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DETERMINATION OF CODLING MOTH, *CYDIA POMONELLA* (LEP.: TORTRICIDAE), GENERATION NUMBERS AND ADULT POPULATION FLUCTUATION IN YASOUJ THROUGH TWO DIFFERENT METHODS

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Abstract - Population fluctuation of adult codling moth was studied in Yasouj through two methods, (1) sex pheromone-baited trapping and (2) collecting of larvae and maintaining them in natural conditions, to determine its appropriate spraying time. In the first method, three generations of adults emerged from mid-April to early June (45 days), late May to mid-July (60 days) and mid-July to mid-September (60 days). Their flight peaks occurred after mid-April, after mid-June and early August. At the time of flight peak, second and third generation adult populations were equal but twice as high as first generation adult population. In the second method, three generations of adults emerged during April (25 days), early June to mid-July (45 days) and mid-July to late August (45 days). Their flight peaks occurred in mid-April, mid-June and end July. Both methods were congruent for showing population fluctuation. Flying periods of the second and third generations were equal but longer than flying period of the first generation. Flying periods in the second method were shorter than flying periods in the first method. Flight peaks of both methods coincided but with slight delay for the first method. Some of first generation larvae remained dormant for one year and emerged as adults next year which shows that some individuals were monovoltine. Spraying time for the study area can be adjusted according to the findings of this study and the type of pesticide which have to be used.

KEY WORDS: Monitoring and forecasting, Codling moth, Population fluctuation, Yasouj

Izvleček – DOLOČANJE ŠTEVILA GENERACIJ JABOLČNEGA ZAVIJAČA, *CYDIA POMONELLA* (LEP.: TORTRICIDAE) IN SPREMLJANJE SPREMINJANJA VELIKOSTI ODRASLE POPULACIJE V YASOUJU Z DVEMA METODAMA

Spreminjanje velikosti populacije odraslih jabolčnih zavijačev v Yasouju smo ugotavljali z dvema metodama, (1) lovom na feromonske pasti in (2) zbiranjem ličink in njihovim gojenjem v naravnih razmerah, da bi določili pravilen čas škropljenja. Po prvi metodi so se tri generacije odraslih osebkov pojavljale od srede aprila do začetka junija (45 dni), od konca maja do srede julija (60 dni) in od srede julija do srede septembra (60 dni). Viški letanja metuljev so bili po sredini aprila, po sredini junija in v začetku avgusta. V času viškov sta bili druga in tretja generacija enaki, a dvakrat večji kot prva generacija metuljev. Po drugi metodi so se tri generacije odraslih osebkov pojavljale aprila (25 dni), od začetka junija do srede julija (45 dni) in od srede julija do konca avgusta (45 dni). Njihovi viški letanja so bili sredi aprila, sredi junija in konec julija. Obe metodi sta bili primerni za spremljanje spreminjanja populacije. Obdobji letanja druge in tretje generacije sta bili enaki, a daljši od obdobja letanja prve generacije. Obdobja letanja po drugi metodi so bila krajša kot po prvi metodi. Viški letanja po obeh metodah so se ujemali, vendar z rahlim zaostankom po prvi metodi. Nekaj ličink prve generacije je ostalo dormantnih celo leto in so se kot metulji pojavile naslednje leto, kar kaže, da je nekaj osebkov monovoltinih. Čas škropljenja v območju raziskav lahko prilagodimo glede na ugotovitve te raziskave in vrste pesticida, ki ga uporabljamo.

