



Sensitivity of an enriched ginger oil based trapping system for *Ceratitis* fruit fly pests (Diptera: Tephritidae)



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ABSTRACT

Enriched ginger oil (EGO) is a new male attractant for *Ceratitis* (Diptera: Tephritidae) species. The relative sensitivity of three *Ceratitis* pest species, *C. capitata*, *C. rosa* and *C. cosyra*, and their distance-dependent responses to EGO were determined using mark-release-recapture trials in three commercial fruit orchards in Mpumalanga, South Africa. Mature males of the three species (9–14 days after adult emergence) marked with fluorescent pigments were released at four distances: 25 m, 50 m, 100 m and 200 m from a centrally located white Delta trap baited with EGO. Different pigment colours were used for the different distances. Two releases were conducted in each orchard with an interval of one month between the releases. Traps were checked the following day, one week, two weeks and a month after release. Specimens captured were examined under a UV light to determine pigment colour on males of *C. capitata*, *C. rosa* and *C. cosyra*. There were no significant differences in recapture rates of the three *Ceratitis* species in the EGO baited trap. Most of the recaptures of all species occurred within a 50 m distance from the EGO baited trap. Most of the recaptures also occurred on the day following release. Based on the recapture rates obtained in this study, the probability of a trapping grid of 5 EGO baited traps per 2.59 km² capturing one or more flies of *C. capitata*, *C. rosa* and *C. cosyra* for a population consisting of 1000 males for each species was estimated at over 95%. The EGO based trapping system would be an effective detection method for *C. capitata*, *C. rosa* and *C. cosyra* in pest free areas and in areas of low pest prevalence.

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1. Introduction

Fruit fly pests (Diptera: Tephritidae) affect the production and trade of commercial fruit worldwide (White and Elson-Harris, 1994). Countries importing fruit commodities that are hosts to fruit flies may require that these commodities derive from areas that are either free of particular fruit fly pests or under low prevalence of established pests. In order to maintain and verify fruit fly free areas and areas of low fruit fly prevalence, surveys using sensitive trapping systems are usually required (FAO, 2006, 2008; Jang et al., 2014).

Fruit flies in the genus *Ceratitis* are indigenous to Africa and a number of *Ceratitis* species are key pests of commercial fruit in Africa (De Meyer, 2001). *Ceratitis capitata* (Wiedemann),

Mediterranean fruit fly (Medfly), is the only species in the *Ceratitis* genus that has expanded its distribution well beyond the African and Indian Ocean region (Malacrida et al., 2007) and is a notorious pest of many commercially grown fruit in countries where it has become established (White and Elson-Harris, 1994). Other polyphagous *Ceratitis* species, like *C. rosa* Karsch and *C. cosyra* (Walker), are important pests of many commercial fruit in the African region and are listed as quarantine pests in Europe and America (Augustin et al., 2012). Ecological and climate models have demonstrated that *C. rosa* and *C. cosyra* could become established beyond the Afrotropical region (De Meyer et al., 2008; De Villiers et al., 2013; Li et al., 2009). As such, in early detection and monitoring programmes for invasive *Ceratitis* species, the use of sensitive trapping systems would provide greater assurance in keeping areas either free of or under low prevalence for these pests.

Attractants recommended in trapping surveys of *Ceratitis* pest species are the male lure trimedlure, *tert*-butyl 4 (and 5)-chloro-*trans*-2-methylcyclohexane-1-carboxylate and food-based

