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Abstract	*Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae), known as Medfly, is a severe agricultural invasive pest in Argentinian fruit-producing regions. The native habitat disturbance and introduction and spread of exotic host plants strongly favored Medfly proliferation. This scenario is common throughout the northern subtropical citrus-growing region. Environmentally friendly strategies to suppress Medfly populations by the National Fruit Fly Control and Eradication Program have currently been taken. One of these actions involves augmentative biological control through releases of the exotic parasitoid *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae). Consequently, the hypothesis that the effectiveness of *D. longicaudata* females, from two different population lines, in controlling Medfly larvae progressively increases as the density of released females increases was tested. One parasitoid line derives from larvae of a biparental Medfly strain. The other hails from irradiated larvae of the genetic sexing Temperature Sensitive Lethal Vienna-8 medfly strain reared at the "BioPlanta San Juan" biofactory. Parasitoids foraged for 24 h on peaches artificially inoculated with naked lab-reared biparental Medfly larvae. Peaches were placed near the roof or on the ground in field cages. Five treatments (20, 40, 80, 160, and 320 females released) and a control (no parasitoids) for each population line were carried out throughout summer and autumn 2016. Host density (200 larvae) remained constant. At 320 released parasitoid females, both *D. longicaudata* population lines highly increased the Medfly mortality in both testing seasons, and foraged skillfully on peaches at both fruit height levels. These data encourage the application of augmentative biological control against Medfly.

Keywords (separated by '-')	Fruit fly biological control - Parasitoid effectiveness - Parasitoid foraging ability - Mediterranean fruit fly - South America
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BIOLOGICAL CONTROL IN LATIN AMERICA

Augmentative Releases of Two *Diachasmimorpha longicaudata* (Hymenoptera: Braconidae) Population Lines Under Field-Cage Conditions to Control *Ceratitis capitata* (Diptera: Tephritidae)

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Abstract

Ceratitis capitata (Wiedemann) (Diptera: Tephritidae), known as Medfly, is a severe agricultural invasive pest in Argentinian fruit-producing regions. The native habitat disturbance and introduction and spread of exotic host plants strongly favored Medfly proliferation. This scenario is common throughout the northern subtropical citrus-growing region. Environmentally friendly strategies to suppress Medfly populations by the National Fruit Fly Control and Eradication Program have currently been taken. One of these actions involves augmentative biological control through releases of the exotic parasitoid *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae). Consequently, the hypothesis that the effectiveness of *D. longicaudata* females, from two different population lines, in controlling Medfly larvae progressively increases as the density of released females increases was tested. One parasitoid line derives from larvae of a biparental Medfly strain. The other hails from irradiated larvae of the genetic sexing Temperature Sensitive Lethal Vienna-8 medfly strain reared at the "BioPlanta San Juan" biofactory. Parasitoids foraged for 24 h on peaches artificially inoculated with naked lab-reared biparental Medfly larvae. Peaches were placed near the roof or on the ground in field cages. Five treatments (20, 40, 80, 160, and 320 females released) and a control (no parasitoids) for each population line were carried out throughout summer and autumn 2016. Host density (200 larvae) remained constant. At 320 released parasitoid females, both *D. longicaudata* population lines highly increased the Medfly mortality in both testing seasons, and foraged skillfully on peaches at both fruit height levels. These data encourage the application of augmentative biological control against Medfly.

Keywords Fruit fly biological control · Parasitoid effectiveness · Parasitoid foraging ability · Mediterranean fruit fly · South America

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Introduction

Habitat degradation in Latin America could be linked with persistence and population growth of invasive species such as *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae), commonly known as the Mediterranean fruit fly or Medfly. This is mainly because extensive areas of native vegetation was replaced with agricultural crops, which led to a mosaic landscape in which natural habitats with different disturbance degrees persist between crops and inhabited rural areas where mostly orchards with exotic fruit species are found (Duyck et al. 2006; Aluja et al. 2012; Schliserman et al. 2014). This environmental scenario is largely common throughout both northwestern and northeastern subtropical regions of Argentina. Several Medfly exotic host fruit species, such as *Citrus aurantium* L. (sour orange), *C. sinensis*

L. (Osbeck) (sweet orange), *C. paradisi* Macfadyen (grapefruit), *C. reticulata* Blanco (mandarin orange) (Rutaceae), *Prunus persica* (L.) Batsch (peach), *P. domestica* L. (plum), *P. armeniaca* L. (apricot), *Eriobotrya japonica* (Thunb.) Lindl. (loquat), *Rubus ideaus* L. (raspberry), *R. fruticosus* L. (blackberry) (Rosaceae), *Vaccinium corymbosum* L. (blueberry) (Ericaceae), *Diospyros kaki* L. (Japanese persimmon) (Ebenaceae), *Ficus carica* L. (fig) (Moraceae), *Mangifera indica* L. (mango) (Anacardiaceae), and *Psidium guajava* L. (common guava) (Myrtaceae), are cultivated commercially in large areas or in backyard orchards for local consumption, although they are also feral fruits that grow in patches of disturbed natural vegetation (Ovruski et al. 2003; Schliserman et al. 2014; Funes et al. 2017). Consecutively, in those Medfly host fruits, the natural control by guilds of native parasitoids is scarce or null (Ovruski et al. 2004; Schliserman et al. 2016). Thus, all those alternative exotic feral fruit species throughout Argentinean northern region make Medfly proliferation feasible, but concurrently make problematic their control (Schliserman et al. 2016).

From an economic perspective, *C. capitata* is one of the most severe agricultural invasive pests in all Argentinian fruit-producing regions, and together with the native *Anastrepha fraterculus* (Wiedemann) or South American fruit fly strongly affects Argentine fruit production and growth, marketing, and export (SENASA 2017). Farmers from northern Argentina use different techniques to control both fruit fly pest species. This is subject to their level of education and the economic power for purchasing farming inputs such as synthetic pesticides, which can be used indiscriminately (Ovruski and Schliserman 2012). Against this background in Argentina, the National Agri-Food and Animal Health and Quality Service (SENASA, Spanish acronym) has implemented the National Fruit Fly Control and Eradication Program (PROCEM, Spanish acronym). This achievement has made the struggle against *C. capitata* one of the foremost priorities in the country, but through more effective, friendly to the environment and human health, integrated control strategies (SENASA 2017). In consequence, the Provincial Fruit Fly Control and Eradication Program of San Juan (ProCEM-San Juan, Spanish acronym) incorporated biological control using hymenopteran parasitoids in 2008 for the control of Medfly populations at the fruit-growing region of Cuyo, central-western Argentina (Suárez et al. 2019a). Regarding this fact, open-field augmentative releases of the Southeast Asian-native parasitoid *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae), produced at "BioPlanta San Juan" mass-rearing facility, have been successfully performed in a Medfly-infested fig crop located in southeastern San Juan province to assess capacity of this exotic parasitoid species to control the target pest (Sánchez et al. 2016). The opiine braconid *D. longicaudata* mass-reared at the "BioPlanta San Juan" is a solitary larval-pupal fruit fly endoparasitoid, commonly used worldwide as a biocontrol agent in augmentative releasing programs (Ovruski et al. 2000; Montoya et al. 2017; de Pedro et al. 2019; Garcia et al. 2020).

