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SHORT COMMUNICATION

First results on synchrony between seasonal pattern of pheromone trap captures of cotton bollworm, *Helicoverpa armigera* and appearance of freshly emerged larvae on developing cobs of corn hybrids

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Abstract Synchrony between development of five corn hybrid varieties of various seasonal growing rates (FAO numbers), seasonal flight pattern of male cotton bollworm, *Helicoverpa armigera* Hb. (Lepidoptera: Noctuidae), as monitored by pheromone traps, and the subsequent appearance of newly hatched larvae on developing cobs were studied at Mezőhegyes, South-Hungary, in 2003 and 2004. The phenological stages of corn hybrids were evaluated using the Iowa State University Scale (R_1–R_5), the flight of male moths was monitored by large capacity, funnel type of pheromone traps and the appearance of freshly hatched larvae on developing cobs were counted by visual inspection. The synchrony between the flight peak of male moths and the peak apperance of L_1 larvae on cobs was investigated by cross-correlation. In 2003 (average daily temperatures 20.7°C, average daily relative humidity 59.9% for the period of 4–18 July) "DK 391", "DK 443" and "Maraton" hybrid varieties already reached silking stage (R_1) by the time when pheromone traps indicated a peak, at 7 July (peak capture at 3-day intervals 755.5 males/traps). The number of L_1 larvae peaked on developing cobs of these varieties also at 7 July (7.0, 4.0 and 3.8 larvae/50 cobs, respectively). The synchrony between the flight peak of male moths and the peak appearance of L_1 larvae on cobs was proven (LAG = 0). A rather similar trend was observed in "Vilma" hybrid variety: it reached R_1 stage at 7 July, and L_1 larvae appeared only a few days later (11 July, 3.8 larvae/50 cobs) (LAG = −1). However, "Maxima" hybrid variety reached R_1 stage 1 week later (14 July) than the time of peak captures. Here L_1 larvae peaked as late as at 18 July (0.8 larvae/50 cobs), i.e., only after the cob had reached R_1 stage (LAG = −3). A reverse order of dates of R_1 stage and peak capture was observed in "Maxima" in 2004 (average daily temperatures; 22.5°C, average daily relative humidities 72.6% for the period of 15 July–6 August): it reached R_1 stage at 19 July, while peak trap captures were recorded at 6 August (peak capture at 3-day intervals 20.5 males/traps). L_1 larvae were found in the highest numbers on 2 August (1.5 larvae/50 cobs), practically in synchrony with peak caputres (LAG = 0). "Káma" reached R_1 stage in 16 July, and L_1 larvae peaked at 2 August (1.3 larvae/50 cobs) (LAG = 0). On the rest of the corn varieties larvae were found only in too numbers for performing statistical analysis. We conclude that in order to predict the appearance of L_1 larvae, the phenological stage of the corn variety and the seasonal flight pattern of moths, as measured by large capacity pheromone traps, should be considered in combination. If the corn variety already reached R_1 stage, L_1 larvae appear on cobs as early as the time of peak flight of moths. However, no young larvae appear on cobs despite of high trap captures, until the corn reaches the R_1 stage. These findings are discussed in the view of specifying optimal timing of a pesticide application.

Keywords *Zea mays* · *Helicoverpa armigera* · Captures · Noctuidae · Hatch of larvae · Timing of pesticide application

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Introduction

It is a key point for the success of survival of phytophagous insects that they should oviposit on their respective host plant when it reaches in course of the season the most suitable phenological stage for the development of their offspring. Basic information gained on how temporal pattern of flight activity of a lepidopterous species, regarded as pest, phenological stage of its cultivated host plant and the event of oviposition and/or period of hatching of L_1 larvae match each others serve as practical guideline on when control measures should be timed. Monitoring the flight of pests by means of pheromone trapping is a generally accepted and widely used method for this purpose (see e.g., Wall 1989). Most strikingly, however, this temporal correlation has not been reported in the literature in case of such an important insect–host plant relationship as the cotton bollworm, *Helicoverpa armigera* Hb. (Lepidoptera: Noctuidae)—corn (*Zea mays* L.) (apart from a single pilot study of Dömötör et al. 2002). To clarify this question is of special importance, because *H. armigera* larvae bore soon inside the cob of corn, so only eggs and freshly emerged larvae can be reached at sprayings by the applied pesticide. What gives even more special importance to this issue is that because of the uncertainty of optimal timing, farmers heavily use pesticides against *H. armigera*, which resulted in the appearance of pesticide-resistant (or even cross-resistant) populations (see e.g., Murray et al. 2005; Ramasubramanian and Regupathy 2004). In a laboratory experiment, resistance against Cry1Ac delta-endotoxin of *Bacillus thuringiensis* was also reported (Akhurst et al. 2003). Recently several authors emphasized the importance of optimal timing in application of pesticides (Szeőke 2001; Györök 2004; Hoffmann et al. 2004) or the egg parasitoids, *Trichogramma evenescens* Westwood and *T. pintoi* Voegele (László 2002), therefore we studied the synchrony between seasonal pattern of pheromone trap captures, phenological stage of cobs and seasonal appearance of freshly emerged as well as of elder stages of *H. armigera* larvae on developing cobs of some corn hybrids of various FAO numbers, in Hungary.

