Journal of Plant Diseases and Protection (2022) 129:853–860
https://doi.org/10.1007/s41348-022-00632-5

ORIGINAL ARTICLE

Efficacy of nanocapsules loaded with *Ectomyelois ceratoniae* (Zeller) (Lepidoptera: Pyralidae) sex pheromone as evaluated in wind tunnel and field trapping experiments

Amir-Hossein Danaye-Tous[1] · Shahriar Jafari[1] · Babak Heidary-Alizadeh[2] · Hossein Farazmand[2]

Received: 29 November 2021 / Accepted: 26 May 2022 / Published online: 14 June 2022

Abstract

The carob moth, *Ectomyelois ceratoniae* (Zeller) (Lep.: Pyralidae), is a worldwide pest of pomegranate. Although synthetic sex pheromone has been used extensively for the management of this pest, the major component (*Z,E*) -9,11,13-tetradecatrienal (trienal) is unstable. We microencapsulated synthetic trienal and demonstrated that a nanocapsule gel formulation was superior to unformulated trienal and other formulations, including the pheromone analogue (*Z,E*) -7,9,11-dodecatrienyl formate, in wind tunnel and field trapping experiments. No significant effect was observed between the total times spent by males from the release site to dispensers in different treatments in wind tunnels. In the field, the trienal nanocapsule formulation provided superior attraction for up to 4 weeks, while no other formulation induced trap catches for longer than 3 weeks. According to our finding, microencapsulation of *E. ceratoniae* pheromone components may provide improved trap lures and may be suitable for application in mating disruption.

Keywords Carob moth · pheromone · Microencapsulation · Pomegranate · (*Z, E*)-9,11,13-tetradecatrienal

Introduction

Pomegranate, *Punica granatum* L., (Myrtales: Punicaceae) originated from central Asia (Holland et al. 2009) and is now a cultivated crop species in many tropical and subtropical regions of the world, particularly India, Iran and Turkey (Kahramanoglu and Usanmaz 2016). In Iran, > 80,000 hectares of pomegranate orchards produce about one million tons of pomegranates each year (Ahmadi et al. 2016).

Carob moth, *Ectomyelois ceratoniae* (Zeller) (Lepidoptera: Pyralidae), is the main pest of pomegranate (Karami et al. 2010), date (Nay et al. 2006), almond (Madge 2014) crops worldwide, and in the Middle East causes 30 – 80% annual crop loss (Shakeri 2004; Mamay et al. 2016). Carob moth females lay their eggs on stamens in the calyx and the larvae feed on petals, pierce the fruit and enter it, creating brown spots on the peel, as well as hollow, cracked and rotten fruit that appears black and moldy and is unmarketable (Mamay et al. 2014). Once the larvae penetrate the fruit chemical pesticides are ineffective (Shakeri 2004). In Iran's Lorestan Province, carob moth is tri-voltine, with infestations persisting from the end of April to mid-November (Naserian et al. 2013).

Control tactics used against the carob moth in Iran include collection and removal of overwintering larvae in rotten pomegranates (Sheikh Ali et al. 2009), stamen removal, application of kaolin clay powder to disrupt oviposition (Mazhab et al. 2014), covering the crown with netting (Rafeie et al. 2011), release of the egg parasitoid *Trichogramma embryophagum* (Hartig) (Rezaei-Azqandi et al. 2015) and use of pheromone mediated mating disruption (Tamhankar et al. 2000; Cichón et al. 2004) or mass trapping (Pezhman and Saeidi 2018).

The main components of *E. ceratoniae* sex pheromone are (*Z,E*)-9,11,13-tetradecatrienal (trienal), (*Z,E*)-9,11-tetradecadienal and (*Z*)-9-tetradecenal (Noorbakhsh et al. 2017a; Varshovi et al. 2018). Instability of trienal leads to low efficiency of mating disruption (Baker et al. 1991). A pheromone analogue, (*Z, E*)-7,9,11-dodecatrienyl formate,

✉ Shahriar Jafari
Shahriar.jafari@gmail.com; Jafari.S@lu.ac.ir

[1] Department of Plant Protection, Faculty of Agriculture, Lorestan University, Khorramabad, Iran

[2] Agricultural Research, Education and Extension Organization (AREEO), Iranian Research Institute of Plant Protection, Tehran, Iran

is only weakly attractive (Avand-Faghih et al. 2012; Dhouibi et al. 2016). Successful pheromone applications require sustained release rates over the period that adult insects are active (Chen et al. 2018). Traditional formulations, such as microencapsulation, rubber septa, plastic tubes, hollow fibers and paraffin oil, can extend the duration of bioactivity (Gordon et al. 2005; Li et al., 2012). However, they provide inadequate control in field trials, and an alternative formulation is sought to retard breakdown of the trienal (Mitchell et al. 1976; Avand-Faghih et al. 2012).

Recent advances in pest control technology include microencapsulation of chemical pesticides and pheromones (Racuciu et al. 2009; Roy et al. 2010; Heidary et al., 2020), employing materials such as polyethylene (Bradley et al. 1995), porous silica glass (Tiboni et al. 2008), zeolites (Muñoz-Pallares et al. 2001) and paraffin wax (Atterholt et al. 1999) to form nanocapsules, nanospheres, micelles, nanogels, liposomes and inorganic nano-cages (Nuruzzaman et al. 2016; Chen et al. 2018) ranging in size from 10 to 1000 nm (Nagavarma et al. 2012). To achieve sustained release rates and efficacy, pheromones can be formulated into nanogels using a low molecular mass gelator (Bhagat et al. 2013), and hydrophobic pheromones can be encapsulated into nanoparticles (Chen et al. 2018; de Matos et al. 2019). Although many studies have been performed on the role of pheromones to control the carob moth, little is known about determining the efficacy of nanocapsule formulation of sex pheromone. Our objective was to evaluate the effect of nanoencapsulation on the stability and attractiveness of the trienal sex pheromone molecule as determined in wind tunnel and field experiments.

