Research Article

Effect of artificial diet containing seeds of five corn hybrids on nutritional performance of *Helicoverpa armigera* (Lep.: Noctuidae)

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Abstract: Nutritional indices of *Helicoverpa armigera* (Hübner) larvae, an economic insect pest of agricultural crops in the world, were studied on five corn hybrids (SC700, SC704, SC500, DC370 and SC260) when incorporated into artificial diets under laboratory conditions at 25 ± 1 °C, relative humidity of 65 ± 5% and a photoperiod of 16:8 (L: D) h. The highest values of consumption index (CI) and relative growth rate (RGR) for the fourth instars were on SC500 (37.21 ± 3.34 and 0.43 ± 0.07 mg/mg/day, respectively) and lowest values of these indices were on SC704 (21.44 ± 2.83 and 0.18 ± 0.04, respectively). Among the five corn hybrids tested, efficiency of conversion of ingested food (ECI) of fifth instars was the highest on SC260 (6.92 ± 0.5%) and the highest value of efficiency of conversion of digested food (ECD) was on SC704 (10.71 ± 3%) while the larvae fed on SC700 had the lowest values of ECI and ECD (3.57 ± 0.3 and 4.39 ± 0.4%, respectively). For the sixth instar larvae, although the lowest CI and approximate digestibility (AD) values were observed on SC260 (3.49 ± 0.17 and 53.89 ± 3.70%, respectively) the highest value of ECI and ECD was on SC260 (9.11 ± 0.6 and 16.54 ± 1.5%, respectively). For all instars (fourth to sixth instars), ECD value of *H. armigera* was the highest on SC260 (10.15 ± 1.08%) and lowest on SC700 (6.32 ± 0.47%). The result of this study pointed out higher nutritional performance of the larvae *H. armigera* reared on SC704 and SC260 and lower performance on SC700 and SC500.

Keywords: Nutritional indices, *Helicoverpa armigera*, corn hybrids, feeding performance

Introduction

Helicoverpa armigera (Hübner) is one of the cosmopolitan and serious pests of different field and vegetable crops in the world (Farid, 1986; Reddy *et al*., 2004). The host plants of *H. armigera* are chickpea (Lal *et al*., 1985; Naresh and Malik, 1986; Deka *et al*., 1987), corn, cotton and ground nuts (Fitt, 1989), okra (Jallow *et al*., 2001), soybean (Naseri *et al*., 2010), bean (Rahimi Namin *et al*., 2014), tomato and many other vegetables (Shukla *et al*., 2005). The larvae of this noctuid pest can feed on different vegetative and reproductive structures of their host plants, and cause economic crop losses (Moral Garcia, 2006).

The growth, development, survivorship and reproduction of lepidopteran insects can be influenced by chemical compositions of host plants (Singh and Mullick, 1997). Food consumption and utilization by insects may be

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affected by the type of host plants on which they feed, and secondary metabolites may have an important role in creating resistance to herbivorous insects (Slansky, 1990; Wang, 1997). The rate of food consumption, digestion and conversion into body matter of an insect directly affect the physiology and behavior (Myres, 1985; Hugentobler and Renwick, 1995), adult and pupal weight, and growth and development in immature stages (Hwang *et al*., 2008). Knowledge of how different corn hybrids are digest and converted into body matter can be obtained by studies on nutritional indices of *H. armigera* larvae on these hybrids. Therefore, as a possible strategy to control *H. armigera*, assessing the nutritional performance of this insect on various host plants is necessary and may be useful.

Review of literature indicated no published information concerning the nutritional indices of *H. armigera* reared on various corn hybrids. Consequently, the goal of this study was to provide new insights on the nutritional indices of *H. armigera* larvae on five corn hybrids when incorporated into artificial diets. some researchers have studied the effects of host plants, other than corn hybrids, on nutritional indices of *H. armigera* (Ashfaq *et al*., 2003; Naseri *et al*., 2010; Soleimannejad *et al*., 2010; Hemati *et al.,* 2012; Baghery *et al.,* 2013; Rahimi Namin *et al.,* 2014) and on growth and food consumption of *Heliothis zea* (Farrar and Kennedy, 1987).