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attractants, which are either liquid protein baits or synthetic food-baits like 3-component Biolure (IAEA, 2003). Male lure based trapping systems are usually more sensitive than systems with food-based attractants and are thus preferred in programmes for early detection of fruit fly pests (Cunningham, 1989a, b; Tan et al., 2014). For *Ceratitidis* pests, such as *C. cosyra*, which do not respond to trimedlure (Grout et al., 2011; White and Elson-Harris, 1994), terpinyl acetate is a recommended male lure (Hancock, 1989; White and Elson-Harris, 1994). In early detection programmes for *C. capitata* in California and South Australia, arrays of trimedlure traps are set out at 1.93 and 6.25 traps per square kilometre, respectively (Meats, 2014). The responses of *C. capitata* males to trimedlure baited traps have been shown to be inversely related to the distance of the flies to the traps, with most of the captures recorded when flies were within 50 m from these traps (Cunningham and Couey, 1986; Lance and Gates, 1994; Shelly et al., 2014; Wong et al., 1982). Using distance-dependent responses of *C. capitata* to trimedlure traps and probability models, Lance and Gates (1994) and Shelly et al. (2014) estimated the minimum detectable population sizes of *C. capitata* for particular trimedlure trapping densities. For the California early detection programme, consisting of 1.93 trimedlure traps per km², the minimum detectable population sizes of *C. capitata* were estimated to range between 2000 and 10000 males per km² (Shelly et al., 2014).

For *Ceratitidis* species, in particular for *C. capitata*, male lures which are more potent than trimedlure have been and continue to be investigated (Flath et al., 1994; Jang et al., 2010; Mwatawala et al., 2013, 2015; Nishida et al., 2000; Shelly, 2013; Shelly and Pahio, 2002). One of the promising new male lures discovered for the *Ceratitidis* group is enriched ginger root oil hereafter referred to as EGO (Mwatawala et al., 2013; Shelly and Pahio, 2002). EGO is a distilled oil from the root of ginger, *Zingiber officinalis* Roscoe, which contains the sesquiterpene hydrocarbon α -copaene and other sesquiterpenes (Shelly and Pahio, 2002). Alpha-copaene was found to be highly attractive to *C. capitata* males, (Flath et al., 1994; Nishida et al., 2000). The exposure to α -copaene and α -copaene containing oils like ginger root oil were shown to increase the mating success of *C. capitata* males, including the temperature sensitive lethal genetic sexing strain of *C. capitata*, which is widely used in Sterile Insect Technique (SIT) programmes across the world (Shelly, 2001; Shelly et al., 2002). Pre-release exposure of sterile *C. capitata* males to ginger root oil was found to increase the effectiveness of SIT programmes targeting *C. capitata* (Shelly et al., 2007) and is currently a common practice in many SIT facilities worldwide. Compared to ginger root oil used in SIT programmes, EGO was reported to contain 20 times more α -copaene (Shelly and Pahio, 2002). In the first field tests with EGO, conducted by Shelly and Pahio (2002), attraction of *C. capitata* males to the lure was found to be lower than that of the standard trimedlure. In the latter study, EGO was evaluated as a paste. EGO as an attractant for *Ceratitidis* species is currently commercially available in South Africa (Insect Science, Tzaneen, South Africa) and is marketed as EGO Pherolure in the form of a polyethylene bulb containing EGO liquid in various volumes. In field tests in Africa (Tanzania and South Africa) where EGO Pherolure was compared to trimedlure, EGO Pherolure was found to have equal or superior attractiveness to trimedlure for *C. capitata* (Manrakhan et al., 2017; Mwatawala et al., 2013, 2015). The field results in Africa have also shown that EGO Pherolure attracted a larger range of *Ceratitidis* species than trimedlure and was equal or superior to trimedlure in attractiveness for important *Ceratitidis* pests, like *C. rosa* and *C. cosyra* (Manrakhan et al., 2017; Mwatawala et al., 2013, 2015).

These recent tests conducted with the promising EGO Pherolure have been carried out on wild *Ceratitidis* populations at unknown densities. However, the relative sensitivity of different *Ceratitidis* pest

species to the EGO lure and the sensitivity of an EGO lure trapping system for key *Ceratitidis* pest species still remain unknown. The first two objectives of this study were to: (1) compare the sensitivity of males of three fruit fly pest species: *C. capitata*, *C. rosa* and *C. cosyra* to EGO and (2) determine the distance-dependent responses of male *C. capitata*, *C. rosa* and *C. cosyra* to EGO, using a mark-release-recapture technique. The third objective of the study was to estimate the detection sensitivity of an EGO Pherolure trapping system for *C. capitata*, *C. rosa* and *C. cosyra*, using field release-recapture results and probability models similar to those used by Lance and Gates (1994) and Shelly et al. (2014).