Materials and methods

Preparing the nanocapsule formulation

Analytical-grade materials used without further purifications were Gelatin (type G-2500) (MilliporeSigma, Merck KGaA, Darmstadt, Germany), gum Arabic (CARLO ERBA Reagents GmbH, Emmendingen, Germany), and thin layer chromatography (TLC) plates (25 TLC glass sheets 20×20, silica gel 60 F_{254}), hydrochloric acid and Tween 80 (Merck KGaA, Darmstadt, Germany). All aqueous solutions were prepared with distilled water. Synthesis of (*Z,E*)-9,11,13-tetradecatrienal (trienal) was performed according to Noorbakhsh et al. (2017b).

Nanoencapsulation was accomplished using complex coacervation (Sliwka 1975; Xing et al. 2005; Lv et al. 2013) of equal parts (w/v) of gelatin and gum Arabic (Danaye-Tous et al. 2020). Firstly, 0.112 g of gelatin was dissolved in 7.5 mL of water making a 1.5 wt % solution. The solution was stirred for 0.5 h, heated to 50 °C, and 75 mg of Tween 80 and 10 mg of the synthesized trienal were added while stirring. Then, 7.5 mL of the gum Arabic 1.5% solution was added, and the mixture was stirred at 200 rpm and sonicated for 1.0 h using a 250 W ultrasonicator (Ultrawave Ltd., Cardiff, UK). The pH of the solution was adjusted to 4.35 using diluted HCl and it was maintained at 50 °C for 1.0 h. Then, the nanocapsules were cross-linked by using formaldehyde (1 mL) at 40 °C. The resulting formulation was rinsed several times with distilled water, centrifugalized at 10,000 rpm for (15 min) and held in a vacuum oven at 30 °C for 12 h to obtain a white solid gel of nanoencapsulated trienal. Although the encapsulation efficiency was not measured chemically, the microcapsules mass product was washed with 1 mL dichloromethane and subjected to thin layer chromatography (hexane/ethyl acetate 4:1) in comparison with trienal, as seen in Fig. 1, three spots of trienal, mix trienal/ eluate microcapsules and elute microcapsules were placed on TLC and put it in TLC-tank with hexane/ethyl acetate eluent. After a few minute, the result showed the most trienal was adsorbed in microcapsules and no trienal was observed in eluate.

Morphology of nanocapsule formulation

The morphology of nanocapsule gel was characterized by depositing the suspension (2 µL) onto Formvar-coated

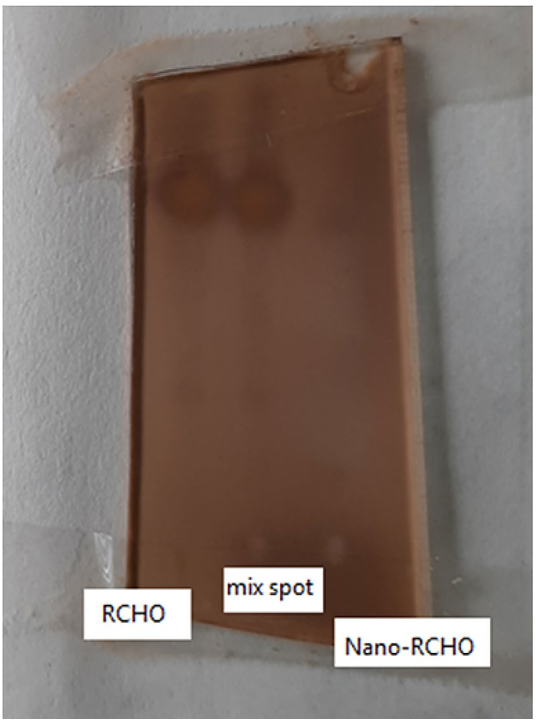

Fig. 1 TLC plate chromatography showing left spot, trienal particles; middle spot, mixture of unformulated trienal and trienal nanocapsules; right spot, trienal nanocapsules

copper grids (CM-300 Philips, Netherlands TEM), excess water was removed by filter paper, and the grids were allowed to dry for 1 h before examination by transmission electron microscopy (TEM) (200 kV Schottky field emitter HR) (Micro to Nano, Ltd., Haarlem, the Netherlands). The size of the nanocapsule particles was measured using purchased software (version 3.2, Soft Imaging System GmbH, Germany). The particle size and distribution of the nanocapsules were analyzed by a laser particle size analyzer (SZ-100z Dynamic Light Scattering and Zeta potential analyzer) (Horiba Ltd., Kyoto, Japan).

Ectomyelois ceratoniae rearing

Rotten pomegranates containing *E. ceratoniae* larvae were collected during November and December 2019 from the pomegranate orchards in western Lorestan Province, Iran. Larvae were excised and reared to maturity on fresh pomegranate fruits held in cages in incubators 29 ± 1 °C and 60 ± 5% RH. Emergent adults were held for 24 h and then used in experiments.

Wind tunnel experiment

Laboratory experiments were conducted in a wind tunnel (50 × 50 × 150 cm), wind speed 30 cm/s, red light (2 lx) held at 29 ± 1 °C and 60 ± 10% RH. The wind tunnel was kept in a room at 29 ± 1 °C, 60 ± 10% RH and under a photoperiod of 16L: 8 D h. To avoid odor contamination the room air was constantly renewed by an external fan. Four treatments, presented in randomized order, were 2.5 mg of synthetic trienal without dispenser in plastic vials, 1 mg of (*Z,E*)-7, 9, 11-dodecatrienyl formate on a rubber septum (Russell IPM, Deeside, UK), 100 mg of polymeric nanocapsule gel containing 2.5 mg of trienal) and control (no dispenser or pheromone). Adult males, 2–3 days old, were tested only once. In each bioassay, a single male was released 120 cm downwind of the test stimulus and given 3 min to initiate flight. Twenty males were tested against each stimulus. If no behavior was observed after 3 min, the moth was discarded. Sequential observations included time from release to take off, duration of upwind flight to 50 cm from the test stimulus, duration taken for close approach to within 10 cm of the test stimulus (Tasin et al. 2006; Borrero-Echeverry et al. 2015; Hatano et al. 2015; Celina et al. 2019), time resting (sitting) on the test stimulus and time taken in attempting to mate with the test stimulus.