Among different techniques to control *H. armigera*, using chemical products is the main option (Kranthi *et al*., 2002), which has led to development of resistance to a number of insecticides, limiting the effective options to its management programs (Khan *et al*., 1993; Naseri *et al*., 2010). Additionally, due to the detrimental effects of the pesticides used on non-target organisms such as parasitoids and predators (Gujar *et al*., 2004; Wakil *et al*., 2009), environmental pollution, as well as secondary pests outbreak (Javed *et al*., 2009), the present research has been done either to identify alternative measures to chemical control or, at least, to reduce application of pesticides while using ecosystem friendly ways such as resistant host plants (Sharma, 2001; Sharma and Ortiz, 2002). Because corn is one of the main host plants of *H. armigera* in Iran, therefore, studying the potential resistance in seeds of various corn hybrids this pest would be useful to design strategies for successful development of the pest management in IPM programs for corn.

Materials and Methods

Corn seeds

Seeds of five corn hybrids including SC700, SC704, SC500, DC370 and SC260 were obtained from the Seed and Plant Improvement Institute, Karaj, Iran.

Rearing of *H. armigera* and experimental procedure

The neonate larvae of *H. armigera* were obtained from a laboratory culture maintained on a cowpea-based artificial diet, as described by Teakle (1991), from Tabriz University, Iran. Artificial diets contained powdered seeds of five corn hybrids (250 g), wheat germ (30 g) as protein and carbohydrate sources, sorbic acid (1.1 g) as an antimicrobial agent, ascorbic acid (3.5 g) as a vitamin source, sunflower oil (5 ml) as a preservative, agar (14 g) as a moisturizer, methyl-p-hydroxy benzoate (2.2 g), formaldehyde 37% (2.5 g) and distilled water (650 ml) (Teakle, 1991). Prepared artificial diets were kept refrigerated for no longer than two weeks before use. The insect culture was established on artificial diets and maintained in a growth chamber at 25 ± 1 °C, 65 ± 5% RH, and a photoperiod of 16:8 (L: D) h.

There were five treatment diets each containing one of the five corn hybrids. The fourth to sixth instars of *H. armigera* (20 larvae of each instar) were individually used to estimate the nutritional indices on five corn hybrids. The procedures used to calculate the indices were similar to those described by Waldbauer (1968). The larval weight, food consumed and feces produced were recorded daily. All indices were estimated using dry-tofresh-weight ratio. To calculate dry weight, the amount of daily feces produced and food remnants by twenty specimens of newly molted fourth, fifth and sixth instars were collected and weighed, then were dried at 60 °C for 48 hours and reweighed. The nutritional indices were calculated using the following formulae (Waldbauer, 1968):

Consumption Index (CI) = E/b

Relative Growth Rate (RGR) = P/Tb

Relative Consumption Rate (RCR) = E/Tb

Efficiency of Conversion of Ingested Food (ECI) = P/E

Efficiency of Conversion of Digested Food (ECD) = P/(E-F)

Approximate Digestibility (AD) = (E-F)/E

where, P = dry weight gain of insect (mg), E = dry weight of food consumed (mg), F = dry weight of feces produced (mg), b = mean dry weight of larva during the feeding period (mg), and T = feeding period (day).

Data analysis

Data from the experiment were subjected to one-way-ANOVA, and means were separated with the least significant difference (LSD) test using statistical software SPSS 16.0. Prior to applying ANOVA, all data generated from nutritional indices were first tested for normality.

Results

There were no significant differences among nutritional indices of fourth instars on the five corn hybrids except for CI and RGR. The highest values of CI (F = 5.17; df = 4, 58; P < 0.05) and RGR (F = 3.25; df = 4, 57; P < 0.05) of the fourth instars were on SC500 (37.21 ± 3.34 and 0.43 ± 0.07 mg/mg/day, respectively) and lowest were recorded on SC704 (21.44 ± 2.83 and 0.18 ± 0.04 mg/mg/day, respectively) (Table 1).