2. Materials and methods

2.1. Study sites

Releases and recaptures were conducted in three commercial orchards in Ehlanzeni district, Mpumalanga, in the north of South Africa. Each orchard was about 2 ha in size. All were located between 680 m and 738 m above sea level. The orchards were on three different farms and were between 7 and 20 km apart. Two of the commercial orchards were *Citrus sinensis* (Valencia orange) orchards (Crocodile Valley Estates: S25° 28' 26.18" E31° 00' 49.53"; Sterkspruit: S25° 26' 11.77" E30° 53' 08.51") and one of the commercial orchards was a *Persea americana* (avocado) orchard (Oewersig: S25° 25' 53.58" E30° 48' 59.46"). The study was carried out between December 2015 and February 2016, which was outside of the citrus and avocado ripening season. No fruit fly control actions were carried out in the orchards at the time of the study. Daily weather data for the study site were obtained from the Agrometeorology Division of the Agricultural Research Council, Stellenbosch, South Africa. The data were collected at weather stations that were either at the study site or within 20 km from the study site. The mean maximum temperature and mean minimum temperature during the study were 31.74 °C ± 0.29 °C and 19.38 °C ± 0.10 °C, respectively. The wind direction was mainly from the east-south east or east-north east. On the day of the first release at all sites, the wind speed varied between 0.94 and 1.14 m per second. On the day of the second release at all sites, the wind speed varied between 0.54 and 0.83 m per second.

2.2. Insect materials and marking

Ceratitidis capitata, *C. rosa* (previously known as the *C. rosa* microsatellite cluster R1, see De Meyer et al. (2015)) and *C. cosyra* used in the study were obtained from colonies maintained at Citrus Research International (CRI), Nelspruit, South Africa for over 200 generations. Colonies were refreshed with wild males reared from fruit every two years.

Each puparial batch of each fruit fly species was divided into four equal lots. Each lot was between 19.5 and 49 ml in volume. Each lot was dyed with a selected fluorescent pigment powder. Fluorescent pigments adhere to the puparia and are then retained in the ptilinum suture during adult emergence (Norris, 1957). These pigments can be readily identified on the adults under ultra-violet light (Norris, 1957). The same fluorescent pigment powder was used for all fruit fly species released at a particular distance in a particular site. Flies dyed with the two most contrasting colours were released closest and furthest from the trap. Four colours were used for four release distances: 25, 50, 100 and 200 m, at each site. A different set of four pigments was used between releases at a particular site in order to reduce effect of pigment colour on recapture of flies from a specific release distance and to have more contrasting colours between release distances. The fluorescent pigment powders used in the study were: Lunar Yellow 27, Astral

Pink 1, Magenta 10, Stellar Green 8 and Blaze 5 of the Swada HMP series, as well as Invisible Blue 70 of the Swada T series (all Swada, Cheshire, UK). In the first release at all sites, flies of all species dyed with Astral Pink 1, Magenta 10, Stellar Green 8 and Lunar Yellow 27 were released at 25 m, 50 m, 100 m and 200 m respectively from the trap. In the second release at all sites, flies of all species dyed with Lunar Yellow 27, Stellar Green 8, Invisible Blue 70 and Blaze 5 were released at 25 m, 50 m, 100 m and 200 m respectively from the trap. Pigments were added directly onto the pupae at the rate of 2 g/L and were evenly distributed by gently swirling the containers with the pupae and pigments. Flies were dyed at about two days before adult emergence. Pupae and emerged flies were kept in aerated cages (54.5 cm × 39.0 cm × 31.0 cm) at a temperature of 27.7 ± 0.0 °C until the release day. Emerged flies were provided with water and a mixture of sugar and yeast extract (Amberex 1003, Juneau, USA) at a rate of 3 parts sugar to 1-part yeast extract. One day before release, males of each species and each fluorescent pigment were then separated from the females and placed in smaller aerated containers (diameter: 12.0 cm, height: 7 cm) for release. Males were provided with water and a mixture of sugar and enzymatic yeast hydrolysate until release. For all species, males of 9–14 days old which were most likely mated (Manrakhan and Lux, 2009) were used in the releases.