KLJUČNE BESEDE: Spremljanje in napovedovanje, jabolčni zavijač, spreminjanje velikosti populacije, Yasouj

Introduction

Iran has been one of the ten top apple producers in the world with average production of 1.7 to 2.7 million tons per year (FAOSTAT, 2014). Codling moth, *Cydia pomonella* L. (Lep.: Tortricidae), is a key pest of apple in various regions of Iran (Rajabi *et al.*, 2007). Its full-grown larvae either pupate and emerge as adults after 10 to 20 days or enter diapause until the following spring. Therefore, codling moth could have different number of generations per year in different regions or even in the same region depending on weather conditions (Rajabi *et al.,* 2007; Kot, 2010). Its control is essential to harvest crop in commercial orchards and there have been many control tactics such as fruit thinning, fruit bagging, trunk banding, sanitation methods (Judd *et al*., 1997; Witzgall *et al*., 2008; Anonymous, 2014), mating disruption, trapping (Knight, 1996, Gut *et al.*, 1998, Light *et al.,* 2001) and sterile insect technique (Vreysen *et al.,* 2010; Dyke, 2010). Nevertheless, insecticides have been the main control method. Since larvae bore into fruit and they are protected from insecticide exposure, insecticide spray should be timed and targeted either at the egg stage or newly-hatched larvae depending on the insecticide used (Alston *et al.,* 2010; Ranjbar Aghdam, 2015). That is the reason why codling moth monitoring has always been one of the widely investigated aspects of this pest (Witzgall *et al*., 2008) and many monitoring systems have been devised and examined including: (1) Fermented molasses mixture (Brunner, not dated), (2) Black light traps (Madsen and Sanborn, 1962), (3) Sampling of larvae or the damage caused by larvae, (4) Female moths placed in a cage inside a trap (Hathaway, 1966), (5) kairomone-baited traps (Knight and Light, 2005; Hicher, 2009), (6) Degree day which predict codling moth development and activity based on the heat unit accumulation (Procter, *et al*., 1986; Roltsch et al., 1999; Jones *et al*., 2013) and (7) pheromone-baited traps. The detection sensitivity and species selectivity of pheromone-baited traps make them ideal tools to detect the presence of insects and monitor their flight period phenology (Brunner *et al*., 2002; Fadamiro, 2004). They have been widely used to monitor pest population density and fluctuation and determine spraying time in fruit orchards (Knight et al., 1999; Rajabi et al., 2007).

Codling moth has not been well studied in Yasouj and control measurements are mostly based on the available results from the other parts of the country. In present study, it was attempted to monitor emergence and population fluctuation of adult codling moth in this region through sex pheromone-baited traps accompanied with collecting, maintaining and rearing codling moth in natural conditions. Both methods were applied simultaneously in the same orchard and results were compared to verify the capability and reliability of sex pheromone-baited traps as a conventional method for monitoring and forecasting codling moth in this region. Usually, monitoring systems are used together to provide better information to growers to increase effectiveness of their control programs.

Materials and Methods

The study was carried out from August 2012 to September 2013 continuously in an abandoned and infested apple orchard in Darshahi village (30° 49′ 42″ N and 51° 22′ 12″ E) that is located thirty kilometers northwest of Yasouj.

A) Trapping using sex pheromone trap (First method)

Three pheromone traps were installed at a height of 1.5 to 2 meters above the ground level within tree canopy in late March 2013. Pheromone trade mark was Scentomos and traps were delta type. Traps were more than 150 meters away from each other. They were being inspected daily and replaced once every seven days. Trapped moths were being removed with a forceps and recorded in each inspection. This observation continued from late March to late September when no more moths were trapped. Finally, mean of trapped adults was calculated for every day and plotted against time to determine generation numbers, flying periods and flight peaks.

B) Collecting, maintaining and rearing larvae in natural conditions (Second method)

B-1) Large numbers of overwintering larvae were collected during late August and September 2012 by collecting maggoty apples and wrapping corrugated cardboard around the trunk of apple trees. These larvae were maintained in natural conditions in the garden and monitored on a daily basis from late March 2013 onwards to follow adult emergence. In fact this group of adult moths was the **first generation of adults** in 2013 whose flying period and peak flight were determined (Table 1).

B-2) In the next year, inspection of orchard started in mid-April on a daily basis to search for signs of codling moth infestation. From May 10th (when the first sign of codling moth infestation was seen) up to June 15th (ten days after emergence of the first adult of second generation), maggoty apples were collected daily and maintained in containers under natural conditions in the orchard. Larvae inside fruits were allowed to complete their development and pupate in corrugated cardboards. These pupae were maintained in natural conditions in the orchard and monitored on a daily basis to record emergence of the **second generation adults** (Table 1).