Based on the growing progress in the *D. longicaudata* mass production (Suárez et al. 2020; Carta Gadea et al. 2020) and the successful open-field pilot release tests (Suárez et al. 2014; Sánchez et al. 2016), Medfly biological control is currently considered a workable complementary control strategy for other Argentinian fruit-growing regions (Núñez-Campero et al. 2020). Therefore, an integrated approach to fruit fly pest management involving biological control throughout the northern Argentinian Citrus-producing areas is currently developed by the national PROCEM (SENASA 2017). Taking into account the aforementioned circumstance, the aim of this study was to assess the augmentative biological control strategy against *C. capitata*, albeit in a simulated way, under field-cage conditions in the subtropical northwestern Argentina, by using *D. longicaudata* from two different population lines: one from "BioPlanta San Juan" biofactory and another one originated in Tucumán. Consequently, the hypothesis that females of *D. longicaudata*, regardless of the population line, are progressively more effective in finding and attacking Medfly larvae infesting peaches as the density of released females in the tested patch increases was tested. This prediction has been based on previous studies (Ovruski et al. 2012; Suárez et al. 2019b) on the performance of *D. longicaudata* to develop on *C. capitata* and for their foraging ability on diverse Medfly host fruit species in northwestern Argentina.

Material and methods

Source of insects and rearing procedures

Parasitoids from a *D. longicaudata* population line reared on third-instar larvae of a biparental *C. capitata* strain (from now on: Dl_{BipCc}) and Medfly adults were produced at the Biological Control Section (BCS) of the Planta Piloto de Procesos Industriales Microbiológicos y Biotecnología (PROIMI) located in San Miguel de Tucumán, northwestern Argentina. Both parasitoid and biparental *C. capitata* adults were held in cubical acrylic glass-structured, voile-covered cages (30 × 30 × 30 cm). The cages were kept in an air-conditioned room at 25 ± 1°C, 75 ± 5% relative humidity (RH), and 12-h photoperiod. The biparental *C. capitata* colony started from wild individuals recovered by harvesting infested feral peaches from trees in a protected wildlife area in Yerba Buena, Tucumán (26°55′S, 65°05′W, 600–800 m). The Dl_{wildCc} colony was originally established from individuals imported from the Mexican Moscafrut Program, where they were reared on irradiated *Anastrepha ludens* (Loew)

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larvae. Parasitoids were provided ad libitum with water and honey every other day. Medfly adults were fed daily ad libitum with a mixture of yeast hydrolysate enzymatic (MP Biomedicals, LLC, Solon, OH) and common refined sugar (Ledesma®, Buenos Aires, Argentina) plus water. Fly larvae were reared on artificial diet based on wheat germ, sugar, brewer's yeast, citric acid, agar–agar, vitamins, minerals, and preserving agents. A second *D. longicaudata* population line was reared on 90 Gy-irradiated third-instar larvae of the genetic sexing Temperature Sensitive Lethal (*tsl*) Vienna-8 *C. capitata* (from now on: Dl_{tslCc}) at the Parasitoid Rearing Laboratory of the "BioPlanta San Juan" mass-rearing facility belonging to the San Juan Biotechnology Center of the Dirección de Sanidad Vegetal, Animal y Alimentos (DSVAA), San Juan, Argentina. The Dl_{tslCc} colony was held in rectangular iron-framed, voile-covered cages (50 × 50 × 60 cm). The Dl_{tslCc} colony was initially established from 200 individuals of the Dl_{BipCc} reared at the BCS-PROIMI's laboratory (Suárez et al. 2019a). Adults parasitoids were provided with honey and water ad libitum and kept at 24 ± 1°C; 65 ± 5% RH and 12 h photoperiod. The colony of *tsl* Vienna-8 *C. capitata* strain was reared at the Medfly Rearing Laboratory of the "BioPlanta San Juan" mass-rearing facility. Larvae of this Medfly strain were reared on an artificial diet based on wheat bran, sugar, yeast, poplar wood chips, hydrochloric acid, sodium benzoate, methyl p-hydroxybenzoate, and water (Suárez et al. 2019a). Puparia parasitized by Dl_{TSLCc} population line were sent from San Juan Biotechnology Center to PROIMI every 5 days between January and April 2016 to carry out the tests in Tucumán. Adult parasitoids of both Dl_{tslCc} and Dl_{BipCc} intended for testing were kept under controlled lab conditions (25 ± 1°C, 75 ± 5% RH, and 12:12 L:D h) in a 6-m^2 room at the BCS-PROIMI's laboratory, and provided with water and honey every other day. Dl_{tslCc} and Dl_{BipCc} cohorts used in the trials were the 145th and 60th generations, respectively.

Experimental site and environmental conditions

The assay series were performed inside field cages under uncontrolled environmental conditions at an experimental citrus crop at the Instituto Nacional de Tecnología Agropecuaria–Estación Experimental Agropecuaria Famaillá (INTA-EEA Famaillá) (27°03′S, 65°25′W, 363 m), in Famaillá, Tucumán province. The climate is humid warm temperate, with a rainy-warm season from October to April, and a dry-cold season from May to September. The average annual temperature is 19.4°C and the average annual rainfall is close to 1000 mm. Percentage of RH and mean temperature recorded during trial dates were provided by digital weather stations (LUFT®, model WS80, China). For each field cage, a weather station was placed inside, while outside of them another weather station protected from the sun and rain was situated. Both the RH and mean temperature inside and outside field cages were recorded every 30 min for 24 h, the time that each replicate lasted.

Experimental setup

Tests were performed inside four cylindrical nylon field cages (3.5 × 3.0 m, diameter × height) placed at the INTA-EEA Famaillá's experimental citrus crop during two seasons in 2016, summer (January–February) and autumn (April–May). Field cages were sheltered from both the sun by a 130-g/m^2 black-shade cloth, UV resistant net (AGROREDES®, San Isidro, Buenos Aires, Argentina), and the rain by a 100-µ translucent high density polyethylene protective cover (AGROREDES®). Both cloth and plastic cover were placed 50 cm above the cage roof, which allowed sunlight to pass through but not rainwater. Inside every field cage were placed six small cylindrical cages (0.5 × 2.0 m, diameter and height) made of white cotton voile fabric. These internal experimental cages (from now on: EC) were reinforced with two thick wire rings, one located at the base and another at the top. Each EC also had a 1.5-m-long zipper in its middle part to enter. A potted peach tree (~1.5 m high) was placed inside each EC to simulate a natural environment. Four peaches, *Prunus persica* (L.) Batsch (Flavor Crest cultivar), were placed within each EC equidistant from each other; in this way, they formed a circle in which each fruit was located towards a different cardinal point. Two field cages contained four fruits in their upper section, next to the roof, while in the other two field cages four peaches were located in the cage floor. Overall, 16 similar-size (50.6 ± 1.9 g and 69.1 ± 2.3 cm^3 weight × volume) and ripeness degree (ripe, reddish-yellowish) peaches were selected for use in the trials per testing date. The fruit was bought in a supermarket, washed first with a sodium benzoate plus methyl p-hydroxybenzoate solution and then only with water. After this, peaches were dried and individually inoculated with 50 5-day-old lab-reared *C. capitata* larvae from the BCS-PROIMI's laboratory. Artificial infestation was carried out by cutting the fruit at its top, equivalent to one-fourth of the total fruit, by using a sterilized scalpel and the half of pulp plus the stone was removed; then the space formed inside the fruit was filled with naked (i.e., without artificial larval diet) lab-reared *C. capitata* larvae from the biparental strain. About 50% of the total fruit volume was engaged by *C. capitata* larvae. Once the fruit was inoculated, both fruit sections, small upper cover and large lower piece, were joined together with 2.5-cm-wide Parafilm "M"® strips (Pechiney Plastic Packaging, Chicago, USA). Each artificially infested fruit represented one oviposition unit. Once inoculated fruits were placed inside the EC, in those field cages whose treatments involved fruits in the upper