2.3. EGO trapping system

The EGO Pherolure dispenser (Insect Science (Pty) Ltd, Tzaneen, South Africa) was used in the EGO trapping system evaluated. The EGO Pherolure is a polyethylene bulb that contains 2 ml of the EGO Pherolure liquid. The EGO Pherolure dispenser was placed inside a plastic basket and fitted in the middle of a white Delta trap (11 cm × 28 cm × 20 cm) (Insect Science (Pty) Ltd, Tzaneen, South Africa) over a sticky liner, which was placed on the floor of the trap (Fig. 1). Traps were hung at 1.5 m above the ground inside the tree canopy on the south-eastern side of the trees.

2.4. Releases and recaptures

The experimental layout for the release-recapture trials consisted of a centrally placed EGO Pherolure baited white Delta trap in the middle of each orchard and release points that were at the four distances: 25, 50, 100 and 200 m from the trap along four cardinal directions (north, south, east and west).



Fig. 1. White Delta trap containing EGO Pherolure dispenser used in the mark-release-recapture trials.

Two releases of each species were carried out in each orchard with a month interval between the releases. One-month interval was deemed sufficient to ensure that flies in the first release would not continue to be captured in the second release. In release-recapture trials on *C. capitata*, Lance and Gates (1994) and Shelly et al. (2014) recaptured most of the flies on the first day and first week after release, respectively.

Equal numbers of males of each species were released at each release point for a particular distance. For each release point in an orchard, 50 males of each species were collected for release such that there would be 200 males of each species released per distance and per site. However, during the second release at Sterkspruit only 25 males of *C. capitata* could be collected for release at each of the four points located at a distance of 200 m from the trap. Also, during the second release at Oewersig, only 35, 30 and 25 *C. capitata* males could be collected for release at each of the four points located at 25 m, 100 m and 200 m from the trap respectively. For the 50 m release distance at Oewersig during the second release, 50 males of *C. capitata* could be collected for release at each of the release points for that distance. Releases of marked flies at each release point were carried out by gently opening the aerated plastic container and allowing the flies to fly out. Those remaining at the bottom of the cage were gently tapped to initiate flight. Flies found dead in the release containers were brought back to the laboratory for identification and counting. The exact number of males of each species released at each release point was calculated as the number of males collected for release minus the number of dead males at the particular release point.

For each release, a new EGO Pherolure dispenser was used inside the white Delta trap. Traps were placed just before release in the middle of the orchard and were serviced the following day, one week, two weeks and a month after release at each site. During each trap service, the sticky liner was removed and replaced with a new liner. The liner was placed inside a plastic container such that the liner remained flat. The plastic containers with the liners were brought back to the laboratory for processing and analysis.

In the release-recapture trials, the sites and release times were considered as replicates. There were therefore six replicates.

2.5. Identification of recaptured flies

Each fly captured on the sticky liner was removed using a pair of weakly-sprung soft forceps. The fly was identified to species and sexed. Males of *Ceratitis capitata*, *C. rosa* and *C. cosyra* captured in the trap in a particular orchard on a particular date were placed in separate Petri dishes, which were then labelled according to species, site and recapture date. The thorax and abdomen of each male of each species were cut out from the insect body using a sharp micro-scalpel, so that only the head remained on the Petri dish. Each head was then dissected anteriorly so that the dye contained in the ptilium suture would be exposed. All forceps were cleaned with paper towel dipped in ethanol after the handling of each fly. The heads of the flies captured were then checked under UV light (TRAC light Pro, Labino, Sweden), peak wavelength of 365 nm, and through a stereo microscope (Leica EZ4D, Leica Microsystems, Germany) at 20× magnification to determine the presence of fluorescent pigment on the ptilinum. The presence of pigment and the pigment colour on each fly species was identified and recorded.

2.6. Data analysis and probability models

The overall effects of species, release distance and recapture period were tested using a log linear regression analysis. Data were first summarised as percentage recapture of males of each fly species for each site, distance released and recapture date owing to

the different numbers of males released at the different distances, sites and release periods. Since there were no captures of marked flies one month after release, the data for this recapture date were omitted from statistical analysis. The percentage recapture data were then converted to count data by multiplying by 100. The count data were log (x+1) transformed. A log-linear model assuming a Poisson data distribution was used (XLSTAT, Addinsoft) to determine the effect of release distance, species and recapture date on recaptures.

