Full Length Research Paper

Efficiency of Methyl eugenol as attractant for Acanthiophilus helianthi Rossi, 1794 (Diptera: Tephritidae)

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Fruit fly is one of the major pests that attack agronomical commodities, as well as the insect becomes a major pest in safflower plantation all over the world. The objective of this research was to study the response of fruit fly on methyl eugenol (ME) used as attractant. The safflower fly is one of the most important pests of safflower in Iran. Losses caused by larval feeding leads to disrupted plant activities, reduction in flower buds, and ultimately to decreased quality and quantity of crop. Infestation of bolls began 15 days after the formation of flower heads. Laboratory studies show that among the three species of fruit flies tested for attractiveness to methyl eugenol, male of *Acanthiophilus helianthi* Rossi (Diptera: Tephritidae) and *Chaetrollia carthami* (Diptera: Tephritidae) showed no significant difference but both are significantly different (P < 0.05) with *Trellia luteolla* (Diptera: Tephritidae). Virgin adult male's *A. helianthi* more than 18 days old were greatly attracted to methyl eugenol.

Keywords: Efficacy, bait trap, control, safflower fly.

INTRODUCTION

Fruit flies (Diptera, Tephritidae) cause large losses to fruits throughout the world, and are recognized today as major insect pests of the horticultural industries (Permalloo, 1989). Their significance is further increased by a growing international trade (Drew and Allwood, 1997). Fruit fly problems in Iran date back to the beginning of this century (Sabzalian et al., 2010). Fruit flies have been the subject of experimentation and control for many years (Hammes, 1980; Anon, 1983; Anon, 1996; Landell Mills, 1991). Despite an intensive programme of biological control (Hammes, 1980), a long term method of control: the Sterile Insect Release Method (Hammes 1980) and the use of insecticides on backyard gardens (Anon, 1985), the high fly populations and the abundance of fruits throughout the year combined to maintain the status of the fruit flies as the major pests of cultivated fruits. Males of many fruit flies are strongly attracted to methyl eugenol, a compound found naturally in a variety of plant species (Metcalf and Metcalf, 1992). Methyl eugenol plays a major role in male mating behavior by serving as a pheromonal precursor. Working with Bactrocera opiliae (Drew and Hardy) and B.

dorsalis (Hendel), respectively, Fitt (1981) and Nishida et al. (1988) reported that males fed on methyl eugenol produced volatiles which contained metabolites of this compound. whereas unfed males lacked these metabolites. Additionally, Shelly and Dewire (1994) showed that wild males of *B. dorsalis* which ingested methyl eugenol exhibited increased signal effort, signal attractiveness, and mating success compared with males not given access to the lure. Additional tests similarly revealed that irradiated males exposed to methyl eugenol gained a mating advantage over unexposed wild males for up to three weeks after feeding on the lure (Shelly, 1995). Moreover, irradiated males exposed to methyl eugenol were less likely to be captured in lure-baited traps than were unexposed irradiated males (Shelly, 1995). These results suggest that the methyl eugenolmale association could potentially be incorporated in the sterile insect technique (SIT). Specifically, the abundance of wild males could be reduced initially via male annihilation, and then lure-fed irradiated males could be released concurrent with continued male annihilation. If successful, this approach would effectively replace wild