Field experiment

A field experiment was conducted in pomegranate orchards, in Bizhenvand Village (33°41′16″ N, 47°50′43″ E), Sarab-e-Dowreh County, Lorestan Province, Iran for 5 weeks from September 7 to October 11, 2019. For all tests, Dispensers were placed in the middle of the sticky bottom (Daegil Co., Gyeongsangnam-do, South Korea) of white delta traps (200 × 200 base × 100 mm high) (Raha Andish Kavan Co., Tehran, Iran). Traps were hung 150 cm over the ground, spaced 50 m apart within the canopy of pomegranate trees. Five treatments (N = 4 replicates) were applied in a randomized complete block design: 2.5 mg trienal in an Eppendorf tube, 1 mg (*Z, E*)-7, 9, 11-dodecatrienyl formate (Russell IPM, Deeside, UK), 100 mg of polymeric nanocapsule gel containing 2.5 mg of trienal, 100 mg if glycerin containing 2.5 mg of the trienal, and unbaited and control traps. Glycerin was tested as a potential alternative method to nanoencapsulation to improve stability of the trienal. Traps were checked weekly and captured males were counted and recorded.

Data analysis

Because there was no response to the control stimulus in the wind tunnel, controls were eliminated from analysis. The proportional responses to the three remaining treatments were analyzed with a chi-square test. Mean durations of behavioral categories exhibited in the wind tunnel and mean responses to different treatments in the field experiment were evaluated with a generalized linear model (GLM), modified for a Poisson distribution for the field data (Lo et al. 2013; Borrero-Echeverry et al. 2015; Reger et al. 2020), followed by Tukey's HSD test (SAS Institute, Cary, NC, USA). In all cases, $\alpha = 0.05$. Graphs were drawn using Graph Pad Prism 8 software (GraphPad Software, San Diego, CA, USA).

Results

Nanocapsule characteristics

As shown in Fig. 2A–C, the nanocapsule trienal particles are relatively spherical and smooth, have a dense core and are partially in a gel. TEM images showed a size range of 300 to 350 nm, but the DLS spectra showed three distinct peaks in diameter (mean ± SD), 184.2 ± 22.7 nm, 1109 ± 234.5 nm and 6293.4 ± 787.4 nm. Because of the agglomeration of nanocapsules, there are three peaks and a broader particle size distribution in DLS spectra (Fig. 3).

Wind tunnel tests

In the wind tunnel experiment, 80% of test male *E. ceratoniae* contacted the gel nanocapsule lure, compared to 70% for (*Z,E*)-7, 9, 11-dodecatrienyl format, and 60% for the unformulated trienal, and no males were attracted to the control. When exposed to the nanocapsule gel stimulus, males

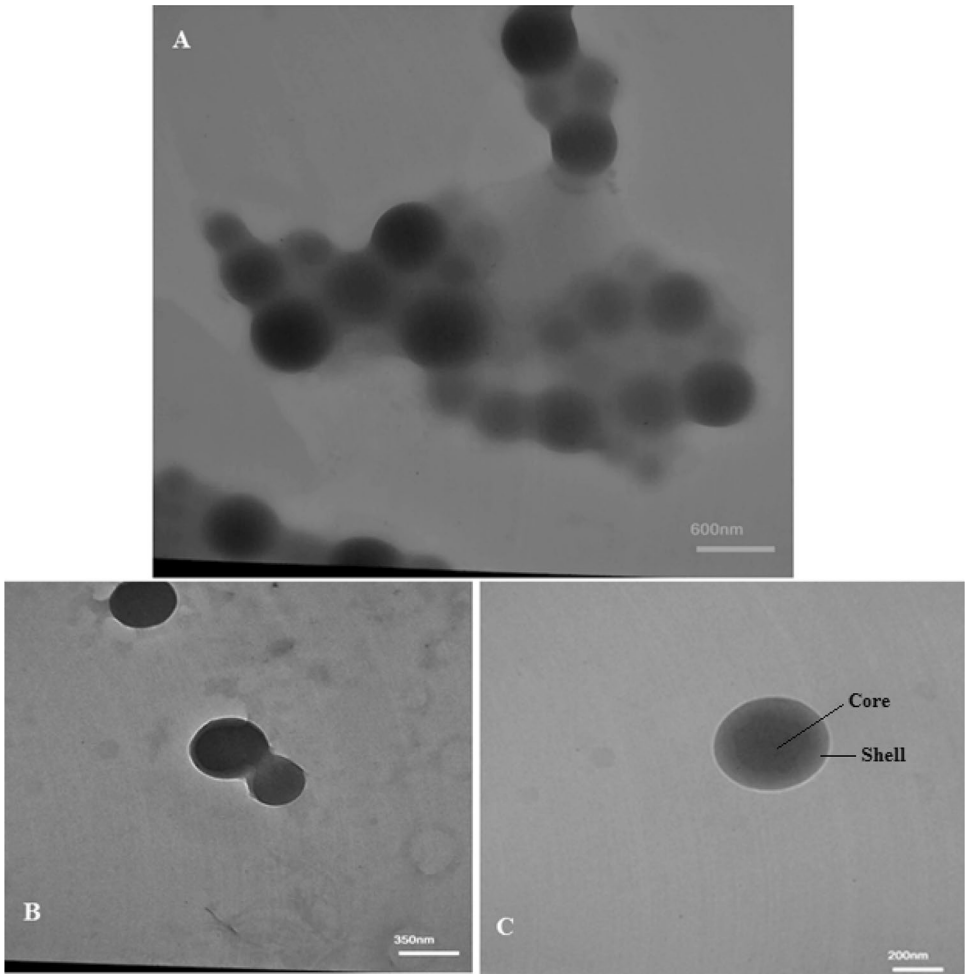

Fig. 2 (A, B, C). Transmission electron microscopy images (TEM) of nanocapsules of carob moth sex pheromone (original) (Adapted from Danaye-Tous et al. 2020)