The probability of detection of *C. capitata*, *C. rosa* and *C. cosyra* with an EGO based trapping grid was estimated using the probability model of Lance and Gates (1994). In order to compare the sensitivity of the EGO trapping system with that of the trimedlure trapping system used for *C. capitata* detection in California (Shelly et al., 2014), a trapping density of five EGO baited traps per 2.59 km² was assumed. The mean proportion of trapped males (*p*) of each species at each release distance (25 m, 50 m, 100 m and 200 m) obtained in this study was used in the calculations. The probability of capturing zero flies for a population density *n*, at a certain distance from a trap was calculated as (1-*p*)^{*n*} for each species. Each distance used in the study was then set as a distance zone which was the distance from a single source of flies to a trap within the grid. The same distance zones used by Shelly et al. (2014) were used in the calculations in this study. The 25 m, 50 m, 100 m and 200 m corresponded to distance zones of 0–37.5 m, 37.5–75 m, 75–150 m and 150–250 m respectively, which thereby represented 0.9%, 12.8%, 24.3% and 62.1% respectively of an area per trap covering a circle of 406 m radius (1 trap covering 0.518 km²). The probabilities of capturing zero flies were weighted across the different distance zones and summed to derive the probability of zero capture of a fly species within a trapping grid. The probability of capturing >0 flies for a population of size *n* was equalled to 1-summed probabilities of capturing zero flies for a fly population of size *n*.

3. Results

3.1. Relative sensitivity and distance-dependent responses of *Ceratitis* males to EGO

The mean overall recapture rates (all releases combined) with the EGO baited trap for *C. capitata*, *C. rosa* and *C. cosyra* were 5.70% ± 0.81%, 8.51% ± 1.30% and 9.49% ± 1.36%, respectively. Despite overall numerical differences in recapture rates of the three species in an EGO baited trap, the differences between the species were not statistically significant (Table 1 and Fig. 2). Males of the three *Ceratitis* species released at a distance of 200 m from the EGO baited trap could be captured within one week. For all species, however, male responses to EGO were significantly higher when they were within 50 m from the male lure than when they were further away (Table 1 and Fig. 2). The distance dependent responses

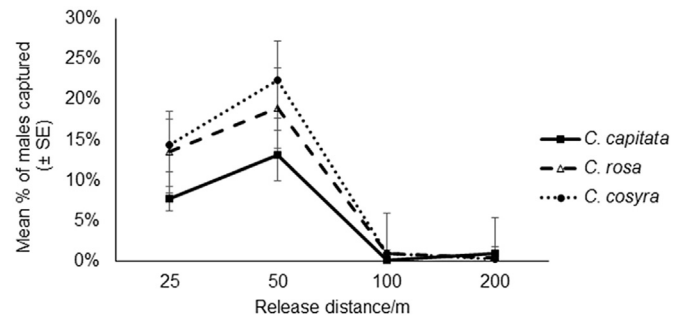


Fig. 2. Mean percentage of males of *C. capitata*, *C. rosa* and *C. cosyra* captured when released at four distances: 25 m, 50 m, 100 m and 200 m from a centrally placed EGO baited white Delta trap in commercial orchards in Mpumalanga, South Africa. Recapture data were pooled from all trap services (recapture dates and sites).

of *Ceratitis* males to EGO did not follow an exponential relationship. For all species, there were numerically higher recaptures of flies when released at 50 m compared to 25 m from the EGO trap, although recaptures of flies when released from these two distances were not statistically significant ($\chi^2 = 0.71$, *P* = 0.40). Recapture rates of all species were significantly higher on the day after release than a week or 2 weeks post release (One week after release: $\chi^2 = 12.66$, *P* = 0.00; Two weeks after release: $\chi^2 = 12.55$, *P* = 0.00) (Table 1 and Fig. 3). There were no recaptures of any of the released flies a month after their release (Fig. 3).