part, inoculated peaches were hung from the center of the EC roof and positioned 1.5 m above EC floor at the canopy level of the potted tree. Each peach was hung from the EC roof by means of a strong nylon fishing rope tied to a metal clip, which in turn was inserted into the thin plastic strip that wrapped the fruit. A circular galvanized wire-framed, voile-covered basket (10 × 2 cm, diameter × height) filled with sterilized 2-mm-thick vermiculite Intersum® (Aislater S.R.L., Córdoba, Argentina) was positioned about 5 cm below the peach. This basket was held by four equidistant nylon fishing ropes and tied to the main string from which the peach was hung. In those field cages whose treatments involved fruits on the bottom, inoculated peaches were placed onto a plastic tripod stand (3 × 2 cm, diameter × height) positioned in a circle around the potted tree inside a square white plastic tray (34 × 34 × 19 cm) located on the EC floor. Each peach was oriented towards a certain cardinal point. The plastic tray had 1 cm of sterilized vermiculite on the bottom as a pupation substrate. Each field cage concurrently included five treatments and one control carried out in individual ECs. Naïve, i.e., non-exposed to host larvae or fruit or both, 7-day-old, mated females from Dl_{BipCc} or Dl_{tslCc} population line were used in tests. Treatments in cage 1 involved different release densities of Dl_{BipCc} females inside ECs with peaches on the top. Treatments were as follows: T_1, 20 Dl_{BipCc} females released; T_2, 40 females released; T_3, 80 females released; T_4, 160 females released; T_5, 320 females released; and a control, no parasitoids were released. Controls were conducted to determine natural *C. capitata* adult emergence rates for which involved inoculated peach but not exposed to parasitoids. Treatments in cage 2 involved the same parasitoid female release densities but from the Dl_{tslCc} population line inside ECs with peaches on the top. Treatments in cages 3 and 4 involved the same parasitoid female release densities from the Dl_{BipCc} and the Dl_{tslCc}, respectively, inside ECs but with peaches on the bottom. The host-parasitoid ratio per treatment was 10, 5, 2.5, 1.25, and 0.63 *C. capitata* larvae per 1 *D. longicaudata* female either Dl_{BipCc} or Dl_{tslCc}. The host density remained constant in all treatments, 200 *C. capitata* larvae per EC. Parasitoid females were released into each EC on leaves of the potted tree's median portion, and they were allowed to forage fruit for 24-h period starting at 12:00 h. After tests were finished, both peaches and the vermiculite from the basket as well as both fruits and the vermiculite from the plastic tray were removed from each EC. In the laboratory peaches, either top or bottom were dissected to remove possible living *C. capitata* larvae, and host puparia were recovered from the basket or from the tray. Larvae recovered from the fruit located on the top and puparia recovered from the basket belonging to a particular treatment were placed into the same plastic cups (10 × 7 cm, diameter × height) with sterilized vermiculite on the bottom as a pupation substrate. Larvae recovered from the fruit located on the bottom and puparia recovered from the tray belonging to a particular treatment were jointly deposited in plastic cups as described above. The top of each cup was tightly covered with a voile piece. The cups were kept in a room air-conditioned at 26 ± 1°C, 80 ± 5% RH, and 10 h photoperiod until adult flies and parasitoids emerged. Both the number and sex of the parasitoids and the number of adult flies were recorded. Treatments and controls were replicated 10 times. For each replicate, new inoculated fruits, new pupation substrate, and new parasitoid females were always used. Tests were performed to comparatively assess the effectiveness of both *D. longicaudata* population lines, Dl_{BipCc} or Dl_{tslCc}, in parasitizing *C. capitata* larvae in a key host fruit species under a natural free-foraging condition and into a variation in parasitoid female release densities. In turn, the augmentative biological control tactic was also evaluated. Furthermore, the effect of infested fruit location, i.e., fruit located in the canopy and fruit fallen on the ground, on effectiveness of both parasitoid population lines was also taken into consideration.

Data analysis

Four biological parameters were considered: (1) percentage of parasitoid emergence was calculated as the number of emerged parasitoids divided by total exposed hosts per 100; (2) percentage of parasitism was calculated as the number of parasitoids emerged plus the number of non-emerged parasitoids (larvae, pupae, or adults) found inside puparia divided by total exposed hosts per 100; (3) percentage of effectiveness, that is the total host mortality inflicted by the parasitoid under natural environmental conditions, which was estimated through Abbot's corrected formula (Rosenheim and Hoy 1989); this formula relates the emerged Medfly population recovered from treatments, in which parasitoid releases were made, with the living host population recorded from the control test; and (4) sex ratio, as the percentage of emerged female parasitoids over the total number of emerged parasitoids per 100. The four parameters were estimated for each parasitoid population line and per testing date. Firstly, univariate two-factor general lineal models (GLMs) with type III error at $p = 0.05$ were performed to identify significant effects of the parasitoid emergence, parasitism, effectiveness, and sex ratio on two interacting fixed factors of the models, namely, parasitoid populations lines (Dl_{BipCc} or Dl_{tslCc}) and female parasitoid release densities with five levels (20, 40, 80, 160, and 320 parasitoid females). Models were distinctly used for the infested fruit localization (canopy or ground) and for both seasons (summer and autumn). Secondly, the effectiveness was analyzed using univariate two-factor GLMs to compare it through

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the interaction of two fixed factors, namely, parasitoid population lines and fruit localization. These models were particularly applied for each treatment (female parasitoid release densities) and for both seasons assessed in the study. Thirdly, the effectiveness was analyzed by means of univariate two-factor GLMs to compare it through the interaction of parasitoid population lines and testing seasons. Models were run for each treatment and for both fruit located at canopy and ground levels. Furthermore, mean temperatures and RH percentages recorded inside each of the four field cages and outside of them were analyzed using univariate one-factor GLMs by testing date and per studying season. A Pearson product moment correlation at $p = 0.05$ was applied to determine the degree of association between RH and temperature during testing dates in both seasons, and to be able to choose if one or both environmental variables can be used as covariate in the statistical analyses on the parasitoid performance. In addition, *t*-student tests at $p = 0.05$ were particularly used to compare both mean temperatures and RH percentages between both seasons. Mean partitioning was performed by Tukey's honestly significant difference (HSD) test at $p = 0.05$. Given lack of normality, data were rank transformed prior to analyses (Conover and Iman 1981), but untransformed means (± SE) were used in all tables and figures. Statistical analyses were performed using STATISTICA software, version 10.0 (StatSoft 2011).

Results

Environmental conditions inside and outside field cages

Both RH and temperature values did not differ significantly between field cages and outdoors by study date, taking into account both summer and autumn (raw and analyzed data are included as an appendix). There was a positive and significant correlation between mean temperature and RH ($r = 0.60$, $N = 4{,}800$, $p > 0.001$). Therefore, the mean temperature outside field cages was only included as a covariate to assess a potential influence of environmental conditions on performance of both parasitoid population lines. There were significant differences between mean temperature and RH throughout the studied periods (temperature and RH in summer vs autumn; 28.5 ± 0.2°C vs 23.7 ± 0.3°C, $t = 280.747$, $df = 79$, $p > 0.001$; 73.1 ± 0.4% vs 57.1 ± 1.0%, $t = 39.444$, $df = 79$, $p > 0.001$).