initiated flight in a mean time of 8.10 ± 0.57 s (mean ± SE), slightly but significantly faster than 9.97 ± 0.49 s for (*Z,E*)-7,9,11-dodecatrienyl, while the take-off time for the unformulated trienal was not significantly different from either of the two ($F_{2, 12} = 4.23$, $P = 0.04$, Fig. 4). There was no difference among treatments for the time spent in oriented flight ($F_{2, 12} = 1.80$, $P = 0.20$), approaching ($F_{2, 12} = 2.62$, $P = 0.11$) or sitting on the stimulus source ($F_{2, 12} = 1.60$, $P = 0.24$) or in the total duration of the orientation sequence ($F_{2, 12} = 0.98$, $P = 0.40$). However, males spent 29.47 ± 0.66 s attempting to copulate with the nanocapsule lure significantly more time than either of the other two stimuli ($F_{2, 12} = 5.41$, $P = 0.02$).

Field tests

In the first week, the highest numbers of male *E. ceratoniae* (2.5 ± 0.29) were captured in traps baited with unformulated trienal; there was no significant difference between this catch and that in traps baited with nanocapsules or (*Z,E*)-7,9,11-dodecatrienyl, which in turn attracted no more males than traps baited with the trienal formulated in glycerin or unbaited control traps ($F_{4, 12} = 10.02$, $P < 0.001$, Fig. 5). After the first week, no males were captured in unbaited control traps. In weeks 2 and 3, the number of males captured by traps baited with trienal nanocapsules caught the highest numbers of males (2.75 ± 0.48 and 3.0 ± 0.41, respectively), but in week 2 the catch in traps baited with nanocapsules was not significantly higher than in traps baited with unformulated trienal ($F_{4, 12} = 12.57$, $P < 0.001$) and in week 3 it was no greater than in traps baited with (*Z,E*)-7,9,11-dodecatrienyl formate ($F_{4, 12} = 13.15$, $P < 0.001$) (Fig. 5). In week 4, traps baited with trienal nanocapsules caught significantly more male *E. ceratoniae* (3.5 ± 0.29) than traps baited with (*Z,E*)-7,9,11-dodecatrienyl or the glycerin trienal formulation ($F_{4, 12} = 50.71$, $P < 0.001$) (Fig. 5). In the fifth week, all traps caught very low numbers of males and there was no difference among treatments ($F_{4, 12} = 1.31$, $P = 0.3219$) (Fig. 5).