3.2. Detection sensitivity of an EGO trapping system

Based on the probability model of Lance and Gates (1994), the probability of capturing more than zero flies for a population of 1000 males of *C. capitata*, *C. rosa* and *C. cosyra* was over 95% when using an EGO trapping grid at 5 traps per 2.59 km² (Fig. 4).

In order to compare the sensitivity of an EGO baited trap with the sensitivity of a trimedlure baited trap for *C. capitata*, the detection probability estimates obtained for *C. capitata* in an EGO baited trap in this study were compared with the detection probability estimates derived from field recapture rates of *C. capitata* in trimedlure baited traps in two previously published studies (Fig. 4). The recapture rates of *C. capitata* males in a trimedlure baited Jackson trap (a white Delta trap as used in this study) obtained by Shelly et al. (2014) in residential areas in Waimanalo and Manoa, Hawaii and the recapture rates of *C. capitata* in a trimedlure baited Steiner trap obtained by Cunningham and Couey (1986) in a macadamia nut orchard in Hawaii were used to derive the probability estimates (Fig. 4).

For a population consisting of 1000 *C. capitata* males, the detection probability (probability of capturing >0 flies) in a commercial citrus and/or avocado orchard was 95.53% with an EGO baited trap. For the same *C. capitata* male population density, the detection probabilities in residential areas and in a commercial non-host orchard were 72.11% and 99.99%, respectively, with a trimedlure baited trap.

4. Discussion

Ceratitis capitata, *C. rosa* and *C. cosyra* were found to be equally attracted to the male lure EGO in this study. This is in contrast to responses of these congeneric species to the existing male lure trimedlure to which *C. cosyra* does not respond and *C. capitata* has a higher response than *C. rosa* (Grout et al., 2011). The release-recapture results of this study support the findings of catches of wild *C. cosyra* and *C. rosa* in EGO baited traps in both Tanzania and

Table 1

Log-linear regression results showing effects of species, release distance and recapture times on recaptures of *C. capitata*, *C. rosa* and *C. cosyra* in an EGO Pherolure baited white Delta trap. The data followed a Poisson distribution (Overdispersion test: *P* = 0.84).

Regression tests	Regression parameters	d.f	χ^2	<i>P</i> > χ^2
Statistic	-2 Log(Likelihood)	7, 208	96.89	<0.0001
	Score	7, 208	85.13	<0.0001
	Wald	7, 208	49.40	<0.0001
Source	Species	2, 208	0.24	0.89
	Release distance	3, 208	43.22	<0.0001
	Recapture date	2, 208	53.42	<0.0001