No significant difference was observed on AD of fifth instars of *H. armigera* fed on different corn hybrids. However, the values of CI (F = 3.45; df = 4, 68; P < 0.05) and RGR (F = 3.79; df = 4, 66; P < 0.05) of the fifth instars were the highest on SC500 (9.83 ± 0.79 and 0.19 ± 0.02 mg/mg/day, respectively). Furthermore, the lowest values of CI and RCR (F = 6.20; df = 4, 62; P < 0.05) were on SC704 (6.34 ± 0.93 and 3.32 ± 0.47 mg/mg/day, respectively). Among various corn hybrids, the recorded value of ECI (F = 3.92; df = 4, 67; P < 0.05) was the highest on SC260 (6.92 ± 0.5%) and the highest value of ECD (F = 3.19; df = 4, 68; P < 0.05) was obtained on SC704 (10.71 ± 3%). However, the larvae fed on SC700 had the lowest values of ECI and ECD (3.57 ± 0.3 and 4.39 ± 0.4%, respectively) (Table 2).

For the sixth instars, although the lowest CI (F = 3.63; df = 4, 51; P < 0.05) and AD (F = 6.57; df = 4, 51; P < 0.05) values were observed on SC260 (3.49 ± 0.17 and 53.89 ± 3.70, respectively), the highest value of ECI (F = 2.76; df = 4, 49; P < 0.05) and ECD (F = 3.07; df = 4, 49; P < 0.05) was on SC260 (9.11 ± 0.6 and 16.54 ± 1.5%, respectively). Moreover, the results indicated that RCR and RGR values of the sixth instar larvae were not significantly different on five corn hybrids tested (Table 3).

Recorded values of ECI and RCR for the whole larval instars (fourth to sixth instars) indicated no significant differences among the five corn hybrids tested. However, CI value (F = 4.84; df = 4, 51; P < 0.05) was the highest on SC500 (17.01 ± 1.24) and lowest on SC704 (10.71 ± 1.20). Our results showed that the highest and lowest values of AD (F = 3.62; df = 4, 51; P < 0.05) were on SC500 (83.48 ± 0.65%) and SC260 (76.00 ± 1.63%). Additionally, ECD value (F = 3.81; df = 4, 51; P < 0.05) of *H. armigera* was the highest on SC260 (10.15 ± 1.08%) and lowest on SC700 (6.32 ± 0.47%). Also, the highest RGR value of the pest (F = 2.89; df = 4, 51; P < 0.05) was observed on SC500 (0.22 ± 0.03 mg/mg/day) and SC260 (0.19 ± 0.02 mg/mg/day) and lowest one was obtained on SC704 (0.11 ± 0.02 mg/mg/day) (Table 4).

Table 1 Nutritional indices of fourth instars of *Helicoverpa armigera* fed on artificial diets prepared by five corn hybrids.

Hybrid	AD (%)	CI	RGR (mg/mg/day)	RCR (mg/mg/day)	ECI (%)	ECD (%)
SC260	92.96 ± 0.74a	31.19 ± 3.59ab	0.36 ± 0.06ab	20.65 ± 3.25a	3.53 ± 1a	3.83 ± 1.3a
SC700	92.87 ± 0.75a	24.06 ± 2.04bc	0.35 ± 0.06ab	15.94 ± 2.09a	3.70 ± 0.7a	3.99 ± 0.7a
SC500	93.98 ± 0.73a	37.21 ± 3.34a	0.43 ± 0.07a	17.24 ± 3.21a	3.17 ± 0.5a	3.41 ± 0.6a
DC370	93.11 ± 0.72a	23.80 ± 2.30bc	0.25 ± 0.04bc	11.73 ± 1.88a	3.60 ± 0.4a	3.89 ± 0.4a
SC704	90.91 ± 1.14a	21.44 ± 2.83c	0.18 ± 0.04c	15.10 ± 2.85a	2.77 ± 0.5a	3.09 ± 0.6a

Means followed by different letters in a column are significantly different (P < 0.05, LSD).
AD = Approximate digestibility, CI = Consumption index, RGR = Relative growth rate, RCR = Relative consumption rate, ECI = Efficiency of conversion of ingested food, ECD = Efficiency of conversion of digested food.

Table 2 Nutritional indices of fifth instars of *Helicoverpa armigera* fed on artificial diets prepared by five corn hybrids.