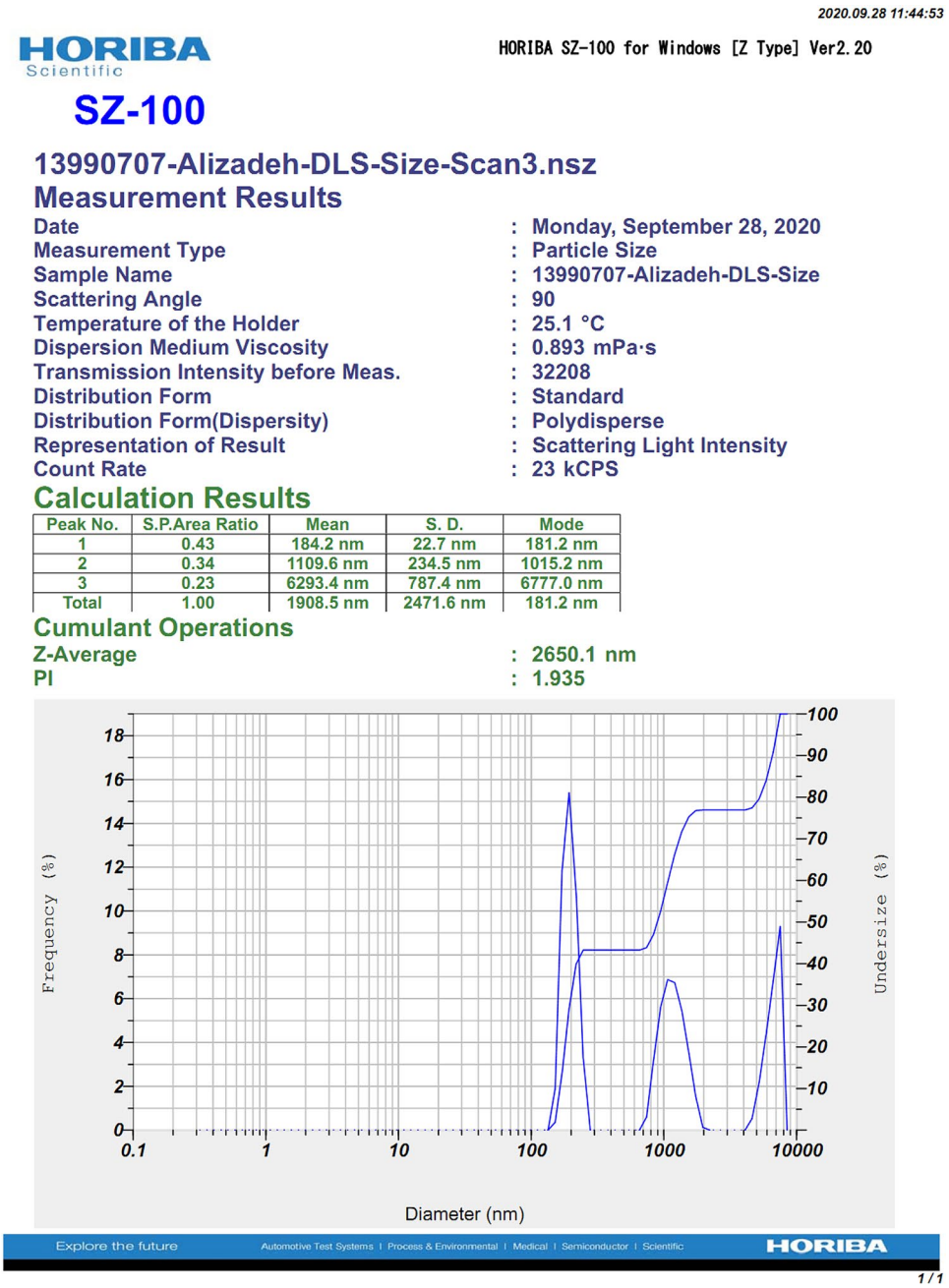

Fig. 3 DLS spectra of nanocapsules of carob moth sex pheromone (original)

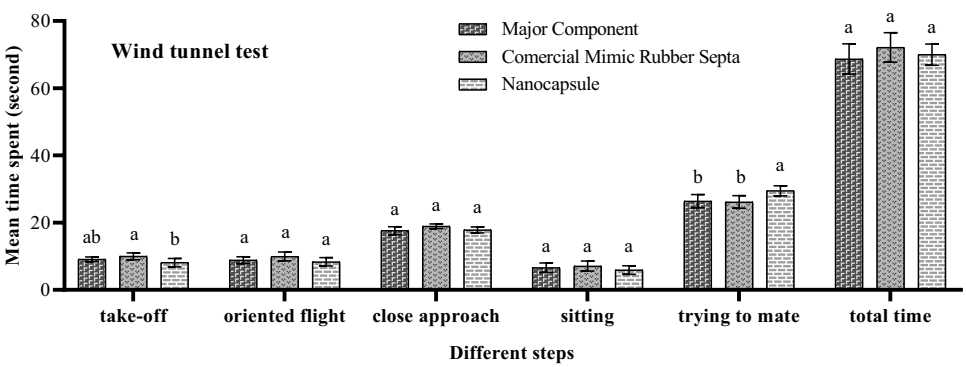

Fig. 4 Mean (±SE) times spent by male *E. ceratoniae* in a wind tunnel to take off, oriented flight, close approach, sitting, trying to mate and total time in different treatments. Bars with different letters for each behavior are significantly different; Tukey's HSD test, $P < 0.05$

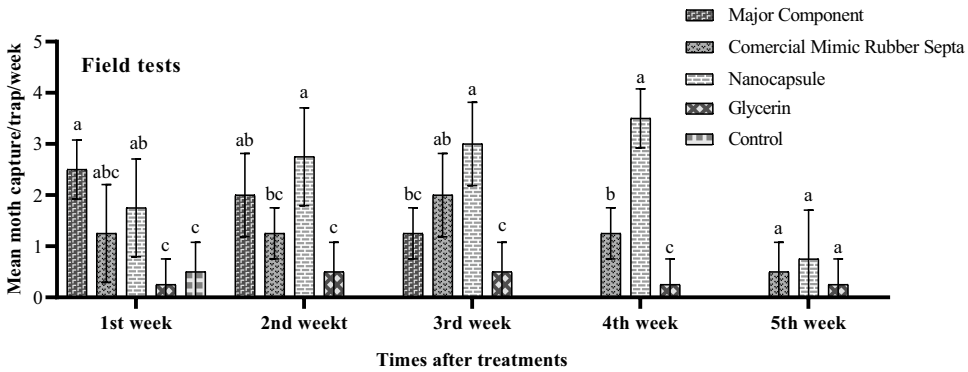

Fig. 5 Mean (± SE) captures of *E. ceratoniae* males per week for each treatment in a field trapping experiment from September 7 to October 11, 2019. Bars with different letters at each time point are significantly different, Tukey's HSD test, $P < 0.05$

Discussion

Our results showed that the nanocapsule gel is superior to other pheromone formulations in attracting male *E. ceratoniae* in a wind tunnel, that traps baited with trienal nanocapsules capture more males than traps baited with other pheromone formulations, and that the trienal nanocapsule gel formulation remains effective in attracting males for up to 4 weeks in the field. Therefore, we can assume that the amount of the remaining pheromone in the capsules in a longer time was still high enough and independently from the agglomeration that is a desired effect of the prepared formulation. The microencapsulated synthetic formulation in this study in addition to increasing the efficiency could be a suitable controlled release formulation, and nanocapsule gel formulation could have potential applications in mass trapping and mate disruption of carob moth in the field.

In this study, the nanocapsules were dispersed in three size ranges, with the largest mean diameter measuring 6,293.4 nm and accounting for 23% of the particle volume, while particles of mean diameters of 184.2 nm and 1,109.6 nm accounted for 43 and 34% of the volume, respectively (Fig. 3). The small particle size of the lower two categories, as well as the spherical shape of all particles, allows for optimal pheromone loading and controlled release (Perlatti et al. 2013; Stadler et al. 2018; Heidary et al. 2022).

Our results support those of Baker et al. (1991), who also demonstrated initiation of flight by male *E. ceratoniae* in a wind tunnel in response to trienal alone. The high percentage (80%) of male *E. ceratoniae* exhibiting mate-seeking behavior in the wind tunnel to trienal nanocapsules compares very favorably with wind tunnel responses of 76.9 – 85.7% of *E. ceratoniae* males to used pheromone of three different populations (Ziaadini et al. 2011). It is superior to the 65% response of male *E. ceratoniae* flying toward an 8:1:1 blend of trienal, (*Z, E*)-9,11-tetradecadienal (dienal) and (*Z*)-9-tetradecenal (monoenal), a 63% response to the above blend with (*Z,E*)-7,9,11-dodecatrienyl formate replacing trienal and respective responses of 50 and 70% to trienal and (*Z,E*)-7,9,11-dodecatrienyl formate alone (Todd et al. 1992).