Materials and methods

Experimental site

For the site of the trials, adjacent small plots of various corn varieties in Mezőhegyes, Southeast Hungary were chosen. No pesticides were used in these plots in course of the 2 years of the experiment (2003–2004).

Corn hybrids

For the experiment, five corn varieties of various seasonal rate of growing were planted. In 2003, "DK 391" (FAO number 300), "DK 443" (FAO number 380), "Maraton" (FAO number 450), "Vilma" (FAO number 510) and "Maxima" (FAO number 580), while in 2004 the same ones with the exception that instead of "Vilma", which were not available any more, the very similar "Káma" (FAO number 510) was used.

Observation period

Observations were carried out during the second flight of *H. armigera* (July–August), when females lay eggs on developing cobs. During this period, phenological stage of the corn was evaluated, trapped moth counted and the number of freshly hatched, as well as older larvae on cobs recorded twice a week.

Phenological stages of corn

Phenological stages of the development of each corn variety were recorded during the experiment twice a week, using the Iowa State University Scale (1996), where R_1 stage means silking, R_2 stage means blistering, R_3 stage means milking, R_4 stage means doughing, R_5 stage means denting, R_6 means physiological maturity.

Monitoring seasonal flight pattern of *H. armigera* by pheromone trapping

Four sticky as well as four non-saturated funnel type of pheromone traps were used (Csalomon® RAG and VARL+) (Plant Protection Institute, Hungarian Academy of Sciences, Budapest, Hungary), both baited with the same type of rubber dispenser (Szőcs et al. 1995), loaded with 300 µg (*Z*)11-hexadecenal and 7.5 µg of (*Z*)9-hexadecenal (Nesbitt et al. 1980; Kehat et al. 1980). Traps were operated during the second flight of *H. armigera*, as during this time the developing cobs reach that phenological stage under the climate of Hungary, which makes them suitable for *H. armigera* females to oviposit on them (16 June–25 August 2003, and 11 June–30 August 2004). Sticky layers were exchanged to a new one, or collecting jars were emptied, and captured moths were recorded twice a week.

Sampling of *H. armigera* larvae in cobs in course of the season

At each sampling, 200 randomly chosen cobs (four groups, each consisting of 50 cobs) of each corn variety were inspected. From these cobs, all lepidopterous larvae were collected. Samplings were made twice a week, on the occasions of inspecting pheromone traps. Collected larvae were identified to species, and the larval stage of *H. armigera* larvae was determined, based on the width of their head capsule (Scsegolev 1951).

Meteorological data

Data of minimum-, maximum- and average daily temperatures, as well as of daily average relative humidities at Pitvaros (situated ca. 10 km from the site of the field trials), were obtained from the Institute of Hungarian Meteorological Service (Budapest).

Statistical analysis

Synchrony of seasonal flight pattern of captures of male moths in large-capacity, funnel type of pheromone traps, and appearance of freshly hatched, as well as of L_2–L_5 larvae were analyzed using the cross correlation function of Statistica StatSoft (1994) program package.

Results

Phenological stages of corn

In 2003, “DK 391”, “DK 443” and “Maraton” reached the R_1 stage at the same time (4 July) (Fig. 1). “Vilma” reached R_1 stage one recording later (7 July), while “Maxima” developed to R_1 stage at 14 July. The same trend of developments was recorded for the R_2 stage, which followed R_1 stage by 2 weeks. This time lag between varieties got smaller by the next stages, and all variety reached R_5 stage nearly at the same time. A very similar trend was recorded in 2004, with the exception that the time lag between varieties at R_1 stage did not totally diminish by R_5 stage.