B-3) Adults of the second generation were being placed in a separate box and provided cotton wads of 10 percent honey solution as food and small apple fruits for egg laying. Fruits were taken out after one or two nights and maintained by date in separate containers in natural conditions. Newly-hatched larvae were allowed to bore into fruits, complete their development and pupate in corrugated cardboards. Albeit, once fruits were being withered, larvae were being transferred to a new and fresh fruit. These pupae were kept in natural conditions in the garden and monitored on a daily basis to record emergence of the **third generation adults** (Table 1).

B-4) The previous process was repeated exactly once again with the third generation adults. All larvae of this generation started to **overwinter** and no adult emerged up to the next April. Finally, the number of adult moths was plotted against time to determine population fluctuation.

Table. 1. Codling moth adult emergence during the year by the second method, with some landmark dates.

Date	Event	Adult emergence in order
Aug 2012	Collecting of larvae	
Sep 2012	Collecting of larvae	
26 Mar 2013	Emergence of first adult of 1st generation	
15 Apr 2013	1st generation flight peak	1st generation
19 Apr 2013	Emergence of last adult of 1st generation	
10 May 2013	Start to collect maggoty apples	
6 Jun 2013	Emergence of first adult of 2nd generation	
15 Jun 2013	Stop collecting maggoty apples 2nd generation flight peak	2nd generation
12 Jul 2013	Emergence of first adult of 3rd generation	
16 July 2013	Emergence of last adult of 2nd generation	3rd generation
30 Jul 2013	3rd generation flight peak	
25 Aug 2013	Emergence of last adult of 3rd generation	

Results

A) Trapping using sex pheromone trap (First method)

The first adult moth was trapped after mid April 2013. There was an intense increase in the number of trapped moths two nights after and then it decreased sharply. Whole May to early June, number of trapped moths was low and with more fluctuation. From June 10th trapped moths increased again, reached its peak on June 20th, and decreased during late June. Whole July, number of trapped moths was low and with more fluctuation. Late July trapped moths started to increase once again and reached its peak end July to early August. 8th September onwards no moth was trapped (Figure 1).

B) Rearing pest on apple fruit in natural conditions (Second method),

B-1) Of overwintering larvae which had been collected during late August and September 2012, nearby 28 percent died. The rest emerged as first generation adults from late March to late April (25 days) and their flight peak occurred in mid-April.

B-2) First brood larvae, which had been collected from infested fruits during early May to mid-June 2013, completed their development and crawled to corrugated cardboards. Of them, about five percent died and four percent remained dormant up to the next year and then emerged as adults. The rest emerged as second generation adults from early June to mid-July (45 days) and their flight peak occurred mid-June. In fact, most of adults emerged from June 5th to 25th and the rest emerged sporadically