The pheromone component analogue (*Z, E*)-7, 9, 11-dodecatrienyl formate is reportedly not very effective as a field trap lure (Avand-Faghih et al. 2012; Dhouibi et al. 2016), but in our experiment it provided trap catches comparable to those in traps baited with trienal nanocapsules in weeks 1 and 3 (Fig. 5). Moreover, the catch rate of 0.17 males/trap/night is almost identical to the rate of 0.168 males/trap/night to traps baited with a commercial (*Z, E*)-7, 9, 11-dodecatrienyl formate lure (Naserian et al. 2016).

Our results showing superior performance of traps baited with unformulated trienal in week 1, and then a decline to zero catches in week 4 are consistent with initial high release of a highly volatile compound and its expected inactivation due to oxidation over time (El-Sayed et al. 2006, 2021). In contrast, traps baited with the trienal nanocapsule rose to a higher level than other treatments by week 2 and were at their highest level and significantly higher than to other traps in week 4 before declining to a very low level by week 5. The performance suggests a slow rise to an optimal controlled release rate and also that microencapsulation protected the trienal from oxidative degradation, as predicted by (Ziaee et al. 2014). The results suggest that further studies are warranted to develop a commercial microencapsulated pheromone lure for use in monitoring *E. ceratoniae* populations as a component of integrated pest management programs, and also that a microencapsulated formulation should be tested for effectiveness in mating disruption.

Acknowledgements We thank the Department of Plant Protection, Lorestan University, Agricultural Research, Education and Extension Organization (AREEO), and the Iranian Research Institute of Plant Protection for providing space, laboratory equipment and financial support. We also thank Dr. J.H. Borden (Simon Fraser University, Canada) for helpful suggestions and review of the manuscript.

References

Ahmadi K, Gholizadeh H, Ebadzadeh HR, Hatami F, Hosseinpour R, Kazemifard R, Abdshah H (2016) Agricultural statistics, garden products, ministry of Jihad-e-Agriculture of Iran, Deputy for planning and economy, Information and communication technology center

Atterholt CA, Delwiche MJ, Rice RE, Krochta JM (1999) Controlled release of insect sex pheromones from paraffin wax and emulsions. J Control Release 57:233–247. https://doi.org/10.1016/s0168-3659(98)00119-9

Avand-Faghih A, Rastegari N, Jafari Nodooshan A (2012) Effect of dispenser type and dose of the pheromone mimic, (Z, E)-7,9,11-dodecatrienyl formate, on trapping of carob moth, *Ectomyelois ceratoniae* (Zeller). In: Proceedings of the 20th Iranian Plant Protection Congress. p. 841 Shiraz: Shiraz University, Iran, 26–29 August 2012

Baker T, Francke W, Millar J, Löfstedt C, Hansson B, Du J, Phelan P, Vetter R, Youngman R, Todd J (1991) Identification and bioassay of sex pheromone components of carob moth, *Ectomyelois ceratoniae* (Zeller). J Chem Ecol 17:1973–1988. https://doi.org/10.1007/BF00992582

Bhagat D, Samanta SK, Bhattacharya S (2013) Efficient management of fruit pests by pheromone nanogels. Sci Rep 3:1294. https://doi.org/10.1038/srep01294

Borrero-Echeverry F, Becher PG, Birgersson G, Bengtsson M, Witzgall P, Saveer AM (2015) Flight attraction of *Spodoptera littoralis* (Lepidoptera, Noctuidae) to cotton headspace and synthetic volatile blends. Front Ecol Evol 3:56. https://doi.org/10.3389/fevo.2015.00056

Bradley SJ, Suckling DM, McNaughton KG, Wearing CH, Karg G (1995) A temperature-dependent predictive model for polyethylene tubing pheromone dispenser release rates. J Chem Ecol 21:745–760. https://doi.org/10.1007/BF02033458

Celina B, Saluso A, Murua MG, Guerenstein PG (2019) Evaluation of sex pheromone formulations to attract *Spodoptera frugiperda* (Lepidoptera: Noctuidae) adult males in Argentina. Rev Soc Entomol Argent 78:7–20

Chen Y, Chen X, Chen Y, Wei H, Lin S, Tian H et al (2018) Preparation, characterisation, and controlled release of sex pheromone-loaded MPEG-PCL diblock copolymer micelles for *Spodoptera litura* (Lepidoptera: Noctuidae). PLoS ONE 13(9):e0203062

Cichón LI, Trematerra P, Coracini MDA, Fernandez D, Bengtsson M, Witzgall P (2004) Sex pheromone of *Argyrotaenia pomilliana* (Lepidoptera: Tortricidae), a leafroller pest of apples in Argentina. J Econ Entomol 97:946–949. https://doi.org/10.1093/jee/97.3.946

Danaye-Tous AH, Jafari S, Heidary Alizadeh B, Farazmand H (2020) Evaluation of field efficiency of the nanocapsule sex pheromone dispensers of carob moth Ectomyelois ceratoniae (Lep: Pyralidae). Plant Pests Res 10(3):49–60

de Matos SP, Lucca LG, Koester LS (2019) Essential oils in nanostructured systems: challenges in preparation and analytical methods. Talanta 195:204–214. https://doi.org/10.1016/j.talanta.2018.11.029

Dhouibi MH, Hermi N, Soudani D, Thlibi H (2016) Biocontrol of the carob moth *Ectomyelois ceratoniae* (Lepidoptera, Pyralidae) in pomegranate and citrus orchards in Tunisia. Int J Agri Innov Res 4:849–856

