aggregation on bottle gourd flowers created mating opportunities
 100 for the beetles.

101 These Cyclocephalini beetles usually remained inside the floral receptacles until
 102 the next morning (Fig 1A-B). Bottle gourd flowers presented gnawing marks or even
 103 major damages resulted from beetles feeding (Fig 1E-F). Of note, pollens of these
 104 flowers were found in the gut of beetles (80% females; 50% males, n = 10) when
 105 dissected (Fig 1G-H).

106

107 **Fig 1. Sexual and feeding behavior of Cyclocephalini beetle, *Cyclocephala***
 108 ***paraguayensis*, in bottle gourd flowers, *Lagenaria siceraria*.** A) Aggregating and
 109 mating on flowers at night; B) *C. paraguayensis* inside senescent flowers in the next
 110 morning of beetle's attraction; C) Non-damaged *L. siceraria* flower in the field; D)
 111 Stereoscopic cut view of a non-damaged *L. siceraria* flower; E) *L. siceraria* flower
 112 damaged by *C. paraguayensis* in the field; F) Stereoscopic cut view of a damaged
 113 flower showing beetle gnawing and excrement signal as a result of feeding activity; G)

114 Fresh pollen grain of *L. siceraria*; H) Pollen grains of *L. siceraria* extracted from *C.*
115 *paraguayensis* gut (black triangles indicate the pollen grains).

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117 **Floral scent identification**

118 Gas chromatography linked to electroantennographic detection (GC-EAD)
119 analysis of airborne volatiles collected from *L. siceraria* showed the presence of a
120 prominent, antennally active peak at 20.5 min (Fig 2).

121

122 **Fig 2. GC-EAD analysis of floral scents extract of *Lagenaria siceraria* by using**
123 **male and female antenna of Cyclocephalini beetle, *Cyclocephala paraguayensis*,**
124 **as sensing elements. “FID” flame-ionization detection.**

125

126 The prominent compound was tentatively identified by GC-MS as a nerolidol
127 isomer, which led us to conduct *cis* and *trans* identification followed by chiral analysis.
128 The EAD active compound was then identified as *trans*nerolidol (i.e., (6*E*)-nerolidol, IR
129 = 1568) by comparing the mass spectra and retention indexes of the natural product
130 and authentic standards (Fig 3a-f). Lastly, chiral analysis using nerolidol standards led
131 to the full characterization of the natural product as (3*S*,6*E*)-nerolidol (Fig 3g).

132

133 **Fig 3. Analysis of bottle gourd flowers, *Lagenaria siceraria*, extract, and synthetic**
134 **isomers of nerolidol by gas chromatography equipped with a chiral column. a) Z**
135 **and E-nerolidol (left to right) isomers of nerolidol (a mix of isomers); b) Z-nerolidol**
136 **isomers; c) Synthetic (3*R*,6*E*)-nerolidol (gift from Dr. Robert Hanus, Czech Republic);**
137 **d) (E)-nerolidol from Sigma-Aldrich; e) Mix of 40 ng from each (3*R*,6*E*)-nerolidol and**
138 **(3*S*,6*E*)-nerolidol; f) Natural extract from VOCs of *L. siceraria* flowers; g) Coinjection of**
139 **“e” and “f” chromatogram extracts.**

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141 Quantification of VOCs from male and female flowers showed that (3S,6E)-
142nerolidol comprised 97% of the volatile bouquet and was released at the rate of ~1.3
143µg/hour under field conditions.

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145 **Field evaluation of synthetic nerolidol**

146 In our first field trial, a total of six adults of Cyclocephalini beetle, *C.*
147*paraguayensis*, landed on nerolidol-treated dummies, whereas no beetle was observed
148on control dummies (two-tailed Exact Binomial test; $P = 0.031$).