Assessment of summer parasitoid releases

Results from the field cages in which peaches were located in the upper portion of each EC, which can be associated with the canopy of the potted tree, are provided first. Female parasitoid release densities (from now on: FPRD) and their interaction with parasitoid population lines (from now on: PPL) had a significant effect on both parasitoid emergence and parasitism (Table 1). Both FPRD and PPL, and their

Table 1 Summary of univariate two-factor GLMs with a covariable (outdoor mean temperature) on the effect of female parasitoid release densities, parasitoid population lines, and their interactions on *Diachasmimorpha longicaudata* adult emergence, parasitism, effectiveness, and female offspring proportion (sex ratio) recorded from third instars of a biparental *Ceratitis capitata* strain inside peaches located at the canopy of potted tree and on the ground under field-cage conditions in summer (January–February 2016); Tucumán, Argentina

Source of variation/fruit location	Biological parameters (dependent variables)									
	df	Error *df*	Parasitoid emergence		Parasitism		Effectiveness		Sex ratio	
			F	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Fruit at the canopy										
Categorical variables:										
Parasitoid release densities (FPRD)	4	389	190.532	< 0.001*	353.404	< 0.001*	418.378	< 0.001*	2.770	= 0.027*
Parasitoid population lines (PPL)	1	389	0.929	= 0.335	2.996	= 0.084	4.578	= 0.033*	1.152	= 0.284
FPRD × PPL	4	389	4.543	= 0.001*	2.725	= 0.029*	4.053	= 0.003*	0.284	= 0.888
Covariable: temperature (outdoor)	1	389	0.044	= 0.507	0.044	= 0.834	0.087	= 0.768	0.008	= 0.976
Fruit on the ground										
Categorical variables:										
Parasitoid release densities (FPRD)	4	389	227.124	< 0.001*	364.633	< 0.001*	424.039	< 0.001*	1.868	= 0.115
Parasitoid population lines (PPL)	1	389	4.129	= 0.043*	2.611	= 0.107	3.654	= 0.057	0.002	= 0.968
FPRD × PPL	4	389	5.935	= 0.001*	4.098	= 0.003*	2.652	= 0.033*	0.721	= 0.578
Covariable: temperature (outdoor)	1	389	0.052	= 0.305	2.686	= 0.102	2.067	= 0.151	0.017	= 0.896

*Statistically significant

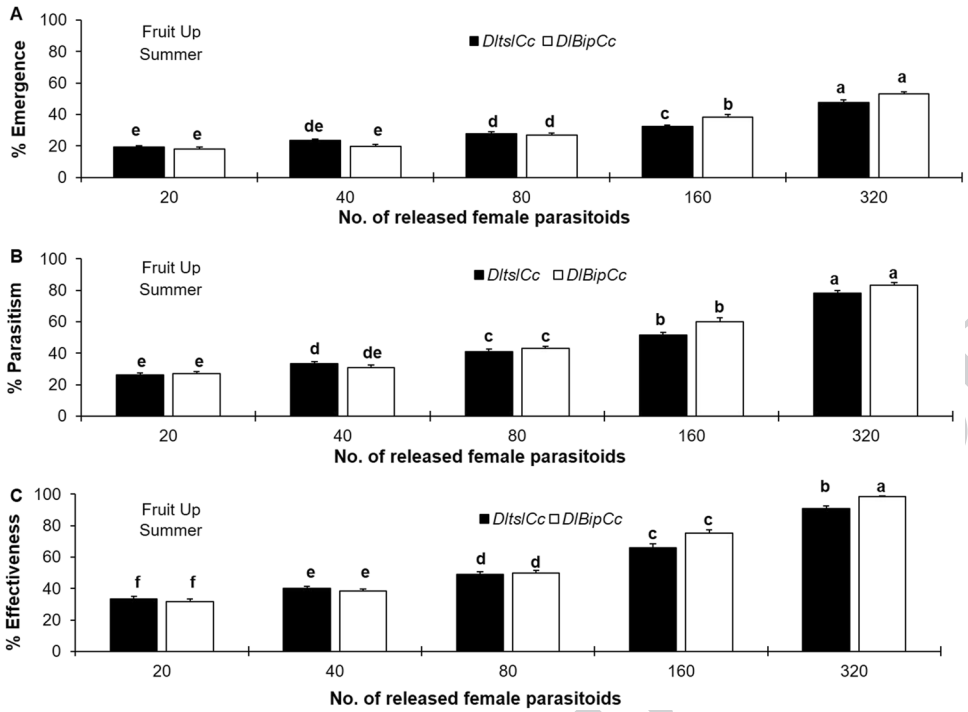

Fig. 1 Mean (±SE) percentage of individuals emergence (**A**), parasitism (**b**), and effectiveness (**C**) recorded from artificially inoculated peaches (*Prunus persica*) with 200 lab-reared third-instar *Ceratitis capitata* larvae from a biparental strain and located at the upper section, next to the roof of field cages, that were parasitized by different *Diachasmimorpha longicaudata* female release densities of two population lines throughout summer 2016; Tucumán, Argentina. Bars with dissimilar letter indicate significant differences (Tukey HSD test, $p = 0.05$). Notations: Dl_{tslCc}, *D. longicaudata* lineage reared on irradiated larvae of the Temperature Sensitive Lethal Vienna-8 *C. capitata* strain; Dl_{BipCc}, *D. longicaudata* lineage reared on non-irradiated larvae of a biparental *C. capitata* strain

interaction also, had a substantial influence on effectiveness (Table 1). Only FPRD had a considerable effect on offspring sex ratio, but the interaction between both categorical factors did not (Table 1). Adult emergence from both Dl_{BipCc} and Dl_{tslCc} significantly increased concurrently with FPRD (Fig. 1(A)). The same pattern described above was recorded for parasitism (Fig. 1(B)). Similarly, the effectiveness increased notably, but gradually, with FPRD growth, although the maximum significant effectiveness was recorded in the Dl_{BipCc} population line (Fig. (C)). Both Dl_{BipCc} and Dl_{tslCc} exhibited a slightly female-biased sex ratio (1.2:1 and 1.1:1 females:male, respectively). When sex ratios among different FPRD were compared, regardless of the PPL and their interaction, there was a slight trend towards a greater female offspring emergence at low FPRD (20, 40, and 80 females) than at high FPRD (160 and 320 females), which was 55.9% vs 51.2% daughters on total offspring, respectively. Regarding results from the field cages in which fruits were placed on the ground of each EC, both the FPRD and the PPL, and their interaction, had a significant influence on parasitoid emergence (Table 1). The FPRD and their interaction with PPL had a considerable effect on both parasitism and effectiveness (Table 1). There was no significant effect on sex ratio by the two categorical factors or their interaction (Table 1). Parasitoid emergence displayed a gradual increase with a greater number of released female parasitoids; Dl_{BipCc} significantly peaked at 160 and 320 released females, while Dl_{tslCc} did at 320 released females (Fig. 2(A)). A similar trend was recorded for parasitism, with substantially higher values for both PPLs at the maximum parasitoid release density (Fig. 2(B)). The same pattern of increase described above was recorded for the effectiveness, which was notably higher in both Dl_{BipCc} and Dl_{tslCc} at 320 released females, although the effectiveness achieved by Dl_{BipCc} at 160 released females was particularly similar to that of 320 females (Fig. 2(C)). Both Dl_{BipCc} and Dl_{tslCc} exhibited a similar and slightly female biased sex ratio (1.2–1 female:male) in fruit in the canopy like fruit on the ground. The covariable (outdoor mean temperature) did not have a significant effect on outcomes of all statistical analyzes. In all treatments, the *D. longicaudata* females' effectiveness was significantly influenced by fruit location (FL), with varied outcomes (Table 2). There was a very slight trend to attack infested peaches in the top in treatments 2 and 5, while in the remaining treatments the trend was to attack fruit located on the ground (Fig. 3). However, the interaction between PPL and FL did not significantly affect the effectiveness of *D. longicaudata* females, except only in