El-Sayed AM, Ganji S, Gross J, Giesen N, Rid M, Lo PL, Kokeny A, Unelius CR (2021) Climate change risk to pheromone application in pest management. Sci Nat 108:47. https://doi.org/10.1007/s00114-021-01757-7

El-Sayed AM, Suckling DM, Wearing CH, Byers JA (2006) Potential of mass trapping for long-term pest management and eradication of invasive species. J Econ Entomol 99:1550–1564

Gordon D, Zahavi T, Anshelevich L, Harel M, Ovadia S, Dunkelblum E et al (2005) Mating disruption of *Lobesia botrana* (Lepidoptera: Tortricidae): Effect of pheromone formulations and concentrations. J Econ Entomol 98:135–142

Hatano E, Saveer AM, Borrero-Echeverry F, Strauch M, Zakir A, Bengtsson M, Ignell R, Anderson P, Becher PG, Witzgall P, Dekker T (2015) A herbivore-induced plant volatile interferes with host plant and mate location in moths through suppression of olfactory signalling pathways. BMC Biol 13:75. https://doi.org/10.1186/s12915-015-0188-3

Heidary M, Jafari S, Karimzadeh J, Negahban M, Shakarami J (2020) The effects of pure and nanocapsulated formulations of *Thymus daenensis* Celak (Lamiaceae) essential oil on life-table parameters of cabbage aphid (*Brevicoryne brassicae* L.) (Hem.: Aphididae). Plant Pest Res 10:15–32. https://doi.org/10.22124/iprj.2020.4289

Heidary M, Karimzadeh J, Jafari S, Negahban M, Shakarami J (2022) Aphicidal activity of urea–formaldehyde nanocapsules loaded with the *Thymus daenensis* Celak essential oil on *Brevicoryne brassicae* L. Int J Trop Insect Sci 42(2):1285–1296. https://doi.org/10.1007/s42690-021-00646-w

Holland D, Hatip K, Bar-Yaakov I (2009) Pomegranate: Botany, Horticulture and Breeding. In: Janick J (ed) Horticultural Reviews, 35th edn. John Wiley and Sons Inc, pp 127–191

Karami A, Mirabzadeh A, Rafeie Karahroudi Z, Tafaghodinia B, Loonie S (2010) Comparing the methods of removing flags and the release of *Trichogramma embryophagum* Hartig (Hym., Trichogrammatidae) against the carob moth (*Ectomyelois ceratoniae* Zeller (Lep: Pyralidae)). Q J Spec Entomol Res 2:271–277

Kahramanoglu I, Usanmaz S (2016) Pomegranate production and marketing. CRC Press

Li QY, Liu JL, Zhao LL, Ma RY (2012) Applications of slow release technique of sex pheromone in pest control. Chin J Biol 28:589–593

Lo PL, Walker JTS, Horner RM, Hedderley DI (2013) Development of multiple species mating disruption to control codling moth and leaf rollers (Lepidoptera Tortricidae). N Z Plant Prot 66:264–269. https://doi.org/10.30843/nzpp.2013.66.5646

Lv Y, Zhang X, Zhang H, Abbas S, Karangwa E (2013) The study of pH-dependent complexation between gelatin and gum Arabic by morphology evolution and conformational transition. Food Hydrocoll 30:323–332. https://doi.org/10.1016/j.foodhyd.2012.06.007

Mamay M, Ikinci A, Unlu L, Dogan E (2014) Infestation rate and host preference of carob moth [*Apomyelois* (=*Ectomyelois*) *ceratoniae* Zeller (Lepidoptera: Pyralidae)] on different pomegranate cultivars. Turkish J Entomol 38:101–110

Mamay M, Unlü L, Yanık E, Dogramaci M, Ikinci A (2016) Efficacy of mating disruption technique against carob moth, *Apomyelois ceratoniae* Zeller (Lepidoptera: Pyralidae) in pomegranate orchards in Southeast Turkey (Şanlıurfa). Int J Pest Manag 69:295–299. https://doi.org/10.1080/09670874.2016.1185552

Madge D (2014) Carob moth eating your profits? In: Proceedings of 16th Australian almond conference. Australia, p 33

Mazhab S, Farazmand H, Vafaei-Shoushtari R, Emami MS (2014) Effect of kaolin and stamen-removing on damage reduction of pomegranate fruit moth, *Ectomyelois ceratoniae* (Lep.: Pyralidae). J Entomol Res 6:183–190

Mitchell ER, Baumhover AH, Jacobson M (1976) Reduction of mating potential of male *Heliothis* spp. and *Spodoptera fuglperda* in field plots treated with disruptants. Environ Entomol 5:484–486

Muñoz-Pallares J, Corma A, Primo-Yufera J (2001) Zeolites as pheromone dispensers. J Agric Food Chem 49:4801–4807. https://doi.org/10.1021/jf010223o

Nagavarma BVN, Yadav Hemant KS, Ayaz A, Vasudha LS, Shivakumar RHG (2012) Different techniques for preparation of polymeric nanoparticles–a review. Asian J Pharm Clin Res 5:16–23

Naserian N, Farazmand H, Vafaei Shoushtari R, Avand-Faghih A, Azadbakht N (2013) The seasonal population fluctuations of pomegranate fruit moth, *Ectomyelois ceratoniae* Zeller and quince moth, *Euzophera bigella* Zeller in pomegranate orchards. J Entomol Res 5:161–170

Naserian N, Farazmand H, Vafaei Shoushtari R, Avand-Faghih A (2016) Study of effective factors in capture of pheromone trap

carob moth, *Ectomyelois ceratoniae* Zeller. J Plant Protect 30:481–487

Nay JE, Boyd EA, Perring TM (2006) Reduction of carob moth in 'Deglet Noor'dates using a bunch cleaning tool. Crop Prot 25:758–765. https://doi.org/10.1016/j.cropro.2005.10.010