males with irradiated males and thereby generate a high proportion of irradiated male by wild female mattings. Effort to overcome the problem is referred better aimed at integrated pest management. Alternative that have prospect for developed is usage attractant (Epsky and Heath, 1998; Manrakhan and Price, 1999; Bueno and Jones, 2002; Gopaul and Price, 2002; Rouse et al., 2005). Attractant is one of tool to monitor pest population and at the same time applicable to depress population Bactroceraspp. (Bueno and Jones, 2002; Michaud, 2003). Enticing Substance that contain single component (males lure) called pharapheromone that only effective to captivate male fruit fly. Methyl eugenol compound have characteristic in common with pharapheromone that can attract male insect (Iwahashi et al., 1996; Manrakhan and Price, 1999). According to Nurdijati and Tan (1996); Kardinan et al. (1999); Miele et al. (2001) and Kothari et al. (2005) basil plant have prospect as methyl eugenol source. The objective of this research was to study the response of fruit fly on methyl eugenol (ME) used as attractant. Safflower (Carthamus tinctorius L.) is an important oilseed crop and an essential component of cropping systems in the dry regions and marginal areas of the world (Sabzalian et al., 2008). Like other crops, safflower suffers from various diseases and insects (Weiss 2000). The most serious safflower pest in Asia and Europe is the safflower fly Acanthiophilus helianthi Rossi (Tephritidae), and sometimes known as the shoot fly or capsule fly (Talpur et al., 1995; Zandigiacomo and lob, 1991). In Asia, the safflower fly devastates most production areas in Iraq (Al-Ali et al., 1977). Pakistan (Talpur et al., 1995), and India (Vaishampayan and Kapoor, 1970; Verma et al., 1974). In Iran, seed-yield loss due to the safflower fly is estimated to be 30-70% for different safflower cultivars (Sabzalian et al., 2010). The safflower fly is a polyphagous insect belonging to the Tephritidae family (Ashri, 1971). Adult flies lay eggs on the inner side of involucral bracts of safflower green heads (Narayanan, 1961; Ashri and Knowles, 1960). Heavy infestations of safflower fly occur during the reproductive phase of the plant, and the fly prefers to lay its eggs inside developing heads throughout the flowering stage (Talpur et al., 1995). Larvae hatch from eggs, penetrate the head bracts, and feed on receptacle tissue or the whole seed (Faure et al., 2004; Jkhmola and Yadav, 1980; Narayanan, 1961; Ricci and Ciriciofolo, 1983). Larval feeding on seeds causes significant losses in seed weight, yield, and seed marketability (Ashri, 1971). The increasing impact of A. helianthi has elicited concern among entomologists who are looking for pest management options. The biology and behavior of A. helianthi has been described by some entomologists in various parts of Iraq (Al-Ali et al., 1977), Pakistan (Rahoo et al., 1997), India (Verma et al., 1974), and Egypt (Hegazi and Moursi, 1983). However, little information is available on the biology of this pest in the dry zone of Iran (Bagheri, 2007), and no information is available for

Gachsaran, Iran. The purpose of this study was to examine efficiency of methyl eugenol as attractant for *A. helianthi*.

MATERIALS AND METHODS

Field and laboratory studies were conducted to assess the efficacy of methyl eugenol on fruit flies. A trial was conducted in a safflower field (1.5 ha) at Agricultural Research Station in Gachsaran (12° 39´ N latitude and 76° 41´ longitudes) in southern Iran during November 2008 to July 2009. The experimental site was 1500 m2 situated almost in the center of the field. Laboratory trials were conducted in doors at ambient temperature (27° ± 2°C).

Trial I: Field trapping of Acanthiophilus helianthi

This study was initiated on 16th May 2008 for a period of four months. Plastic traps (10 cm × 10 cm) with circular openings measuring 2.4 cm in diameter at both ends were used for trapping the fruit-flies. This round hole trap design was adopted for they were effective in trapping fruit-flies (Ibrahim et al., 1979). A total of nine traps were placed at strategic positions in the field. Each trap was baited with a mixture of 0.5 ml methyl eugenol, 0.5 ml of Malathion EC 56 and 2 ml of Sucrose solution soaked in cotton rolls. The Traps were hung to the branches of plants at a height of 80 cm from the ground. At the chosen height, there was no effect on capture of fruit flies (Hooper and Drew, 1979). Collection of the fruit flies and recharging of the poisoned baits were made every 4th day between 6 - 7 pm. The flies were sexed and identified.

Trial II: Laboratory studies

Three different species of fruit-flies viz: A. helianthi, C. carthami and T. luteolla were reared from infested flower heads of safflower. The third instars larvae were allowed to pupate in nylon-meshed cages (82 cm X 66 cm X 66 cm) filled with infestation flower heads. The newly emerged adults were provided with water, sugar solution (10%) and protein hydrolysate. Thirty male fruit-flies of the same species which had been kept in captivity with females for ten days were tested for their response to methyl eugenol. A total of 90 male flies belonging to three different species were released in a Perspex cage (1 × 1 × 1 m). A small trap (8 cm X 12 cm) of similar shape to the field trap was used in each cage. The trap was baited with three drops of methyl eugenol, one drop of Malathion and 1 ml of sucrose solution. Recordings were made at hourly intervals for four consecutive hours on all the tested species of the fruit flies. In a further trial to eva-

Species sex	Total N	Average	catch/ trap/ month
Acanthiophilus helianthi	Male	35959	500.47
	Female	76	
Chaetrollia carthami	Male	26	0.38
	Female	2	
Trellia luteolla	Male	5	0.13
	Female	5	

Table 1. Fruit flies, three species attracted to methyl eugenol in safflower fields from 15.4.2008to 15.8.2008.