149 In the second field experiment, we evaluated the response of adults of *C.*
150*paraguayensis* to nerolidol-baited traps in two different locations: Cassilândia, State of
151Mato Grosso do Sul and Piracicaba, State of São Paulo. In both locations, traps baited
152with nerolidol (a mix of isomers) captured significantly more beetles than control traps.
153In Cassilândia, 280 adults of *C. paraguayensis* were captured by nerolidol-baited traps
154during the tests, with no beetles captured in control traps (two-tailed Exact Binomial
155test; $P < 0.001$; Fig 4A). Although total captures in Piracicaba ($n = 32$) were lower than
156in Cassilândia, nerolidol-baited traps captured significantly more beetles than control
157traps (two-tailed Exact Binomial test; $P < 0.001$). Interestingly, the sex ratio of captured
158beetles did not differ significantly from the 1:1 ratio in both Cassilândia (51.4% females;
15995% Clopper-Pearson exact confidence interval: 0.454-0.574; $P = 0.632$) and
160Piracicaba (41.4% females; 95% Clopper-Pearson exact confidence interval: 0.235-
1610.610; $P = 0.353$).

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164**Fig 4. Mean number of Cyclocephalini beetle, *Cyclocephala paraguayensis*,**
165**caught per replicate in field experiments conducted in Cassilândia, MS, Brazil.**

166Treatments: Nerolidol (a mix of isomers) = mixture of four isomers of nerolidol; (3S,
1676E)-nerolidol = synthetic nerolidol, which is identical to the stereoisomer of nerolidol
168produced by *L. siceraria* flowers. A) Mean number of adults of *C. paraguayensis* caught
169per replicate of nerolidol-baited traps and controls. *Treatment is significantly different
170at $P < 0.05$ according to Exact binomial test goodness-of-fit; B) Mean number of adults
171of *C. paraguayensis* caught by traps containing synthetic nerolidol isomers. Treatments
172had a significant effect on attraction of Cyclocephalini beetles (Fiedman's $Q_{2,30} =$
17320.002, $P < 0.001$; $n = 10$ replicates). Means followed by the same letter are not
174significantly different (REGWQ test, $P > 0.05$).

175

176 In the third field tests, we compared captures of *C. paraguayensis* in traps
177baited with nerolidol (a mix of isomers), (3S,6E)-nerolidol, and control traps.
178Throughout these experiments, a total of 1,146 beetles were captured in nerolidol traps
179(i.e., traps baited with either enantiopure nerolidol, or a mix of isomers). By contrast,
180only two individuals (males) were caught in control traps. Although treatment and
181control were significantly different ($Q_{2,30} = 20.002$; $P < 0.001$; Fig 4B), there was no
182statistical difference in catches by the mix of isomers or (3S,6E)-nerolidol (Fig 4B). We,
183therefore, concluded that the other isomers were neither attractants nor inhibitors. Of
184note, the sex ratio of the captured beetles in the two nerolidol treatments were
185somewhat female-biased (mix of isomers = 54.4% female, 95% Clopper-Pearson exact
186confidence interval: 0.509-0.578; $P = 0.012$; and (3S,6E)-nerolidol = 58.4% female,
1870.544-0.622; $P < 0.0001$). However, as we did not measure the sex ratio of the field
188populations while conducting bioassays, it is not possible to assert that traps captured
189significantly more female than male beetles.

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192 Discussion

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194 Our results suggest that (3S,6E)-nerolidol mediates aggregation of the
195 Cyclocephalini beetle, *C. paraguayensis*, on bottle gourd flowers, *L. siceraria*. We
196 showed that this sesquiterpene alcohol represents the major VOC emitted by the floral
197 scents of *L. siceraria*, and field tests demonstrated that nerolidol-baited traps captured
198 significantly more beetles than control traps. Surprisingly, field tests showed captures
199 in traps baited with racemic nerolidol (a mix of four isomers) did not differ significantly
200 from those in traps having the enantiopure nerolidol as lure. Thus, the three other non-
201 natural isomers of nerolidol are neither attractant nor deterrent.

202 We, therefore, concluded that *C. paraguayensis* rendezvous in *L. siceraria* is
203 mediated by nerolidol. There is no evidence in the literature indicating that chemical
204 communication in *C. paraguayensis* involves sex pheromones. If it does, it is unlikely
205 that they need a long-range aggregation-sex pheromone (sensu Cardé 2014) [19] as
206 nerolidol mediates aggregation of both sexes. It is conceivable, however, that *C.*
207 *paraguayensis* utilizes a hitherto unknown short-range, or contact sex pheromone for
208 species and/or sex recognition.