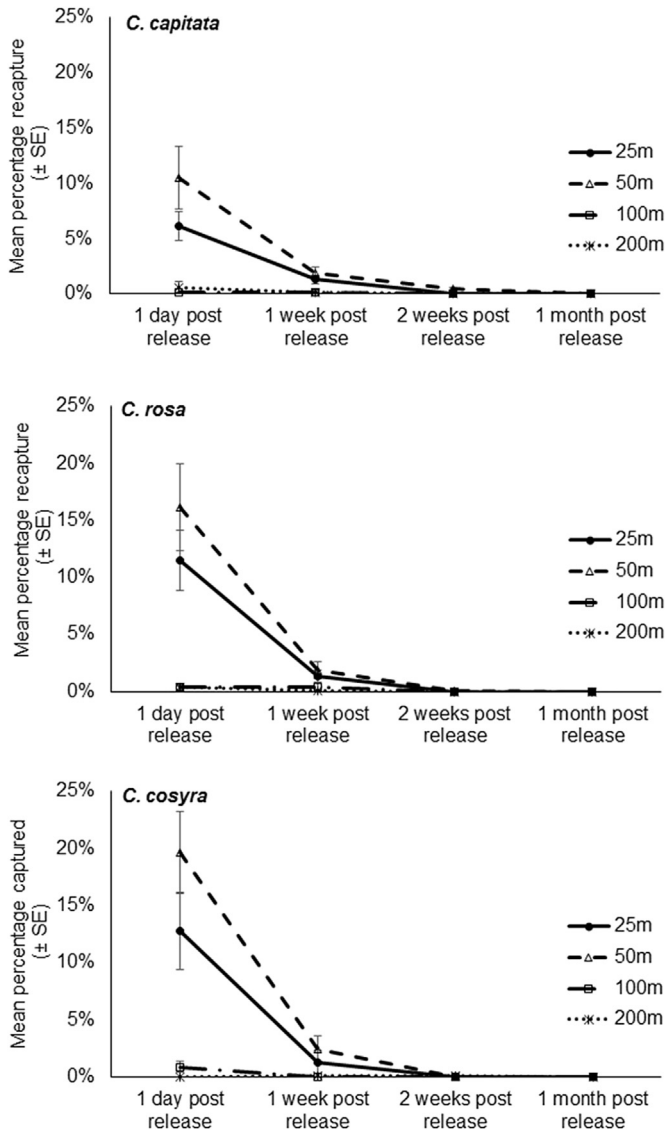


Fig. 3. Captures of males of *C. capitata*, *C. rosa* and *C. cosyra* across time when released at four distances: 25 m, 50 m, 100 m and 200 m from a centrally placed EGO baited white Delta trap in commercial orchards in Mpumalanga, South Africa.

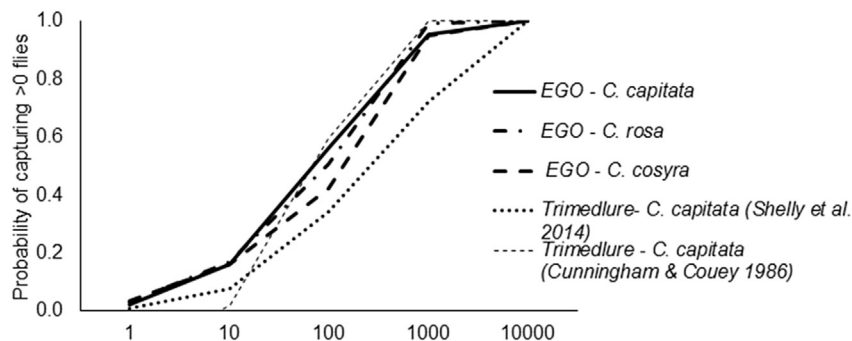


Fig. 4. Probability of capture of more than zero males of different population sizes of (1) *C. capitata*, *C. rosa* and *C. cosyra* in a trapping grid of 5 EGO baited traps per 2.59 km², (2) *C. capitata* in a trapping grid of 5 trimedlure baited traps per 2.59 km² based on field release-recapture results obtained by Shelly et al. (2014) in 3 residential sites in Waimanalo and Manoa, Hawaii and (3) *C. capitata* in a trapping grid of 5 trimedlure baited traps per 2.59 km² based on field release-recapture results obtained by Cunningham and Couey (1986) in a macadamia nut orchard, Hawaii. The probability model of Lance and Gates (1994) was used to derive the probability estimates.

South Africa (Manrakhan et al., 2017; Mwatawala et al., 2013, 2015). Our results here also support the conclusion of other studies that EGO targets a wider taxonomic spectrum than trimedlure (Mwatawala et al., 2013, 2015). Trimedlure, unlike EGO containing α -copaene, is an anthropogenic male lure (Tan et al., 2014). Trimedlure was developed mainly as an attractant for *C. capitata*, and its development followed the screening of a series of esters of carboxylic acid (Cunningham, 1989a). Exposure of *C. capitata* males to trimedlure was found to confer a mating advantage (Shelly, 1999) to these males, possibly through increases in calling (Shelly et al., 1996) although the mechanism behind this has not been fully elucidated. Alpha-copaene is a component of a number of plants and has been extracted from leaves of various citrus species (Nishida et al., 2000). Alpha-copaene, similar to trimedlure, has been shown to confer a mating advantage to *C. capitata*, possibly through elevated calling levels (Shelly, 2001). Ginger root oil, containing α -copaene, was also found to confer a mating advantage to *C. rosa* (Quilici et al., 2013). The similar levels of responses of *C. capitata*, *C. rosa* and *C. cosyra* to EGO could therefore result from similar advantages gained in all these species with regards to their mating behaviour.