Noorbakhsh S, Heydari Alizadeh B, Farazmand H, Saber M (2017a) Comparative study of the sex pheromone of carob moth, Apomyelois ceratoniae (Zeller 1839) (Lepidoptera: Pyralidae) from four regions of Iran using headspace solid phase micro extraction–gas chromatography/mass spectrometry. Turkish J Entomol 41:61–73. https://doi.org/10.16970/ted.61775

Noorbakhsh S, Heidary Alizadeh B, Saber M, Farazmand H (2017b) Improved Synthesis of (Z, E)-9,11,13-Tetradecatrienal, the sex pheromone of the carob moth *Apomyelois (=Ectomyelois) Ceratonia*e (Lepidoptera: Pyralidae). J Chem Res 41:199–201. https://doi.org/10.3184/174751917X14894997017450

Nuruzzaman M, Rahman MM, Liu Y, Naidu R (2016) Nanoencapsulation, nano-guard for pesticides: a new window for safe application. J Agri Food Chem 64:1447–1483. https://doi.org/10.1021/acs.jafc.5b05214

Perlatti B, de Souza Bergo PL, Fernandes JB, Forim MR (2013) Polymeric nanoparticle based insecticides: a controlled release purpose for agrochemicals. In: Trdan S (ed) Insecticides development of safer and more effective technologies. IntechOpen

Pezhman H, Saeidi K (2018) Evaluation of light traps, pheromone traps and light–pheromone traps for mass trapping of *Ectomyelois ceratoniae* (Lepidoptera: Pyralidae) and determination the best pheromone traps density to decrease the pest damage in a pomegranate orchard. J Entomol Res 10:13–28

Racuciu M, Miclaus S, Creanga D (2009) The response of plant tissues to magnetic fluid and electromagnetic exposure. Rom J Biophys 19:73–82

Rafeie B, Farazmand H, Goldasteh Sh, Sheikh Ali T (2011) Investigating the impact of various fruit covering in reducing the damage of carob moth in pomegranate, *Ectomyelois ceratoniae* Zeller (Lep: Pyralidae), in Saveh region. J Entomol Res 3:1–11

Rezaei-Azqandi S, Kazazi M, Abdul Ahadi F (2015) *Ectomyelois ceratoniae* zeller (Lep., Pyralidae) and its control procedures in Iran. J Appl Environ Biol Sci 5:743–747

Reger J, Wenger J, Brar G, Burks C, Wilson H (2020) Evaluating response of mass-reared and irradiated navel orangeworm, *Amyelois transitella* (Lepidoptera: Pyralidae), to crude female pheromone extract. Insects 11:703

Roy D, Cambre JN, Sumerlin BS (2010) Future perspectives and recent advances in stimuli-responsive materials. Prog Polym Sci 35:278–301. https://doi.org/10.1016/j.progpolymsci.2009.10.008

Shakeri M (2004) A review on investigations on pomegranate neck worm in Iran, A proceeding on evaluation of finding and current problems associated with *Spectrobates ceratoniae* management in pomegranate. In: Ministry of Jihad-e-Agriculture, organization of research and education, Yazd Agriculture and Natural Resources Research Center, Iran, pp 18–30

Sheikh Ali T, Farazmand H, Vafaii Shoshtari R (2009) The impact of flag removing method in reducing the damage of *Ectomyelois ceratoniae* Zeller (Lep, Pyralidae) Saveh region. J Entomol Res 1:159–167

Sliwka W (1975) Microencapsulation. Angew Chemie Int Ed 14(8):539–550

Stadler T, Buteler M, Valdez SR, Gitto JG (2018) Particulate nanoinsecticides: a new concept in insect pest management. In: Begum G (ed) Insecticides: agriculture and toxicology. InTech, pp 83–105

Tamhankar AJ, Gahukar RT, Rajendran TP (2000) Pheromones in the management of major lepidopterous and coleopterous pests of cotton. Integr Pest Manag Rev 5:11–23. https://doi.org/10.1023/A:1009675920393

Tasin M, Bäckman AC, Bengtsson M, Varela N, Ioriatti C, Witzgall P (2006) Wind tunnel attraction of grapevine moth females, *Lobesia botrana*, to natural and artificial grape odor. Chemoecology 16:87–92. https://doi.org/10.1007/s00049-005-0332-6

Tiboni A, Coracini MDA, Lima ER, Zarbin PHG, Zarbin AJG (2008) Evaluation of porous silica glasses as insect pheromone dispensers. J Braz Chem Soc 19:1634–1640. https://doi.org/10.1590/S0103-50532008000800026

Todd JL, Millar JG, Vetter RS, Baker TC (1992) Behavioral and electrophysiological activity of (Z, E)-7,9,11-dodecatrienyl formate, a mimic of the major sex pheromone component of carob moth, *Ectomyelois ceratoniae*. J Chem Ecol 18:2331–2352. https://doi.org/10.1007/BF00984953

Varshovi F, Heidari Alizadeh B, Farazmand H, Oliali Torshiaz A, Sirjani M (2018) Extraction and identification of the sex pheromone of different populations of carob moth, *Ectomyelois ceratoniae*. Pestic Plant Prot Sci 4:165–177. https://doi.org/10.22092/jppps.2018.118518

Ziaadini M, Goldansaz SH, Ashouri A, Ghasempour A (2011) Study on the sexual behavior and male cross-attraction among three geographical populations of the carob moth, *Ectomyelois ceratoniae* under laboratory conditions. Iranian J Plant Prot Sci 42:151–161

Ziaee M, Moharramipour S, Mohsenifar A (2014) MA-chitosan nanogel loaded with *Cuminum cyminum* essential oil for efficient management of two stored product beetle pests. J Pest Sci 87:691–699

Xing F, Cheng G, Yi G, Ma L (2005) Nanoencapsulation of capsaicin by complex coacervation of gelatin, acacia, and tannins. J Appl Polym Sci 96:2225–2229. https://doi.org/10.1002/app.21698