Monitoring seasonal flight pattern of *H. armigera* by pheromone trapping

Captures in the funnel type of pheromone traps followed a typical flight pattern characterized by a sharp peak in both years (7 July 2003, 755.5 males/traps/3 days; 6 August 2004, 20.5 males/traps/3 days) (Fig. 1). Captures at the sticky traps indicated the same trend, however, showed no peak but rather a more-or-less steady captures during the period of mass-flight, reaching the very limited capture-capacity of the sticky surface. First captures appeared in both types of traps at the same time in 2003, while funnel traps caught the first specimens by one recording earlier in 2004 than that of the sticky type.

Sampling of *H. armigera* larvae in cobs in course of the season

In 2003, freshly hatched (L_1) larvae appeared at July 7 in “DK 391”,”DK 443” and “Maraton” hybrid varieties (7.0, 4.0 and 3.8 larvae/50 cobs, respectively) (Fig. 1). In case of these varieties L_1 larvae were highest right at the occasion when they were first detected. In “Vilma”, peak captures were recorded at R_1 stage, and L_1 larvae appeared on the subsequent occasion (11 July, 3.8 larvae/50 cobs). In “Maxima”, L_1 larvae appeared, and peaked on the subsequent inspection after R_1 stage was recorded (18 July, 0.8 larvae/50 cobs). In case of all corn hybrid varieties, the peak of L_2–L_3 larvae followed that of the peak of L_1 larvae in rather regular intervals.

In 2004, infestation level was under detection limit in case of “DK 391”,”DK 443” and “Maraton” hybrid varieties. In “Káma”, L_1 larvae appeared on 30 July. Here L_1 larvae were found in highest numbers at 2 August (1.3 larvae/50 cobs). In “Maxima”, L_1 larvae were first found at 30 July. They peaked on 2 August (1.5 larvae/50 cobs).

Synchrony of seasonal flight pattern of moths and appearance of larvae

Results of statistical analyses of investigating synchrony of seasonal flight pattern of captures of male *H. armigera* moths in funnel type of pheromone traps, and appearance of freshly hatched, as well as of L_2–L_3 larvae in various corn hybrids in 2003 and in 2004 are given in the Table 1. As for 2003, the synchrony between the flight peak of male moths as measured by funnel type of pheromone traps and the peak apperance of L_1 larvae on cobs in “DK 391”, “DK 443” and “Maraton” hybrid varieties was proven by cross-correlation (LAG = 0), while a slight shift for “Vilma” and a marked shift for “Maxima” hybrid varieties were found. In 2004, synchrony was found in case of both “Káma” and, in contrast to the previous year’s results, also in “Maxima” hybrid varieties.

Meteorological records

In course of 4–18 July 2003, the average daily temperatures varied from 17.3 to 26.0°C (average 20.7°C), maximum daily temperatures varied from 24.8 to 33.9°C (average 27.9°C), minimum daily temperatures varied from 10.1 to 20.3°C (average 13.7°C), while average