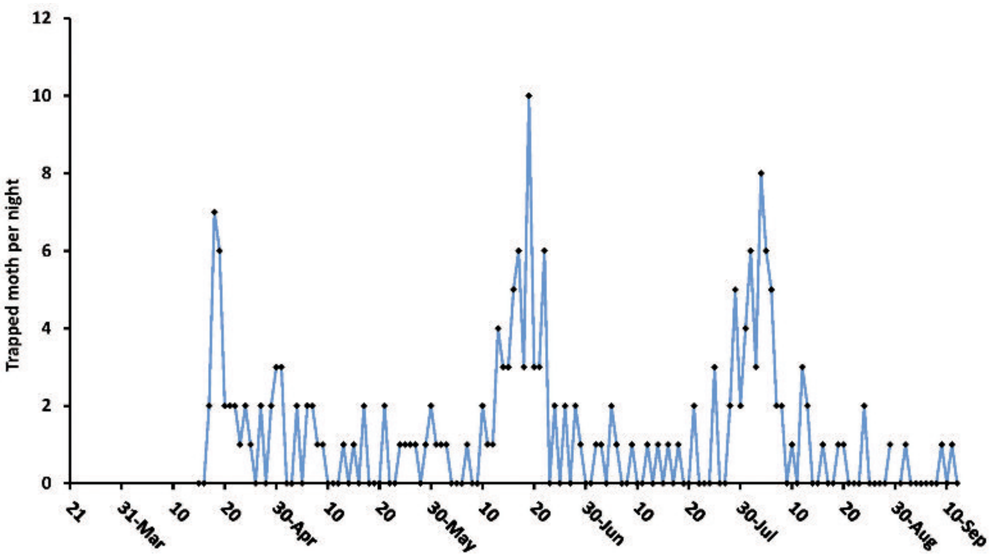

Fig. 1. Adult codling moth trapping through first method (pheromone-baited trap) in the year 2013 in Yasouj.

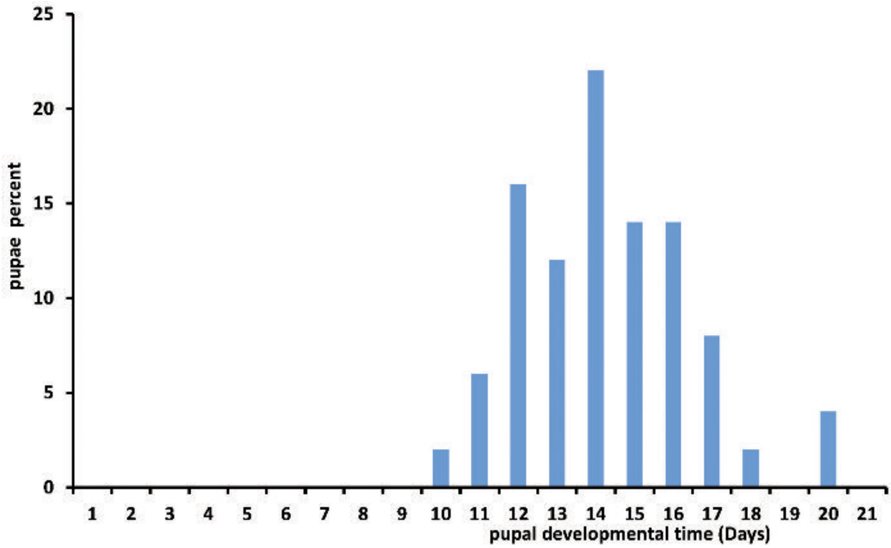

Fig. 2. Developmental time of first brood pupae of codling moth in the year 2013 in Yasouj.

during July. In this part, pupal developmental time was also measured which varied from 10 to 20 days and its mean was 14.28 days. This population showed a normal distribution for pupal developmental time in such a way that a large portion of population (25 percent) had 14 days of pupal developmental time and the rest of population was distributed normally on both sides. Pupal developmental time for 2 percent of population was 10 days and for 4 percent of population was 20 days (Figure 2).

B-3) Of second brood larvae, which were the offspring of second generation adults, about 16 percent died and the rest emerged as third generation adults during mid-July to late August (45 days). Flight peak occurred in late July (Figure 3).

B-4) Third brood larvae (offspring of third generation adults) entered diapause, started to overwinter and no adults emerged up to the next April.

Discussion

Codling moth is the most destructive insect pest of apple orchards in Kohgiluieh and Boyer Ahmad province, although no systematic survey has been carried out to estimate its damage. Chemical insecticides have dominated attempts to control this pest in the region; nevertheless, farmers are unhappy and always complaining about chemical control inefficiency which is mainly rooted in inappropriate spraying time. This pest has not been studied sufficiently in the region and control decisions mostly rely on the available results from the other parts of the country. In view of establishment of new apple orchards, codling moth could be a serious problem in this region in future. Hence, monitoring of codling moth and determination of its optimal spraying