Hybrid	AD (%)	CI	RGR (mg/mg/day)	RCR (mg/mg/day)	ECI (%)	ECD (%)
SC260	79.44 ± 2.49a	7.02 ± 0.65bc	0.16 ± 0.02ab	5.38 ± 0.73b	6.92 ± 0.5a	8.82 ± 0.8a*
SC700	81.37 ± 2.61a	9.14 ± 0.82ab	0.12 ± 0.02bc	8.32 ± 0.95a	3.57 ± 0.3b	4.39 ± 0.4b
SC500	86.80 ± 1.62a	9.83 ± 0.79a	0.19 ± 0.02a	5.66 ± 0.57b	5.95 ± 0.6a	7.01 ± 0.8ab
DC370	79.09 ± 3.24a	9.46 ± 1.18ab	0.09 ± 0.02c	5.25 ± 0.78b	5.28 ± 0.7ab	6.99 ± 1ab
SC704	77.11 ± 3.28a	6.34 ± 0.93c	0.10 ± 0.02bc	3.32 ± 0.47c	6.19 ± 0.6a	10.71 ± 3a

Means followed by different letters in a column are significantly different (P < 0.05, LSD).
AD = Approximate digestibility, CI = Consumption index, RGR = Relative growth rate, RCR = Relative consumption rate, ECI = Efficiency of conversion of ingested food, ECD = Efficiency of conversion of digested food.

Table 3 Nutritional indices of sixth instar larvae of *Helicoverpa armigera* fed on artificial diets prepared by five corn hybrids.

Hybrid	AD (%)	CI	RGR (mg/mg/day)	RCR (mg/mg/day)	ECI (%)	ECD (%)
SC260	53.89 ± 3.70b	3.49 ± 0.17b	0.06 ± 0.01a	2.31 ± 0.23a	9.11 ± 0.6a	16.54 ± 1.5a
SC700	69.53 ± 2.70a	4.64 ± 0.31a	0.05 ± 0.01a	2.19 ± 0.16a	7.15 ± 0.6b	10.72 ± 1.2b
SC500	70.30 ± 2.33a	4.79 ± 0.38a	0.06 ± 0.01a	3.29 ± 0.37a	6.33 ± 0.5b	9.02 ± 0.7b
DC370	73.08 ± 3.00a	4.53 ± 0.27a	0.06 ± 0.01a	2.99 ± 0.65a	6.49 ± 0.9b	10.94 ± 2.2b
SC704	66.84 ± 2.98a	4.52 ± 0.23a	0.06 ± 0.01a	2.33 ± 0.19a	7.61 ± 0.7ab	11.78 ± 1.4b

Means followed by different letters in a column are significantly different (P < 0.05, LSD).
AD = Approximate digestibility, CI = Consumption index, RGR = Relative growth rate, RCR = Relative consumption rate, ECI = Efficiency of conversion of ingested food, ECD = Efficiency of conversion of digested food.

Table 4 Nutritional indices of whole larval instars (fourth to sixth instars) of *Helicoverpa armigera* fed on artificial diets prepared by five corn hybrids.

Hybrid	AD (%)	CI	RGR (mg/mg/day)	RCR (mg/mg/day)	ECI (%)	ECD (%)
SC260	76.00 ± 1.63e	13.96 ± 1.31b	0.19 ± 0.02a	9.55 ± 1.17a	6.35 ± 0.39a	10.15 ± 1.08a
SC700	81.06 ± 1.67c	12.78 ± 0.67bc	0.17 ± 0.02ab	9.04 ± 0.78a	4.95 ± 0.39a	6.32 ± 0.47e
SC500	83.48 ± 0.65a	17.01 ± 1.24a	0.22 ± 0.03a	8.08 ± 1.16a	5.43 ± 0.45a	6.82 ± 0.54d
DC370	81.59 ± 1.72b	11.92 ± 0.77bc	0.16 ± 0.02ab	6.30 ± 0.78a	5.75 ± 0.76a	7.39 ± 0.99c
SC704	78.72 ± 1.67d	10.71 ± 1.20c	0.11 ± 0.02b	9.90 ± 2.76a	5.66 ± 0.33a	7.86 ± 0.56b

The means followed by different letters in a column are significantly different ($P < 0.05$, LSD).
AD = Approximate digestibility, CI = Consumption index, RGR = Relative growth rate, RCR = Relative consumption rate, ECI = Efficiency of conversion of ingested food, ECD = Efficiency of conversion of digested food.