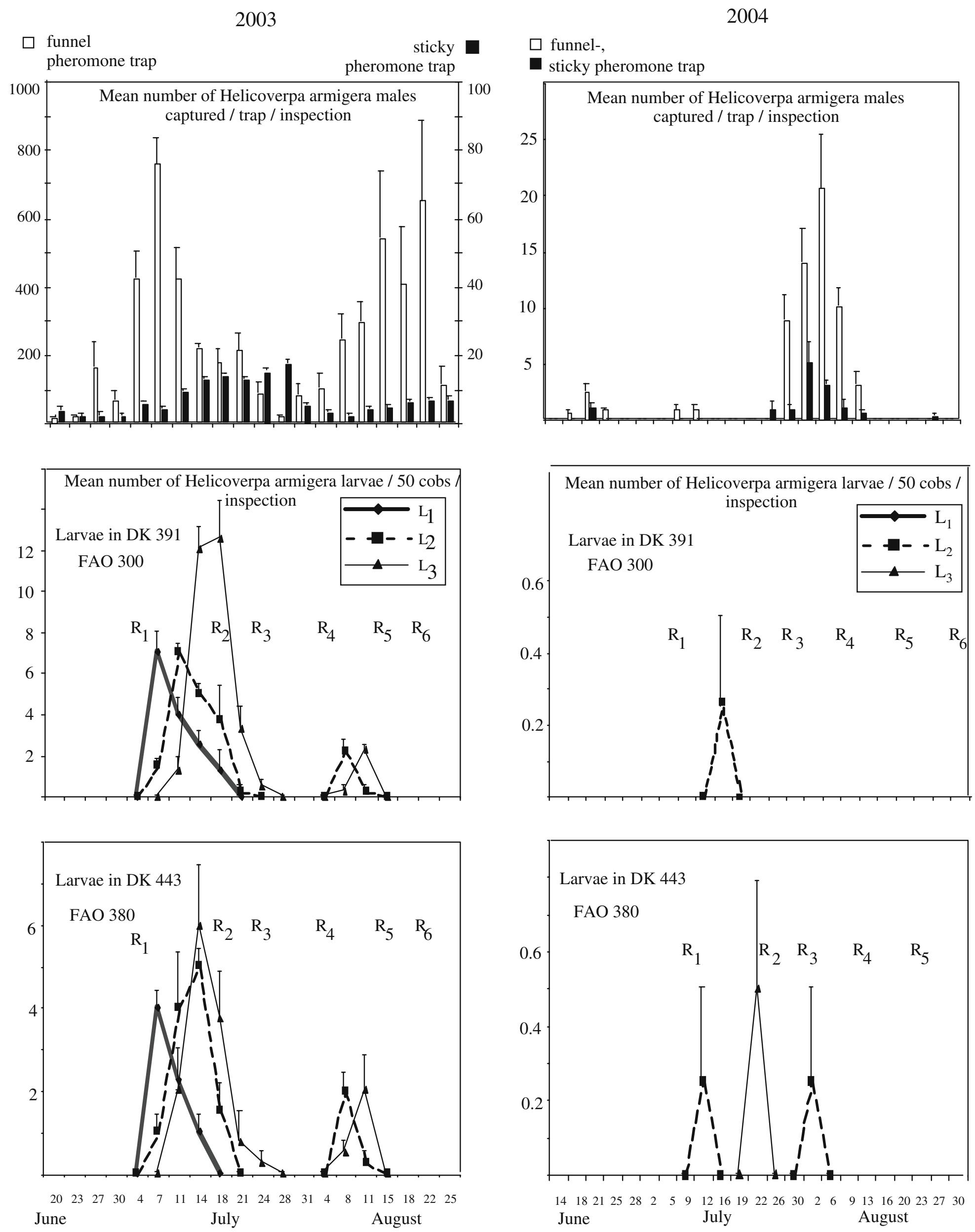

Fig. 1 Seasonal flight pattern of the second flight of *Helicoverpa armigera* in a corn field at Mezőhegyes, Hungary, in 2003 and 2004, as measured by large-capacity pheromone traps, as well as of sticky traps; and average number of L_1–L_3 larvae found in cobs of "DK 391", "DK 443", "Maraton" "Vilma", "Káma" and "Maxima" corn hybrids. Columns show the average number of males trapped between recordings (twice a week), bars denote SE (standard error). *On line charts*, *values* indicate average number of larvae found in 50 cobs at a recording, *bars* denote SE. R_1–R_6 denote phenological stage of corn

daily relative humidities ranged from 48 to 80% (average 59.9%).

For the period of 15 July–6 August 2004, the following values were measured: average daily temperatures ranged from 14.3 to 27.2°C (average 22.5°C), maximum temperatures ranged from 15.7 to 35.7°C (average 29.4°C), minimum temperatures ranged from 10.7 to 19.7°C (average 16.0°C), while relative humidities ranged from 52.0 to 96% (average 72.6%).

Discussion

We found in this study that L_1 *H. armigera* larvae appeared in developing cobs of corn at the same time when peak captures were recorded by large capacity, funnel type of pheromone traps, in case of those corn varieties which have reached R_1 phenological stage by the flight period of moths ("DK 391", "DK 443", "Maraton"). This is in a line with our earlier study