209 In this study we showed that *C. paraguayensis* also uses bottle gourd flowers of
210 *L. siceraria* for nourishment given that pollen grains were found in the gut epithelium of
211 males and females. The energy-rich pollen grains may increase the fitness of these
212 Cyclocephalini beetles for mating and oviposition [4]. Our findings do not provide
213 evidence for a mutualistic insect-plant relationship, as previously suggested for other
214 anthophilous cyclocephalines [10,12]. The floral biology traits of cucurbits seem to be
215 more suitable for pollination by bees [20], and flowers are particularly successful
216 pollinated by hawkmoths [18]. Additionally, *C. paraguayensis* visit caused damage to *L.*
217 *siceraria* flowers, although the effects of these damages on the development of fruits
218 remain to be investigated.

219 **Methods**

220 **Sexual and feeding behavior of insects**

221 The behavior of Cyclocephalini beetle, *C. paraguayensis*, was observed in
222 bottle gourd flowers of *L. siceraria* in a growing area owned by the University of São
223 Paulo, Piracicaba, Brazil (22°42'47.1" S, 47°37'32.7" W). We initially recorded the
224 arrival time of the first beetles on flowers of *L. siceraria*, so the observations in the
225 subsequent days started 30 min earlier. Flowers with insects were marked, and the
226 following parameters were evaluated every 30 min from 7:00 p.m. to 11:00 p.m.: sex of
227 visited flowers; the total number of insects landed per flower; and the number of mating
228 couples per flower. We repeated these observations for 4 d in the same *L. siceraria*
229 area.

230 Additionally, a sample of 10 males and females of *C. paraguayensis* present
231 within the flowers were taken to verify the presence of pollen in the gut content. Insects
232 were dissected with sharp forceps, and pieces of the midgut and/or the hindgut
233 epithelium were removed and immersed in 0.9% sodium chloride solution. Next, the
234 material was analyzed under a light microscope at 20X magnification. Fresh pollen
235 from *L. siceraria* flowers was prepared in glass slides with the saline solution as
236 reference material.

237

238 **Sampling floral VOCs**

239 VOCs from *L. siceraria* flowers were conducted outdoors in the above-
240 mentioned bottle gourd plantation and the laboratory through dynamic headspace
241 extraction. In the field, VOCs were collected from individual male and female *L.*
242 *siceraria* flowers recognized according to morphological features [17]. These flowers
243 were enclosed within a 17 × 20 cm polyester oven bags (Wyda®, Sorocaba, São Paulo,
244 Brazil). Collectors consisted of glass pipets packed with 30 mg of the adsorbent
245 polymer Hayesep® Q (80/100 mesh; Supelco, Bellefonte, PA, USA), which were
246 attached to the bag outlets. Charcoal-filtered air was pushed with a portable pump
247 through the bag and collectors at ~200 mL/min. Bottle gourd flowers were aerated from

2487:30 p.m. to 08:30 p.m., which corresponded to the active period of the beetles.
 249 Trapped VOCs were eluted from collectors using 0.3 mL of distilled hexane into 2 mL
 250 amber vials that were stored at -30 °C until analysis. Aerations of empty bags were
 251 made in parallel to monitor system contaminants. In the same fashion, VOCs were
 252 collected from four detached flowers inside a vertical glass jar (55 cm long × 8 cm id)
 253 under laboratory conditions (23 ±2 °C; 60 ±10% RH). Collectors attached to the jar
 254 outlets were packed with 150 mg of adsorbent polymer. Charcoal-filtered air was
 255 pushed through jars and collectors at ~300 mL/min, and collections were made
 256 overnight for up to 11 h. Trapped VOCs were eluted from collectors using 1.5 mL of
 257 distilled hexane into 2 mL amber vial. To use these extracts for GC-EAD analysis, the
 258 resulting aliquots were then concentrated to 500 µL under a gentle flow of N₂.