Discussion

The growth and development, survivorship, fecundity and the activity of digestive enzymes in alimentary canal of an insect can be affected by secondary biochemicals of host plants (Bernys and Chapman, 1994; Wang *et al*., 2006; Fathipour and Naseri, 2011). Nutritional requirements of insects for growth, tissue maintenance, fecundity and energy are obtained during feeding and the others are synthesized by the organisms (Chapman, 1998).

In the present study, significant differences were observed among nutritional indices, especially ECI and ECD, of *H. armigera* larvae fed on the artificial diets based on different corn hybrids. The significant differences obtained for the nutritional indices of fourth to sixth instars of *H. armigera* on five corn hybrids may probably be due to the differences in feeding performance of this pest during larval development. For the fourth and fifth instars, the highest value of CI was on SC500, indicating that the rate of food intake relative to the average weight of the larvae during the feeding time was the highest in larvae feeding on diet composed of this hybrid. However, CI values were the lowest on SC704, suggesting lower performance of food intake by the fourth and fifth instars relative to the larval weight. Another possible reason for this reduction could be because of higher larval weight on SC704 compared with the other hybrids.

Higher RGR value of fourth and fifth instars of *H. armigera* on SC500 indicated that the rate of increase in larval weight per gram body weight per day was the highest on this hybrid. It is reported that the nutritional requirements of an insect are positively correlated to body biomass and the duration of development (Phillipson, 1981; Schroeder, 1981). Therefore, lower RGR value of fifth instar on DC370 may be due to either decreased food consumption or increased duration of the larval period when the ingested food must be allocated to maintenance metabolism (Lazarevic and Peric-Mataruga, 2003).

The ECI and ECD values of fifth instars were the lowest on SC700, suggesting that a lower proportion of ingested and digested food was converted into energy, thus the consumed secondary allelochemicals had a chronic toxicity (Koul *et al*., 2004) effect on the larvae reared on this corn hybrid. Decrease in ECI value of *H. armigera* larvae may be due to the reduced efficiency to convert food consumed into growth. It is recognized that the degree of food utilization depends upon the digestibility of food and the performance that is the rate at which digested food is converted into body biomass (Batista Pereira *et al*., 2002).

Among the five different corn hybrids, despite the fact that the lowest CI and AD values of sixth instars occurred on SC260, the highest values of ECI and ECD also occurred on this hybrid. It would be suggested that because the highest larval and feces weights of *H. armigera* sixth instars were obtained on SC260, the values for CI and AD were lowest on this hybrid. Nevertheless, since higher weight gain of the larvae was on SC260, the ECI and ECD values were the highest on this hybrid compared with the others. In our study, the ECI and ECD values of fourth instars of *H. armigera* were not significantly different on the five corn hybrids, which are in disagreement with the findings of Naseri *et al*. (2010) and Soleimannejad *et al*. (2010), who noted that the ECI and ECD values of fourth instars of *H. armigera* were significantly different on various soybean varieties. Also, Baghery *et al.* (2013) reported that the ECI and ECD values in fourth instars of *H. armigera* were significantly affected by artificial diets containing seeds of different host plants. Differences in nutritional quality of host plant species used for larval feeding can be the explaination for such discrepancy. The results of the current study indicated that the mean ECI and ECD values of *H. armigera* fifth instars fed on five corn hybrids were lower than the values reported for *H. armigera* larvae reared on different soybean varieties (Naseri *et al*., 2010; Soleimannejad *et al.,* 2010) and those reared on artificial diets containing seeds of five host plants (Baghery *et al.,* 2013). Therefore, it can be concluded that the host varieties tested by above-mentioned researchers are more nutritive for growth and development of *H. armigera* fifth instar than the tested hybrids of corn in our study.