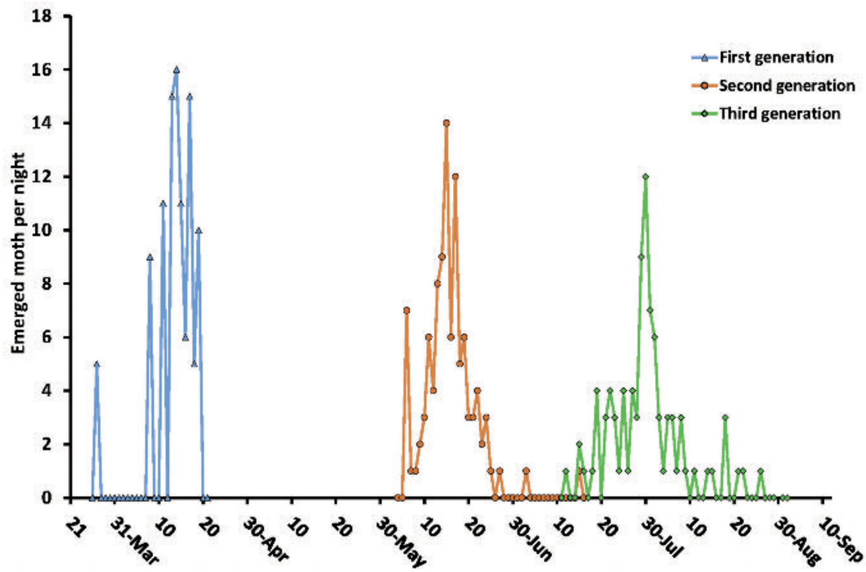

Fig. 3. Adult codling moth emergence through second method in the year 2013 in Yasouj.

time seems to be necessary. In present work, Sex pheromone-based traps accompanied with collecting, maintaining and rearing pest in natural conditions were applied to monitor adult emergence and fluctuation.

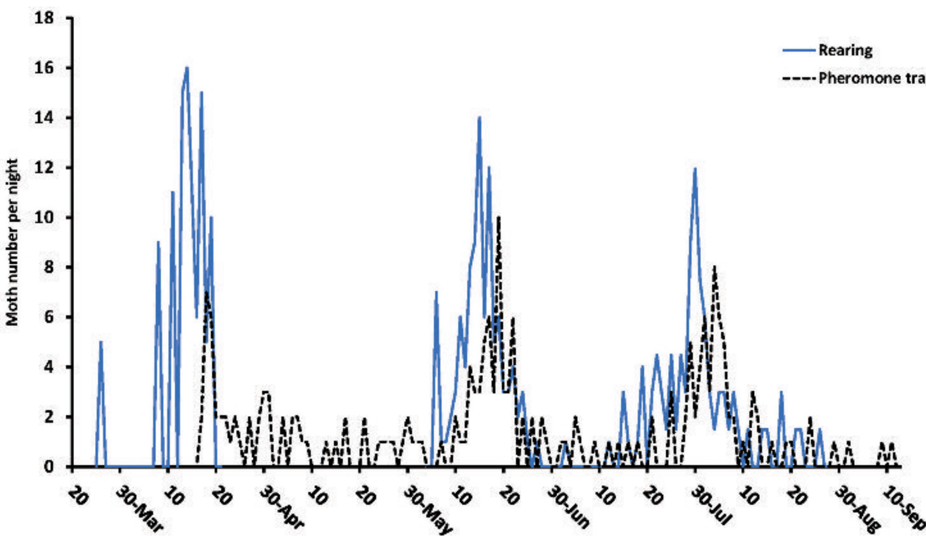

Fig. 4. Congruence of two methods in determination of population fluctuations of adult codling moth.