259

260 **GC- EAD recordings**

261 Extracts of VOCs from *L. siceraria* flowers were analyzed by GC-EAD, i.e., gas
 262 chromatography (Shimadzu GC-2010 gas chromatograph, Shimadzu Corp., Kyoto,
 263 Japan) linked to an electroantennographic detector system (EAD, Ockenfels Syntech,
 264 Buchenbach, Germany) using *C. paraguayensis* antenna as the sensing element. The
 265 GC was equipped with a low polarity stationary phase column (30 m × 25 µm × 25 mm;
 266 Rtx-5; RESTEK, Bellefonte, PA, USA) connected to a stainless steel crosshead split
 267 with two deactivated columns for conducting effluent to FID and EAD detectors at the
 268 ratio of 1:3. Helium was used as the carrier gas at a linear velocity of 30.3 cm/s, and
 269 98.3 kPa was set for column head pressure. EAD preparations consisted of male or
 270 female excised antennae, and electrodes were made of glass pipette/gold wire filled
 271 with a Beadle-Ephrussi Ringer solution (3.75 g NaCl, 0.175 g KCl, 0.14g CaCl₂·2H₂O,
 272 500 mL H₂O). The base of the antenna was inserted into the indifferent electrode, and
 273 the tip of antennal lamellae was connected to the recording electrode for signal
 274 acquisition (Universal Probe, signal amplification 10x). The antenna preparation was

275 inserted into a glass duct connected to the EAD deactivated column tip, carrying a
276 humidified air stream at 0.3 mL/min. Two microliter aliquots of laboratory-collected
277 volatile extracts were injected in splitless mode at 250 °C, and the GC oven was set at
278 40 °C for 5 min and then increased to 250 °C at 10 °C/min. FID and EAD signals were
279 recorded by Syntech GcEad software (version 4.6).

280

281 **Chemical analysis**

282 The EAD active compound in the VOCs was identified by gas chromatography
283 coupled to a mass spectrometer (GC-MS) with a Shimadzu GCQP-2010 Ultra gas
284 chromatograph (Shimadzu Corp., Kyoto, Japan), equipped with a non-polar stationary
285 phase column (30 m × 25µm × 25 mm; Rxi-1MS; RESTEK, Bellefonte, PA, USA). One
286 microliter of the extract was injected at 250 °C in split mode (1:3). The GC oven
287 temperature was the same for GC-FID/EAD analysis (above). The helium carrier gas
288 was maintained at a flow of 1.3 mL/min in linear velocity at 41.1 cm/s and 70.7 kPa
289 pressure. Both ion source and quadrupole temperatures were set at 250 °C. Mass
290 spectra were recorded in electron impact mode (70 eV) from *m/z* 35-280. Compounds
291 were identified by their retention indexes, mass spectra comparison with Library NIST
292 11, and injection of authentic standards (Sigma-Aldrich). The target compound was
293 quantified by GC-FID with a Shimadzu GC-2010 gas chromatograph (Shimadzu Corp.,
294 Kyoto, Japan) equipped with a non-polar stationary phase column (30 m × 25µm × 25
295 mm; Rtx-1; RESTEK, Bellefonte, PA, USA) following the same set-up of GC-FID/EAD
296 analysis. We added 3 µL of a 1 mg/mL solution of octadecane as an internal standard
297 in all samples for quantification.

298 The absolute configuration of flower-produced nerolidol was determined using
299 an Agilent 6890GC with flame ionization detector equipped with a permethylated β-
300 cyclodextrin stationary phase chiral column (30 m × 25µm × 25 mm; HP-CHIRAL-20B;
301 J&W Scientific, Folsom, CA, USA). Synthetic nerolidol (a mixture of isomers) and

individual geometric isomers were purchased from Sigma-Aldrich to obtain retention time of *cis* and *trans*-nerolidol. A sample of synthetic (3*R*,6*E*)-nerolidol was kindly donated by Dr. Robert Hanus (Institute of Organic Chemistry and Biochemistry Academy of Sciences of the Czech Republic). All compounds solution in hexane (40 ppm) were injected and detected at 250 °C. Initial oven temperature was 85 °C for 1 min and then increased to 190 °C at 2 °C/min. For both chiral identification and field assays, we used the (*E*)-nerolidol from Sigma-Aldrich, which was previously determined to be comprised of almost 100% (3*S*,6*E*)-nerolidol [21], and also confirmed in our GC analysis (Fig 3d).