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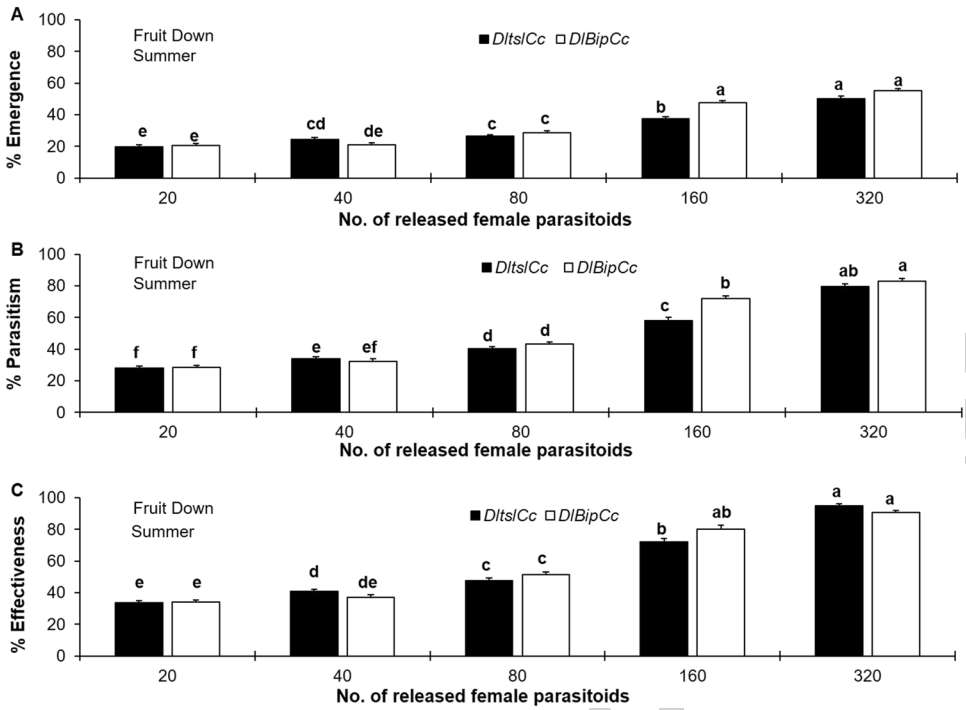

Fig. 2 Mean (±SE) percentage of individuals emergence (A), parasitism (b), and effectiveness (C) recorded from artificially inoculated peaches (*Prunus persica*) with 200 lab-reared third-instar *Ceratitis capitata* larvae from a biparental strain and located on the ground of field cages that were parasitized by different *Diachasmimorpha longicaudata* female release densities of two population lines throughout summer 2016; Tucumán, Argentina. Bars with dissimilar letter indicate significant differences (Tukey HSD test, $p = 0.05$). Notations: Dl_{tslCc}, *D. longicaudata* lineage reared on irradiated larvae of the Temperature Sensitive Lethal Vienna-8 *C. capitata* strain; Dl_{BipCc}, *D. longicaudata* lineage reared on non-irradiated larvae of a biparental *C. capitata* strain

treatment 4 (Table 2) in which Dl_{tslCc} females tended more to parasitize host larvae in peaches located on the ground, but the same did not happen with Dl_{BipCc} females.

Assessment of autumn parasitoid releases

In coincidence with summer results, both the FPRD and their interaction with PPL had a noteworthy influence on parasitoid emergence, parasitism, and effectiveness recorded from both peaches located in upper and lower sectors of ECs (Table 3). The interaction between both categorical factors had no influence on offspring sex ratio (Table 3). The emergence (Fig. 4(A)), parasitism (Fig. 4(B)), and the effectiveness (Fig. 4(C)) recorded for both Dl_{BipCc} and Dl_{tslCc} from peaches in the upper portion of each EC increased gradually concurrently with FPRD. A similar pattern of increase was also recorded from fruit in the lower portion of each EC for the emergence (Fig. 5(A)), parasitism (Fig. 5(B)), and the effectiveness (Fig. 5(C). However, the emergence was significantly analogous at both low parasitoid release values and high release densities (Fig. 5(A)) . The effectiveness of Dl_{BipCc} was substantially similar at higher release densities (160–320), although this did not occur with Dl_{tslCc} since its maximum value was recorded at 320 released parasitoids (Fig. 5(C)). Both Dl_{BipCc} and Dl_{tslCc} had a slightly female-biased sex ratio (1.3–1.2:1 and 1.3–1.1:1 females:male, respectively) in fruit in the canopy like fruit on the ground. In all treatments, the effectiveness was considerably affected by FL, but the interaction between PPL and FL did not significantly influence it (Table 4). In the first four release densities, *D. longicaudata* females were more effective on fallen fruit, but it was the reverse at 320 released parasitoids (Fig. 6). The comparison between the effectiveness of both Dl_{BipCc} and Dl_{tslCc} based on the testing seasons and their interaction with PPL did not show significant differences between summer and autumn (Table 5). In coincidence with summer results, the covariable did not have a significant effect on variable responses in overall used GLMs.

Discussion

Earlier research carried out in Tucumán under field cage during middle spring, November, and early December 2011, assessed the capability of the Dl_{BipCc} population line in parasitizing *C. capitata* larvae under a free-foraging

Table 2 Summary of univariate two-factor GLMs on the effect of parasitoid population lines and fruit location, and their interactions on *Diachasmimorpha longicaudata* effectiveness recorded per treatment from a third instar of biparental *Ceratitis capitata* strain inside peaches located at the canopy of potted tree and on the ground under field-cage conditions in summer (January–February 2016); Tucumán, Argentina

Source of variation (Categorical variables)	*df*	Error *df*	Treatments									
			T_1 (20 released parasitoids)		T_2 (40 released parasitoids)		T_3 (80 released parasitoids)		T_4 (160 released parasitoids)		T_5 (320 released parasitoids)	
			F	*P*	*F*	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Parasitoid population lines (PPL)	1	156	0.219	= 0.640	1.835	= 0.177	2.795	= 0.096	0.263	= 0.608	13.530	< 0.001*
Fruit location (FL)	1	156	35.505	< 0.001*	56.462	< 0.001*	137.764	< 0.001*	45.656	< 0.001*	2039.840	< 0.001*
PPL × FL	1	156	0.220	= 0.641	0.911	= 0.341	2.457	= 0.119	10.132	= 0.002*	2.570	= 0.111

*Statistically significant

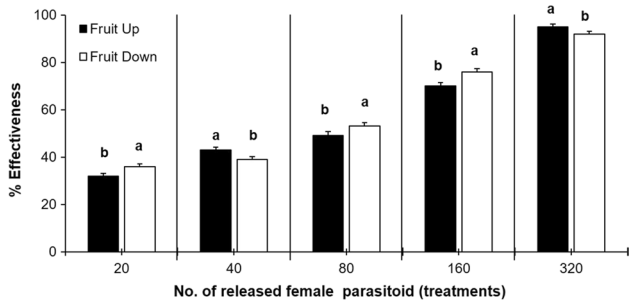

Fig. 3 Comparison between the mean (± SE) effectiveness of *Diachasmimorpha longicaudata* females released at five different densities to suppress medfly larvae infesting peaches located either in the upper (near to roof) or lower sector (on the ground) of field cages throughout summer 2016; Tucumán, Argentina. Data of both *D. longicaudata* lineages were pooled. Letters show comparisons between fruits up and down within the same parasitoid density. Bars with dissimilar letter indicate significant differences (Tukey HSD test, $p = 0.05$)