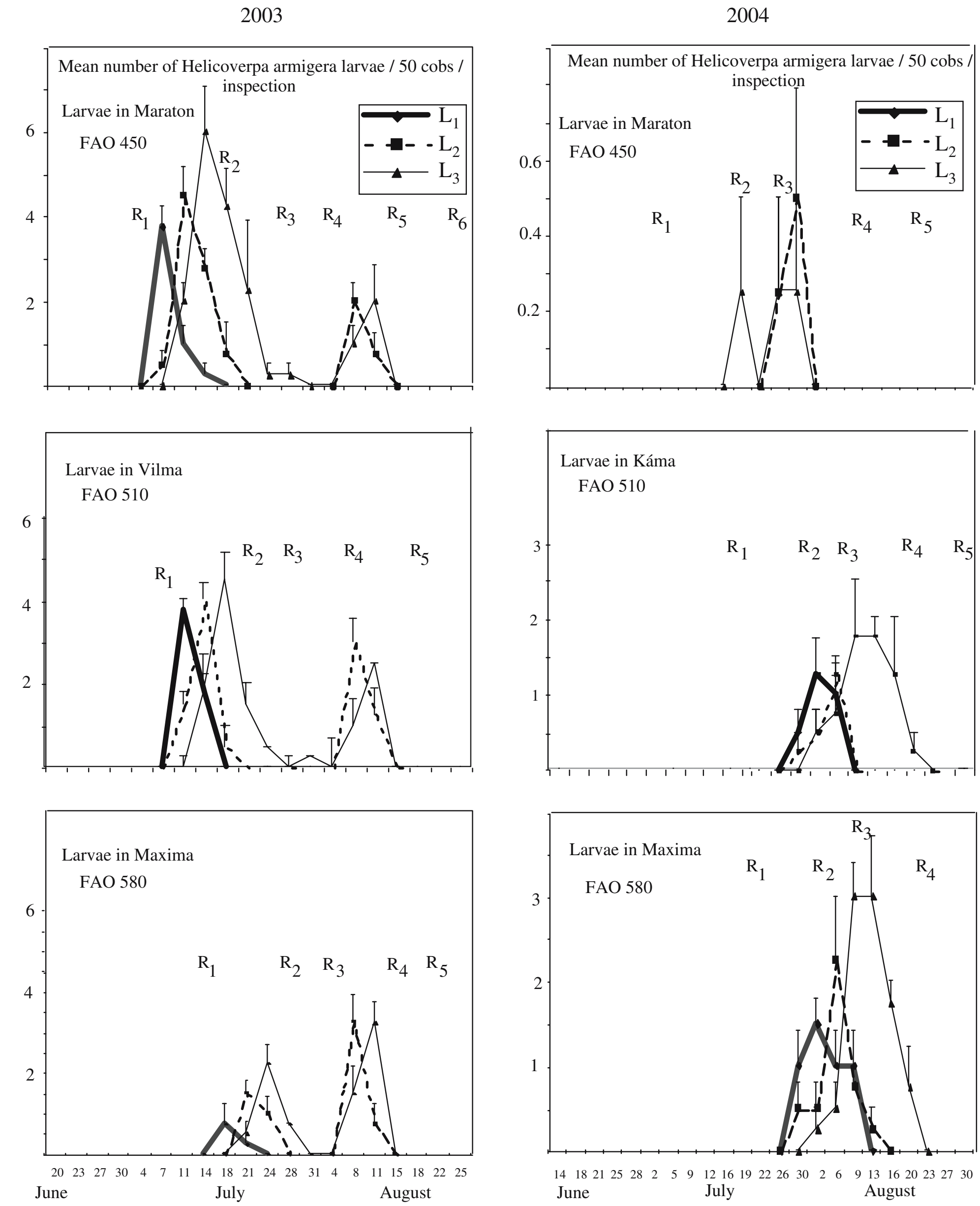

Fig. 1 continued

(Dömötör et al. 2002), when "Norma" corn variety was used at Ócsa (which is situated in the same lowland area of Hungary, as the site of the present trails). Further similarities between the present and published (Dömötör et al. 2002) trials are that corn reached the silking stage by the time peak captures of *H. armigera* was recorded (2001), and weather conditions were also very similar. Moreover, the same methodology was used that time as in the present study. Therefore the published (Dömötör et al. 2002) and present results on synchrony between trap captures and appearance of L_1 larvae, under the condition that corn reached R_1 phenological stage by the time of peak flight, corroborate each other.

Another situation was found in the present study in case of "Maxima", which has not reached R_1 stage by the flight period of moths in 2003. Here L_1 larvae appeared only when it reached R_1 stage. However, in 2004, when by the time of flight of moths "Maxima" already reached R_1 stage, L_1 larvae appeared on cobs without delay. So, it was obviously the "too early" phenological stage of the corn "Maxima" which delayed the appearance of L_1 larvae on cobs in 2003, which is apparently a consequence that *H. armigera* females did not lay eggs on these cobs until these cobs reached R_1 stage.