311

312 **Field bioassays of synthetic flower volatiles**

313 We conducted three independent field trials. In the first tests, we mimicked the 314 *L. siceraria* flowers by using flower-shaped dummies made from white filter paper, 315 which were impregnated with 100 mg of neat nerolidol (a mixture of isomers, Sigma- 316 Aldrich). Control dummies had no compound. Dummies were attached to wood sticks 317 (25 cm long), which were inserted into the ground, so the dummies were at 23 cm 318 high. Nerolidol-treated and control dummies were randomly distributed in three pairs, 319 with each pair containing one dummy of one treatment. Dummies were spaced 3 m 320 apart, and each pair of dummies was placed ~ 5 m from the other. Bottle gourd flowers 321 dummies were checked for visiting beetles from 06:00 p.m. to 09:00 p.m. Soon after 322 landing, Cyclocephalini beetles were removed from dummies to avoid potential 323 intraspecific attraction. This bioassay was carried out in the same *L. siceraria* grove in 324 Piracicaba, SP, Brazil (see above) for 2 d (March 12-13, 2018), with dummies 325 replacement made for each replicate.

326 The second field tests were conducted in the same area in Piracicaba and a 327 pasture area owned by the Mato Grosso do Sul State University, Cassilândia, MS, 328 Brazil (~ 800 km from Piracicaba; 19°05'35.8" S, 51°48'52.5" W). We used custom-

made, cross-vane intercept traps (black polystyrene panels: 18 × 24 cm), with 0.5-L plastic jars attached to the bottom of the trap. Both control and treatment traps were loaded with 200 ml of a solution of water and dish detergent to kill the captured Cyclocephalini beetles. Traps were hung from L-shaped PVC pipe frames (1.5 m long) that were supported by 1-m reinforcing steel bars hammered into the ground. The lures were pieces of filter paper (2 × 6 cm) loaded with 50 mg of neat nerolidol (a mixture of isomers). Control lures were filter papers with no compound. Lures were hung in the central opening slot of traps with a diaper pin 1-h prior to the sunset. Treated and control traps were randomly distributed. Seven even pairs of traps were deployed in Piracicaba and 10 pairs in Cassilândia. Traps were spaced 20 m apart, and pairs of traps were set ~30 m from each other. Traps were serviced for captured beetles in the morning. For every replicate date, the lures were replaced, and the treatment traps were switched in position to avoid positional effects. Overall, the bioassay was repeated for 5 d in Piracicaba (October 17-22, 2018) and for 1 d in Cassilândia (October 1st, 2018).

The third bioassay was conducted in the same area in Cassilândia (October 1-2, 2019), which tested the following treatments: (i) 5 mg of neat (3*S*,6*E*)-nerolidol (*trans*-nerolidol, Sigma-Aldrich); (ii) 20 mg of neat nerolidol (a mixture of isomers), considering 5 mg per isomer; and (iii) control (filter paper with no compound). Treatments were assigned randomly to traps within five blocks. Evaluations were carried out every morning for two days. Lures were replaced 1-h before the sunset, and treatment traps were changed one position within blocks to control for positional effects.

Captured cyclocephalini specimens were sent to Dr. Paschoal Coelho Grossi (Federal Rural University of Pernambuco - UFRPE) for taxonomy. A voucher specimen of Cyclocephalini beetle, *C. paraguayensis*, was deposited in the collection of the Museum of Entomology (#6945), Department of Entomology and Acarology (University of São Paulo, Piracicaba, Brazil). Collection of beetles in the field were conducted

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358Environment. This work was submitted to the Genetic Heritage Management Council
359by SisGen within project registration #AFC4FA2.

360

361 **Statistical Analysis**

362 Data from experiments 1 and 2 were analyzed with the two-tailed Exact
363Binomial test to verify whether the ratio of beetles attracted to nerolidol and control
364differed significantly from the expected 1:1 ratio at 5% probability [22]. The test was
365performed from a spreadsheet provided by
366<http://www.biostathandbook.com/exactgof.html> (accessed January 17, 2019).

367 Differences between treatment means from experiment 3 were analyzed using
368the nonparametric Friedman's Test (PROC FREQ, option CMH; SAS Institute, 2011),
369because data did not pass the assumptions of ANOVA [23]. Replicates were defined by
370block and collection dates. Replicates with no capture of *C. paraguayensis* in any trap
371were excluded from analyses. Once an overall significance was detected by the
372Friedman's test, pairs of means were compared using the REGWQ multiple range test,
373which controls for type I experiment-wise error rate [24]. For all bioassays, the sex ratio
374of attracted adults of *C. paraguayensis* was compared to a nominal proportion of 0.5
375with 95% Clopper-Pearson Exact Confidence Intervals [25].

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386

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- 460

Figure 1

Figure 2

Z-nerolidol

E-nerolidol

Figure 3

A **B**

Figure 4