Citrus species choice condition (Ovruski et al. 2012). Subsequently, renewed studies carried out between late spring (November) and early summer (December) 2014 and 2015 showed that both lab-reared Dl_{BipCc} and Dl_{tslCc} population lines were comparably effective in attacking *C. capitata* larvae in host fruits with different physical and chemical features, such as peaches and oranges (Suárez et al. 2019b). Outcomes of the present study verified now that progressively increasing the number of released parasitoid females considerably favored the effectiveness of both Dl_{BipCc} and Dl_{tslCc} population lines to control *C. capitata* infesting peach, while host larvae density was kept constant. These outcomes recorded in field cages suggest that augmentative releases of that *D. longicaudata*'s lineage reared on *tsl* Vienna-8 Medfly larvae at the "BioPlanta San Juan" biofactory substantially decreased *C. capitata* adult emergence in an ecological region with subtropical climatic conditions. This finding is in agreement with open-field augmentative releases of the Dl_{tslCc} population line performed to assess their effectiveness on the regulation of wild Medfly populations at ecologically isolated, irrigated, fruit-growing areas of San Juan (Suárez et al. 2014; Sánchez et al. 2016). Interestingly, the effectiveness achieved by both *D. longicaudata* population lines in the present study was congruent at different densities of released parasitoids. When comparing the maximum evaluated released female parasitoid density (320 parasitoids per replicate) throughout both testing seasons, Dl_{tslCc} was significantly more or equally or less effective than Dl_{BipCc} to parasitize *C. capitata* larvae, but always its effectiveness was higher than 90%. This happened even though both mean emergence and parasitism percentages were particularly lower. Host mortality inflicted by *D. longicaudata* females due to other causes may adversely

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Table 3 Summary of univariate two-factor GLMs with a covariable (outdoor mean temperature) on the effect of female parasitoid release densities, parasitoid population lines, and their interactions on *Diachasmimorpha longicaudata* adult emergence, parasitism, effectiveness, and female offspring proportion (sex ratio) recorded from third instars of a biparental *Ceratitis capitata* strain inside peaches located at the canopy of potted tree and on the ground under field-cage conditions in autumn (April–May 2016); Tucumán, Argentina

Source of variation/fruit location	Biological parameters (dependent variables)									
	df	Error *df*	Parasitoid emergence		Parasitism		Effectiveness		Sex ratio	
			F	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Fruit at the canopy										
Categorical variables:										
Parasitoid release densities (FPRD)	4	389	231.409	< 0.001*	299.519	< 0.001*	436.957	< 0.001*	4.272	= 0.002*
Parasitoid population lines (PPL)	1	389	0.179	= 0.673	0.409	= 0.523	0.128	= 0.720	3.554	= 0.059
FPRD × PPL	4	389	2.616	= 0.035*	3.569	= 0.007*	5.099	< 0.001*	1.936	= 0.888
Covariable: temperature (outdoor)	1	389	0.184	= 0.668	1.257	= 0.263	3.115	= 0.078	0.001	= 0.971
Fruit on the ground										
Categorical variables:										
Parasitoid release densities (FPRD)	4	389	305.260	< 0.001*	430.899	< 0.001*	539.690	< 0.001*	0.379	= 0.823
Parasitoid population lines (PPL)	1	389	0.071	= 0.789	0.005	= 0.107	0.097	= 0.756	0.086	= 0.769
FPRD × PPL	4	389	3.684	= 0.006*	6.459	< 0.001*	3.835	= 0.004*	2.327	= 0.056
Covariable: temperature (outdoor)	1	389	2.606	= 0.107	3.066	= 0.081	0.958	= 0.328	0.012	= 0.913

*Statistically significant

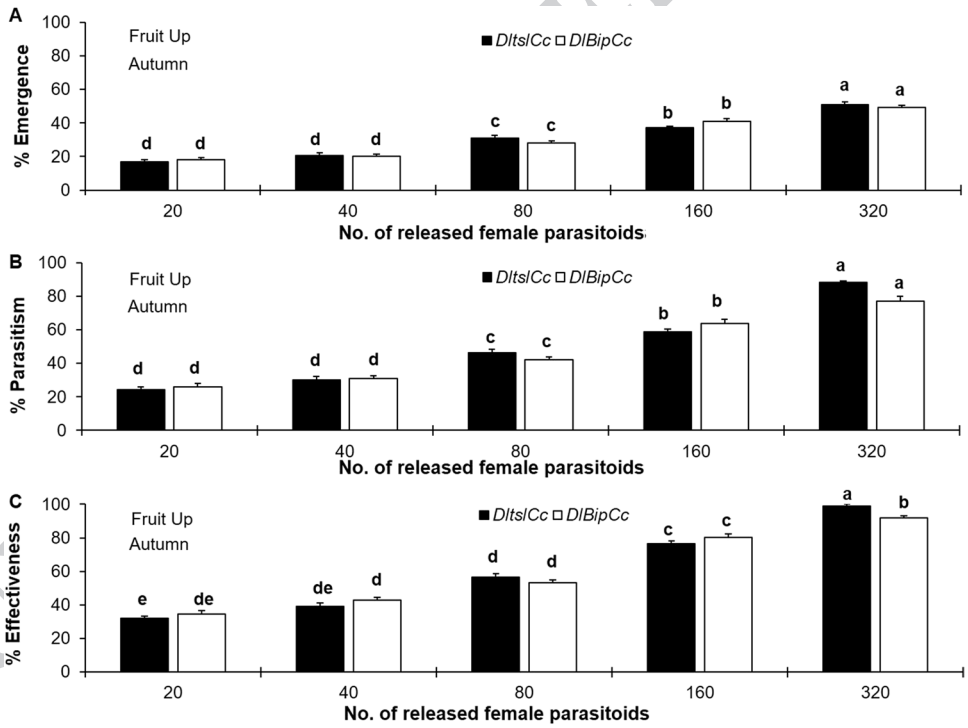

Fig. 4 Mean (±SE) percentage of individuals emergence (**A**), parasitism (b), and effectiveness (C) recorded from artificially inoculated peaches (*Prunus persica*) with 200 lab-reared third-instar *Ceratitis capitata* larvae from a biparental strain and located at the upper section, next to the roof of field cages, that were parasitized by different *Diachasmimorpha longicaudata* female release densities of two population lines throughout autumn 2016; Tucumán, Argentina. Bars with dissimilar letter indicate significant differences (Tukey HSD test, $p = 0.05$). Notations: Dl_{tslCc}, *D. longicaudata* lineage reared on irradiated larvae of the Temperature Sensitive Lethal Vienna-8 *C. capitata* strain; Dl_{BipCc}, *D. longicaudata* lineage reared on non-irradiated larvae of a biparental *C. capitata* strain