In both methods, results were indicative of three generations per year in this region. There are many papers, which have reported three generations per year for this pest (Rajabi *et al.,* 2007; Daneshnia *et al.,* 2012; Ranjbar Aghdam, 2015). It was seen that four percent of first generation larvae remained dormant for one year and emerged as adults in the next year. This implies that codling moth has diversity in voltinism. Rajabi (1978) reported between 8 to 12 percent of first brood larvae entered diapause. The incidence of diapause in first generation larvae indicates an influence of some other factors on diapause induction (Mansour, 2007). According to Brown *et al*. (1979), first generation larvae have a higher diapause rate (thus they are univoltine) when they are reared in apples crowded with larvae. In this work, maggoty apples were maintained together which might be the reason for this phenomenon. However, diapause induction in first generation larvae could serve as a protection mechanism for codling moth population in front of unfavorable conditions.

Flight peaks occurred during mid-April, mid-June and the end of July. Flight peaks of both methods coincided but with slight delay for the first method (Fig. 4). This slight delay in flight peak coincidence could be explained by the time interval between emergence and trapping of adults in the first method which does not exist for the second method. In second method, adult emergence and recording were at the same day while there might be a time interval between emergence and trapping (recording) in the first method. It could be concluded that the second method is more precise than first method for showing flight peaks. In the second method, adults were recorded on the same day they emerged while in the first method, trapped adults might have emerged some days back.

Flying periods of second and third generations were approximately equal but they were 15 to 20 days longer than first generation flying period (Fig. 1, 3). Emergence of first generation adults in a shorter period could be justified by source of emerging adults. Second and third generation adults are coming out of a gradual egg laying during season which will be led to a longer flying period and overlapping of generations (Table 1). While first generation adults are emerging from overwintering larvae which have been a long time (from September to April) full grown larvae. Most probably they would be uniform enough to emerge as adults in a shorter period.

Adult generations emerged during 45, 60 and 60 days in first method (Fig. 1) and 25, 45 and 45 days in second method (Fig. 3). As we can see for the same generation, flying periods in first method were 15 to 20 days longer than that in second method. This difference could be attributed to maintaining and rearing pest in an even condition in second method. In second method, place and condition of growth and development was similar for all members of each developmental stage (including eggs, larvae or pupae) to complete their embryonic, larval and pupal periods simultaneously and emerge as adults in a shorter period. While outside larvae and pupae are living in diverse places which have significant differences. For example, pupae or overwintering larvae on different sides of the same trunk experience different environmental conditions. Therefore, their developmental rate will be different which in turn leads to gradual adult emergence in nature. Normally, more gradual emerging of adults entails the longer flying period and overlapping of generations. The exact time of generation overlapping could not be determined by the first method. There was no overlapping between first and second generations in the second method, but overlapping between second and third generations occurred during second ten days of July (Table 1).

In first method, second and third generation adult populations were equal but twice as high as first generation adult population especially at the time of flight peak (Fig. 1). This could be due to mortality of overwintering larvae during winter. It was mentioned that nearby one third of overwintering larvae died in second method.

Monitoring both males and females, showing exact time of generation overlapping, right and sharp detecting of flight peaks, detecting monovoltine and bivoltine individuals and precise detecting of first adult emergence in all generations especially in first generation could be considered as advantages of second method. However, this method is more time consuming and labor-intensive.

Finally, both methods were good enough to monitor pest in order to determine its generation numbers and flight peaks which can help us to determine optimal spraying time. Spraying time could be adjusted based on population fluctuation and insecticide to be used. As a usual, insect growth regulators (e.g., Diflubenzuron and Tebufenozide) should be applied before egg laying or egg hatching. Contact and ingested insecticides (e.g., spinosad, organophosphates, pyrethroids, and carbamates) should be applied after egg hatching to kill larvae as they emerge from eggs.

Conclusion

Both methods were congruent for monitoring of flight period and population dynamics of codling moth adults. Second method was more precise but time consuming and labor-intensive. In this region, codling moth was trivoltine whose adults emerged from April to late May or early June, late May or early June to mid-July and mid-July to mid-September in order. Flight peaks occurred in mid-April, mid-June and end July or early August. About four percent of first generation larvae entered diapause until the next year, which is indicative of monovoltine individuals inside population. Finally, optimal spraying time could be anticipated based on the obtained population fluctuation and type of insecticide which have to be used.

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