The flight pattern indicated by the sticky type of traps showed a roughly similar trend as that of

Table 1 Synchrony of seasonal flight pattern of captures of male *H. armigera* moths in large-capacity, funnel type of pheromone traps, and appearance of freshly hatched, as well as of L_2–L_3 larvae in various corn hybrids in 2003 and in 2004, at Mezőhegyes, South-Hungary

Year	Larval stage	Corn variety	LAG	*r*	SE	Significance ($P = 5\%$)
2003	L_1	DK 391	0	0.8424	0.2887	SIG
2003	L_2	DK 391	−1	0.8136	0.2357	SIG
2003	L_3	DK 391	−3	0.7858	0.2582	SIG
2003	L_1	DK 443	0	0.8544	0.3015	SIG
2003	L_2	DK 443	−2	0.7730	0.2425	SIG
2003	L_3	DK 443	−2	0.8247	0.2500	SIG
2003	L_1	Maraton	0	0.8503	0.3015	SIG
2003	L_2	Maraton	−1	0.7522	0.2357	SIG
2003	L_3	Maraton	−2	0.8149	0.2500	SIG
2003	L_1	Vilma	−1	0.8311	0.3333	SIG
2003	L_2	Vilma	−2	0.5505	0.2500	SIG
2003	L_3	Vilma	−3	0.6820	0.2673	SIG
2003	L_1	Maxima	−3	0.7313	0.3780	SIG
2003	L_2	Maxima	−9	0.5632	0.4082	N.S.
2003	L_3	Maxima	−10	0.5370	0.4472	N.S.
2004	L_1	Káma	0	0.8568	0.3015	SIG
2004	L_2	Káma	0	0.8832	0.3015	SIG
2004	L_3	Káma	−2	0.9051	0.3162	SIG
2004	L_1	Maxima	0	0.8933	0.2887	SIG
2004	L_2	Maxima	0	0.9151	0.2774	SIG
2004	L_3	Maxima	−2	0.9138	0.3162	SIG

measured by funnel types of traps. However, sticky type of traps caught moths in less numbers than funnel type of traps when the population density of moths was low (2004), which did not enable to make reliable picture on flight pattern. Even more importantly, sticky traps got soon saturated with captured moths when the population density became high (2003), which made the flight pattern curve arbitrarily flat during mass-flight, and therefore the flight peak could not be noticed. Because of these disadvantages, sticky traps were not found to be suitable for obtaining reliable information on the seasonal flight patern of *H. armigera*.

The appearance of freshly hatched larvae at peak flight of moths indicated that the male moth attracted to the pheromone traps were in mature stage for copulation, and females laid eggs without delay. From these eggs larvae hatched within a few days. This is in a line with the statement of Scsegolev (1951), i.e., the egg stage of *H. armigera* lasts only 3–4 days in the summer. Data on the length of larval and pupal development of *H. armigera* under various, constant temperatures under laboratory conditions, provided by Cayrol (1972), demonstrate how markedly developmental time of both larvae and pupae depend on temperature. A more recent study further underlines this, reporting that at 27°C the duration of egg stage of *H. armigera* on corn was 3 days (Liu et al. 2004). Similarly, Rothschild et al. (1982) reported that peak flight of *H. armigera*, as measured by pheromone traps, coincided with peak egg laying on cotton, in Australia. In chickpea (*Cicer arietinum* L.), however, Srivastava and Srivastava (1995) reported that the number of young *H. armigera* larvae peaked 2 weeks after the peak flight was detected by pheromone trap. Our observations that L_1 larvae hatched virtually without delay under the weather conditions of the site of the trials in Southern-Hungary, in both 2003 and 2004, indicate that these cases should be considered as expected phenomena in this region.

Our results indicated that peak captures of the second flight of *H. armigera*, as measured by large-capacity pheromone traps, coincided with the appearance of freshly hatched larvae in cobs in those corn varieties which already reached R_1 phenological stage. On those corn varieties, however, which developed slower (higher FAO number), young larvae appeared in cobs just at the time when they reached R_1 stage, provided that mass flight preceded this period. Also, both the seasonal flight pattern of moth, as measured by large-capacity pheromone traps, and phenological stage of the corn should be considered in order to specify proper timing of a pesticide application against freshly hatched larvae. When pheromone traps indicate mass-flight in a cornfield being in R_1 (or in a more advanced) phenological stage, application of pesticide measurement (if the population denstiy of the pest necessitates it) should be carried out without delay under climates of similar to, or warmer than that of Southern-Hungary's. Also our findings give a guideline to farmers how to time control measures (pesticides, or egg parasitods, as biocontrol agents), which in turn means using less pesticides with higher efficacy.

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