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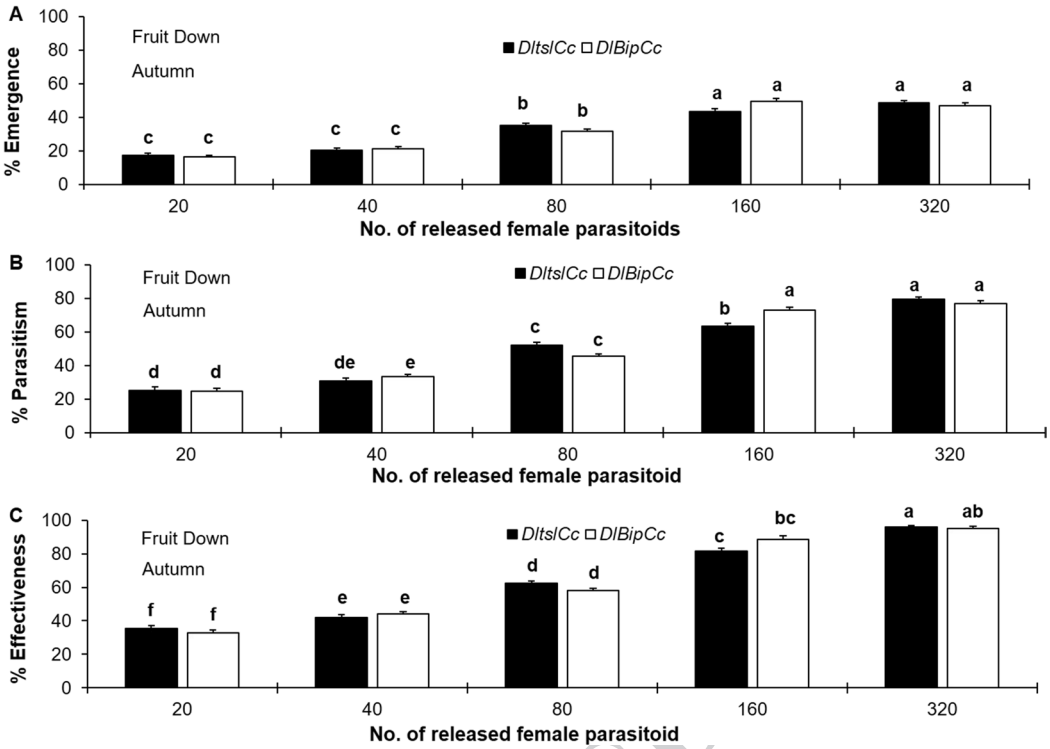

Fig. 5 Mean (±SE) percentage of individuals emergence (A), parasitism (b), and effectiveness (C) recorded from artificially inoculated peaches (*Prunus persica*) with 200 lab-reared third-instar *Ceratitis capitata* larvae from a biparental strain and located on the ground of field-cages, that were parasitized by different *Diachasmimorpha longicaudata* female release densities of two population lines throughout autumn 2016; Tucumán, Argentina. Bars with dissimilar letter indicate significant differences (Tukey HSD test, $p=0.05$). Notations: Dl_{tslCc}, *D. longicaudata* lineage reared on irradiated larvae of the Temperature Sensitive Lethal Vienna-8 *C. capitata* strain; Dl_{BipCc}, *D. longicaudata* lineage reared on non-irradiated larvae of a biparental *C. capitata* strain

Table 4 Summary of univariate two-factor GLMs on the effect of parasitoid population lines and fruit location, and their interactions on *Diachasmimorpha longicaudata* effectiveness recorded per treatment from a third instar of biparental *Ceratitis capitata* strain inside peaches located at the canopy of potted tree and on the ground under field-cage conditions in autumn (April–May 2016); Tucumán, Argentina

Treatments											
Source of variation (Categorical variables)	*df*	Error *df*	T_1 (20 released parasitoids)		T_2 (40 released parasitoids)		T_3 (80 released parasitoids)		T_4 (160 released parasitoids)		T_5 (320 released parasitoids)	
			F	*P*	*F*	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Parasitoid population lines (PPL)	1	156	0.012	=0.913	1.521	=0.219	6.842	=0.010*	30.554	<0.001*	0.803	=0.371
Fruit location (FL)	1	156	24.661	<0.001*	81.940	<0.001*	423.889	<0.001*	375.391	<0.001*	292.182	<0.001*
PPL×FL	1	156	1.918	=0.168	0.006	=0.979	1.342	=0.248	1.249	=0.265	0.746	=0.389

*Statistically significant

influence both adult emergence and parasitism values. These mortality factors, such as both stinging activity without oviposition and superparasitism, may increase host larva and/or pupa mortality rates. This would explain the high effectiveness values recorded for both parasitoid population lines. *Diachasmimorpha longicaudata* has an innate trend to superparasitize host larvae not only under rearing conditions (González et al. 2010; Montoya et al. 2012a) but also in natural environmental situations (Ovruski et al. 2012; Montoya et al. 2013). Additionally, *D. longicaudata* females may cause severe damage to host larvae due to the numerous punctures on them as a result of ovipositor probing's or by excessive number of ovipositions (Montoya et al. 2000a). Similar to that found

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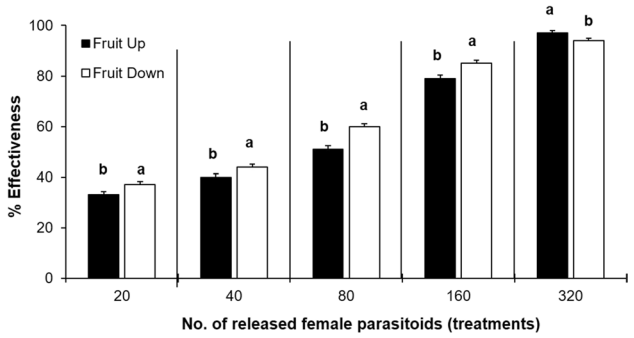

Fig. 6 Comparison between the mean (±SE) effectiveness of *Diachasmimorpha longicaudata* females released at five different densities to suppress medfly larvae infesting peaches located either in the upper (near to roof) or lower sector (on the ground) of field cages throughout autumn 2016; Tucumán, Argentina. Data of both *D. longicaudata* lineages were pooled. Letters show comparisons between fruits up and down within the same parasitoid density. Bars with dissimilar letter indicate significant differences (Tukey HSD test, $p=0.05$)

in this study, Montoya et al. (2000b, 2007) recorded just over 70% of control on *Anastrepha ludens* (Loew) and *A. obliqua* (Macquart) populations by open-field releases of *D. longicaudata* females mass-reared on *A. ludens* larvae, but with only a 50% of parasitism caused by this opiine species in different fruit-growing regions of México. This enhanced mortality is highly relevant for those biological control programs using exotic parasitoid species that are not able to establish themselves or whose establishment is doubtful in a particular area or region, because they can be used to control target pest outbreaks (de Pedro et al. 2019). The exotic *D. longicaudata* has been able to successfully establish in the subtropical areas of the northern Argentina as a direct result of early sporadic releases. However, it was recovered in low numbers from both *C. capitata* and *A. fraterculus* puparia 40 years after their first releases in that region (Schliserman et al. 2003; Oroño and Ovruski 2007). Distinctively, *D. longicaudata* was recovered from *C. capitata* puparia from different host fruit species a few weeks after its release in semi-arid and irrigated fruit-producing valleys of San Juan, but to date its establishment in the central-western region of Argentina has not been confirmed (Suárez et al. 2014; Sánchez et al. 2016). Regarding the offspring sex ratio, data of the present study agreed with Sánchez et al. (2016), due to the fact that in all treatments throughout both testing seasons *D. longicaudata* yielded a moderately female-biased offspring. Parasitism and progeny sex ratio are among the foremost quality control parameters used for *D. longicaudata* mass releases (Messing et al. 1993; Purcell 1998; Montoya et al. 2012b). This finding is relevant when taking into account future mass releases of this opiine parasitoid species under open-field conditions.