When comparing our study with a similar mark-release-recapture study by Cunningham and Couey (1986) on *C. capitata* in a trimedlure baited trap in a *Macadamia* sp. F. Muell. orchard, we found that the overall recapture rate of *C. capitata* in the EGO baited trap (5.7%) was much lower than the overall recapture rate of *C. capitata* in the trimedlure baited trap (an estimated 28.3%). The recapture rate of *C. capitata* in the EGO baited trap in this study was, however, higher than recapture rates of *C. capitata* in trimedlure baited traps in similar studies by Lance and Gates (1994) and Shelly et al. (2014) in residential areas (recaptures ranging between below 1% and 3%). Lance and Gates (1994) also used sterile *C. capitata* males which could have different responses to male lures compared to non-sterile laboratory reared flies and wild flies (Wong et al., 1982). In this study, we used uniform orchard environments and non-sterile flies. Recapture rates of either wild or sterile *C. capitata* males in EGO baited traps in heterogeneous environments could possibly have been different to recapture rates obtained in this study.

Since we did not directly compare responses of *C. capitata* to EGO and trimedlure, it is difficult to suggest which would be a more effective attractant for *C. capitata*. In a recent study using fresh EGO in a similar dispenser as used in this study, Mwatawala et al. (2015) found that *C. capitata* had a higher response to EGO than to

trimedlure. However, results from other studies using the same EGO dispenser showed that when EGO and trimedlure were evaluated over a longer period (with aging of lures factored in), similar levels of catches of *C. capitata* were obtained in EGO and trimedlure baited traps (Manrakhan et al., 2017; Mwatawala et al., 2013). Shelly's (2013) results on the effect of lure age on responses of *C. capitata* males to EGO and trimedlure showed that as the lures aged, *C. capitata* males responded more to trimedlure traps than to EGO traps. Lengthening the field efficacy of EGO would greatly increase its effectiveness for *C. capitata*.

The results of this study indicated that *C. capitata*, *C. rosa* and *C. cosyra* effectively responded to the EGO lure from a distance of 50 m, although responses of the three fruit fly species could still be recorded when they were 200 m from an EGO trap. Recapture rates of flies released at the two closest distances to the centrally placed trap (25 m and 50 m) were similar. The lack of differences in recapture rates between 25 m and 50 m release distances could have reflected experimental conditions or experimental error. In the field, odour plumes from pheromones and other attractant sources were found not to be continuous and to rather occur as a series of bursts which vary in strength and duration downwind (Murlis and Jones, 1981). Concentration of odour, however, was found to reduce with distance from the source (Murlis and Jones, 1981). Environmental factors, such as wind, confounded by fly dispersal, could have led to flies perceiving similar odour plumes within the 50 m radius and as such could have shadowed potential differences in recapture rates between the 25 m and 50 m release distances. The possible experimental error in our study could have been the misidentification of fluorescent pigment powders used for the release distances that were closest to the trap. However, the trends of recaptures with release distances were found to be more or less the same for the first and second releases even with the use of different sets of pigment colours. For distances further than 100 m, contrasting colours were used to reduce possibility of errors in pigment colour recording. As such the maximum sampling range of at least 200 m for EGO cannot be contested. The EGO lure sampling range falls within the sampling range of trimedlure for *C. capitata* found in other studies (with most recaptures of *C. capitata* occurring between 46 m and 60 m from a trimedlure baited trap) (Cunningham and Couey, 1986; Lance and Gates, 1994; Shelly et al., 2014).

Based on the results of the probability model used in this study, the EGO lure based trapping system would be an effective detection method for *C. capitata*, with comparable or higher sensitivity to the trimedlure based trapping system. The advantage of the EGO lure trapping system is that it will also be a sensitive detection method for two other potentially invasive fruit fly pests, *C. rosa* and *C. cosyra*.

In conclusion, an EGO lure based trapping system could serve as an alternate attractant for area-wide detection programmes targeting *Ceratitis* pests. Thresholds of catches of different *Ceratitis* pest species should be developed for this trapping system as a guidance for control actions in areas of low pest prevalence. Further research should be conducted to test the sensitivity of the EGO lure based trapping system in a more heterogeneous environment, like residential areas. Moreover, the field longevity of commercially available dispensers of EGO lure should be tested.

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