Although the highest ECD value of fifth instars of *H. armigera* was recorded on SC704 and SC260, the lowest digestive amylolytic and proteolytic activity of the fifth instars of this pest were reported on hybrid SC704 (Naseri and Razmjou, 2013). It could be suggested that the fifth instars of *H. armigera* fed on SC704 probably required low amounts of digestive enzymes to digest starch and protein contents of this hybrid. However, the results of the present research indicated that the fifth instars reared on hybrids SC700 and SC500 had lower ECI and ECD values than the other hybrids tested. Additionally, data from the whole larval instars (fourth to sixth instars) showed lowest value of ECD on SC700 as compared to other hybrids, indicating the least larval performance for the conversion of digested food to their biomass. Furthermore, studying on population growth parameters of *H. armigera* on the same five corn hybrids, Arghand *et al.* (2012) reported that the intrinsic rate of increase (r_m) of this pest was the lowest on SC700, suggesting unsuitability of this hybrid for growth, development and feeding of *H. armigera*.

Variation in the nutritional indices of *H. armigera* fed on various corn hybrids might be because of differences in nutrients needed by the pest. The unsuitability of some hybrids may be as a result of the presence of some secondary biochemicals in these hybrids, which act as antibiotic agents or absence of primary nutrients necessary for the development of *H. armigera* larvae. Improved knowledge of nutritional performance of *H. armigera* reared on different corn hybrids can lead to a better understanding in selection of partially resistant corn hybrids to *H. armigera*, which will help us in IPM of this pest.

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تأثیر غذای مصنوعی محتوی بذور پنج هیبرید ذرت بر کارایی تغذیه‌ای کرم غوزه پنبه
Helicoverpa armigera (Hübner) (Lep.: Noctuidae)

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چکیده: شاخص‌های تغذیه‌ای لاروهای کرم غوزه پنبه به‌عنوان آفت اقتصادی محصولات کشاورزی در دنیا، بر روی پنج هیبرید ذرت (SC700، SC704، SC500، DC370 و SC260) در قالب رژیم غذایی مصنوعی تحت شرایط آزمایشگاهی با دمای ۱ ± ۲۵ درجه سلسیوس، رطوبت نسبی ۵ ± ۶۵ درصد و دوره نوری ۱۶ ساعت روشنایی و ۸ ساعت تاریکی بررسی شد. بالاترین میزان شاخص مصرف (CI) و نرخ رشد نسبی (RGR) لاروهای سن چهارم روی SC500 (به‌ترتیب ۳/۳۴ ± ۳۷/۲۱ و ۰/۰۷ ± ۰/۴۳ میلی‌گرم بر میلی‌گرم بر روز) و کمترین میزان روی SC704 (به‌ترتیب ۲/۸۳ ± ۲۱/۴۴ و ۰/۰۴ ± ۰/۱۸ میلی‌گرم بر میلی‌گرم بر روز) بود. بین هیبریدهای مختلف ذرت، بیشترین بازدهی تبدیل غذای بلعیده شده (ECI) لاروهای سن پنجم روی SC260 (۰/۵± ۶/۹۲ درصد) و بیشترین بازدهی تبدیل غذای هضم شده (ECD) روی SC704 (۳ ± ۱۰/۷۱ درصد) به‌دست آمد. با این حال، لاروهای تغذیه کرده روی SC700 کمترین مقادیر ECI و ECD (به‌ترتیب ۰/۳ ± ۳/۵۷ و ۰/۴ ± ۴/۳۹ درصد) را داشتند. برای لاروهای سن ششم، با وجود اینکه کمترین میزان CI و شاخص هضم شوندگی (AD) روی SC260 (به‌ترتیب ۰/۱۷ ± ۳/۴۹ و ۳/۷۰ ± ۵۳/۸۹) بود، بالاترین میزان ECI و ECD نیز روی این هیبرید (به-ترتیب ۰/۰۶ ± ۹/۱۱ و ۱/۵ ± ۱۶/۵۴درصد) برآورد گردید. برای مجموع سنین لاروی چهارم تا ششم، شاخص ECD روی SC260 بیشترین (۱/۰۸ ± ۱۰/۱۵ درصد) و روی SC700 (۰/۴۷ ± ۶/۳۲ درصد) کمترین مقدار محاسبه شد. نتایج این تحقیق نشان داد که کارایی تغذیه‌ای لاروهای *H. armigera* روی SC704 و SC260 بیشتر و روی SC700 و SC500 کمتر از سایر هیبریدها بود.

واژگان کلیدی: شاخص‌های تغذیه‌ای، *Helicoverpa armigera*، هیبریدهای ذرت، کارایی تغذیه‌ای