Results of this research showed that the females of both Dl_{BipCc} and Dl_{tslCc} comparably had the ability to forage infested peaches indistinctly over the tree canopy and on the ground beneath the tree. Similar parasitoid effectiveness rates were achieved in both host fruit location in both

AQ3 **Table 5** Summary of univariate two-factor GLMs with a covariable (outdoor mean temperature = T°C) on the effect of parasitoid population lines (=PPL) and testing seasons (=TS), and their interactions on *Diachasmimorpha longicaudata* effectiveness recorded per treatment from third instars of a biparental *Ceratitis capitata* strain inside peaches located at the canopy of potted tree and on the ground under field-cage conditions between January and February 2016, and April and May 2016; Tucumán, Argentina

Treatments												
Source of variation/fruit location	*df*	Error *df*	T_1 (20 released parasitoids)		T_2 (40 released parasitoids)		T_3 (80 released parasitoids)		T_4 (160 released parasitoids)		T_5 (320 released parasitoids)	
			F	*P*	*F*	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Fruit at the canopy												
Categorical variables:												
PPL	1	155	0.012	=0.913	0.348	=0.556	10.436	=0.001*	7.117	=0.008*	0.992	=0.878
TS	1	155	0.023	=0.877	0.109	=0.741	0.243	=0.623	0.035	=0.853	0.024	=0.358
PPL×TS	1	155	3.016	=0.084	1.493	=0.223	1.424	=0.623	1.075	=0.301	0.848	=0.084
Covariable (T°C)	1	155	0.992	=0.321	0.041	=0.838	1.014	=0.315	2.189	=0.141	0.992	=0.321
Fruit on the ground												
Categorical variables:												
PPL	1	155	0.138	=0.710	8.951	=0.003*	0.622	=0.432	18.172	<0.001*	0.254	=0.615
TS	1	155	0.993	=0.320	0.483	=0.488	1.757	=0.187	1.027	=0.313	1.458	=0.229
PPL×TS	1	155	0.755	=0.386	1.400	=0.239	2.381	=0.845	0.045	=0.833	1.127	=0.289
Covariable (T°C)	1	155	0.958	=0.329	0.757	=0.386	0.621	=0.432	3.312	=0.071	2.342	=0.127

*Statistically significant

aforementioned microhabitats. This finding matches with *D. longicaudata*'s foraging behavior that involved different fruits infested by *A. ludens* (García-Medel et al. 2007; Miranda et al. 2015) and by *C. capitata* (Harbi et al. 2018; Suárez et al. 2019b). However, in the present study, there was a significant but slight trend to forage fruit in the canopy at the highest tested female release density (320 parasitoids) in both summer and autumn. These data do not agree with outcomes from previous reports on guava orchards infested by *Bactrocera dorsalis* (Hendel) larvae in Hawaii (Purcell et al. 1994) and on mango orchards infested by *A. ludens* and *A. obliqua* larvae in Mexico (Montoya et al. 2000b), in which *D. longicaudata* females showed a strong trend to forage over fallen fruits. These varied preferences on the fruit location may be the result of either different or combined circumstances that influence the female parasitoid stimulation in the host searching behavior. Examples of these factors may include suitable host sizes and ages commonly present in such fruit that generate enough vibration cues but also chemical cues that stimulate the female to lay eggs (Duan and Messing 2000). *Diachasmimorpha longicaudata* may be able to detect volatile emissions from *C. capitata* larvae during the host-location procedure from a short distance, and during egg-laying activity chemical cues from host larvae have an influence on female ovipositor-probing behavior (Buonocore Biancheri et al. 2019). It is well known that fruigivorous tephritid larvae, which includes *C. capitata*, release para-ethylacetophenone, a volatile that stimulates attraction of *D. longicaudata* females, and encourages both probing and oviposition behaviors (Stuhl et al. 2011). The fruit ripeness degree is another essential factor that may influence the preference of the *D. longicaudata* female to forage fruit on ground or at canopy level. The short-range orientation of *D. longicaudata* females to the host would be regulated by a blend of odors emanating from both the host larva and the ripe fruit (Messing and Jang 1992; Eben et al. 2000; Silva et al. 2007; Segura et al. 2012). Additionally, Stuhl et al. (2011) emphasized the diversity of chemical stimuli involved in the *D. longicaudata* female attraction to find the host larva and to oviposit in it.

Interestingly, the environmental conditions evaluated during the study dates in both testing seasons, mean temperature and RH, did not influence the effectiveness of both Dl_{BipCc} and Dl_{tslCc} to attack *C. capitata* larvae. Despite the significant difference in mean temperature and RH that was recorded between the release dates in the summer and the equivalent ones in the autumn, both *D. longicaudata* population lines were similarly effective to control Medfly. During testing dates in autumn, mean temperature and RH were 4.8°C and 16.0%, respectively, lower than the corresponding ones in summer. Different authors (Meirelles et al. 2015; Harbi et al. 2018; de Pedro et al. 2019) pointed out that *D. longicaudata* seems to be highly resistant to extreme climatic conditions, mainly to temperature variations. In turn, Meirelles et al. (2015) checked that individuals of *D. longicaudata* lab-reared from *C. capitata* larvae had a lower thermal threshold (~ 8°C) than those reared in *A. fraterculus* (~ 13°C). In the present study, only the mean temperature was tested as a covariate in the statistical models since there was a significant and high positive correlation with RH. The average temperatures recorded during the two testing seasons, which varied approximately between 24 and 29°C, are included within a suitable temperature range for the *D. longicaudata* female's oviposition activity and for the development of the immature stages (Meirelles et al. 2015). Precisely, in seasons with mean temperatures above 25°C, *D. longicaudata* could develop more rapidly than at temperatures below 25°C (Meirelles et al. 2013).

To summarize, *D. longicaudata* releases at a ratio of less than one host larva inside the fruit per parasitoid female, which is equivalent to 320 released parasitoid females per EC, highly increased the *C. capitata* mortality in both testing seasons, early summer and middle autumn, under field-cage conditions in northwestern Argentina. Nevertheless, at a host:parasitoid ratio of about 1.3 Medfly larvae per *D. longicaudata* female under infested fruit located on the ground conditions in both studying seasons, Dl_{BipCc} females caused a host mortality similar to that ratio of 0.6:1 host:parasitoid. Either way, *D. longicaudata* females of both tested parasitoid population lines generally showed a linked effectiveness pattern to kill Medfly larvae when peaches were locating in both the upper and lower sectors of the field cages. This would be signaling that female foraging activity of both Dl_{BipCc} and Dl_{tslCc} may not be distinctly defined in view of the host fruit height level preference, which was canopy vs. ground. This fact increases the *D. longicaudata* action range based on the capacity of successful search for and locating the host in two different microhabitats. Furthermore, adults of both Dl_{BipCc} and Dl_{tslCc} were able to get a good performance as biological control agents of *C. capitata* in the trials under local climatic conditions at the testing times. This finding together with data published by Suárez et al. (2019b) clearly suggest that Dl_{tslCc} reared under mass-rearing conditions at the "BioPlanta San Juan" biofactory may be very useful to control *C. capitata* through augmentative releases between middle spring and middle autumn, that is, during at least eight months of the year, in the subtropical region of northwestern Argentina. Throughout this period, both *C. capitata* and *A. fraterculus* accurately increase their natural populations by using feral fruits into both large and small wild forest areas with different degrees of disturbance surrounding commercial fruit crops, as well as in deserted citrus orchards (Schliserman et al. 2014, 2016). In this regard, open-field augmentative releases from either Dl_{BipCc} or Dl_{tslCc} population lines within an

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area-wide integrated Medfly management approach (Hendrichs et al. 2007; Montoya et al. 2007) should be encouraged. Finally, outcomes of the present study support the use of augmentative biological control against *C. capitata* in fruit-growing regions of Argentina, as in other American countries affected by this invasive pest. However, an important step to be considered in the foreseeable future is the use of Neotropical-native parasitoid species, such as the figitid larval parasitoid *Ganaspis pelleranoi* (Brèthes) (Buonocore Biancheri et al 2019) and the diaprid pupal parasitoid *Coptera haywardi* (Ogloblin) (Núñez Campero et al. 2020), in combination with the exotic *D. longicaudata* as a viable alternative to improve Medfly control.

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Author Contribution MJBB, LCS, DSK, and SMO conceived the research and wrote the manuscript. MJBB and SMO conducted experiments. MJBB, LCS, and SMO analyzed data and conducted statistical analyses. All authors read and approved the manuscript.

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Declarations

Conflict of Interest The authors declare no competing interests.

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