Table 2. Laboratory study showing cumulative number of fruit flies attracted to methyl eugenol

Fruit flies species	Total no	Total no of flies caught within			
	1hr.	2hr.	3hr.	4hr.	
Acanthiophilus helianthi	29	38	46	52	43.3a
Chaetrolia carthami	38	45	49	57	47.5ab
Trelli aluteola	1	3	4	5	4.2c

Means followed by the same letters are not significantly different at 5 % level as determined by Duncan's Multiple Range Test

luate the stage of adult *A. helianthi* attracted to methyl eugenol, the fruit-flies were reared using artificial diet (Tanaka et al., 1996). Twenty virgin male flies of varying ages of 4, 8, 12, 16 and 20 days were placed in separate cages containing methyl eugenol solution, malathion and sucrose solution. The number of flies caught in the traps was recorded at hourly intervals for four hours. Both laboratory trials were replicated four times using the Completely Randomized Design. The results were analyzed and the means were separated by the Duncan Multiple Range Tests.

RESULTS AND DISCUSSION

During the period of four months (May 2008 to August 2008) the total number of fruit flies caught was 36035. The dominant fruit flies were the males of A. helianthi (Table 1). The number of female flies caught was extremely low. This finding was similar to that of Steiner et al., (1965) that showed that methyl eugenol rarely attract females. Besides *A. helianthi*, the other two species caught were C. carthami and T. luteolla, though the last two species were significantly (P < 0.05) few in number.

Analysis showed that there was no significant difference in response to methyl eugenol in the laboratory between ten-day-old males of *A. helianthi* and *C. carthami* but, when compared to *T. luteolla*, their attraction to methyl eugenol was found to be significantly different (Table 2). The percentage of *A. helianthi*, *C. carthami* and *T. luteolla* caught were 43.3%, 47.5% and

4.2% respectively. The higher catch of *A. helianthi* in the field could be due to their abundance in the open, as in for the laboratory, the study showed *A. helianthi* and *C. carthami* were equally attracted to methyl eugenol. Less than 50% of the total number of flies from each species was attracted to methyl eugenol even after four hours. Dissection of the male flies showed them to have well developed testes which suggest that a considerable number of flies are indifferent to methyl eugenol. This suggestion conforms to that of Umeya et al. (1973).

The fruit-flies showed marked seasonal fluctuations with the peak periods in July and early August following boll-set). Bateman (1973) reported an increase of fruit fly population at the onset of flower heads ripening. After all the bolls had been bagged, there was a decline in the population. This could possibly be due to absence of flower heads for oviposition which subsequently resulted in a reduced fly population in the field. Observation of flower heads in the field showed that the fly oviposited as early as 15 days after flower heads set. The flies prefer to oviposit on ripe flower heads but in their absence they oviposit on green flower heads .

The placing of the traps also influences the amount of fruit-flies caught. Traps placed at the periphery of the field had higher counts than those placed in the center. This suggests that peripheral traps had better access to wind movement and methyl eugenol is capable of attracting flies at a distance of 0.6 km. (Steiner, 1952).

The attraction of *A. helianthi* to methyl eugenol is relative to the age of the flies (Table 3). The attraction was greatest when the flies were 20 days old. Since methyl eugenol is a sex attractant, it will attract flies of a

Age (days)	n					
		1hr.	2hr.	3hr.	4hr.	
4	80	4	5	5	5	% 6a
8	80	4	8	0	0	% 10ab
12	80	14	21	25	25	% 31c
16	80	38	44	47	47	% 58d
20	80	67	71	72	72	% 9c

Table 3. Laboratory study showing cumulative number of virginAcanthiophilus helianthi to methyl eugenol

Means followed by the same letters are not significantly different at 5 % level as determined by Duncan's Multiple Range Test

specific physiological age. Umeya et al. (1973) observed that male fruit flies were not attracted to methyl eugenol until the ninth day, suggesting that sexual maturity may play a prominent role. The knowledge of physiological ages of flies which are responsive to the attractant is important in a control programme where sex attractants are used.

CONCLUSION

Acanthiophilus helianthi is the main pest of safflower. The laboratory studies showed varying responses of fruit-flies to methyl eugenol. Male of *A. helianthi* and *C. carthami* were equally attracted to the sex attractant but T. luteolla was less attracted to the chemical. Evidently, with virgin males of *A. helianthi* the response to the attractant increased with the age of the flies.

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