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Original article Effects of different host plants on larval nutritional indices and eco-safe management tools against Agrotis ipsilon (Hufnagel) (Lepidoptera: Noctuidae)

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The current study aimed to evaluate the change in nutritional indices in the body of 4th, 5^{th} and 6^{th} instar larvae of Agrotis ipsilon as a result of feeding larvae on potato leaves compared with feeding larvae on castor bean leaves (as standard). The results indicated that the consumption index (CI) was increased in 4th, 5th and 6th instar larvae fed on potato leaves on all days except the 2nd day of 5th instar larvae compared with castor leaves. However, re lative growth rate (RGR) was decreased in 4th, 5th and 6th instar larvae fed on potato leaves on all days except the 2nd day of 4th instar larvae compared with castor leaves. In addition, approximate digestibility (AD) increased on all days of the 4th, 5th and 6th instar larvae fed on potato leaves compared with castor leaves. In general the efficiency of conversion of ingested food to biomass (ECI) was decreased in 4th, 5th, and 6th instar larvae fed on potato leaves on all days except the 2nd day of 4th, 5th& 6th instars compared with castor bean leaves. Also, the efficiency of conversion of digested food to biomass (ECD) was increased in 4th, 5th, and 6th instar larvae fed on potato leaves compared with castor bean leaves. All of these results point to the difference in food utilization leading to difference in nutritional indices. As well as, the 4th instar larvae treated with magnetic and non-magnetic Zn elements and clove oil, the results cleared the effectiveness of unmagnetized Zn element rather than the other tested materials. This means that the magnetism decreased the efficiency of Zn element and clove oil when applied to A. ipsilon larvae.

ABSTRACT

Graphical abstract



The black cutworm, *Agrotis ipsilon* (Hufnagel) is one of the most agricultural pests of crops, feed on various vegetables.



feeding larvae on potato leaves

feeding larvae on castor bean leaves (as standard).

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1. Introduction

The black cutworm, Agrotis ipsilon (Hufnagel) (Lepidoptera: Noctuidae), is a major polyphagous destructive worldwide pest. It causes a high level of economic loss to a wide range of crops through the damage of roots that consumes cotton, corn, wheat, and many vegetables [1]. The pest feeds during the early stages of plants by cutting down leaves, and also, at the base of the stems [2]. Regarding, integrated pest management, the basic problem is needed highly effective insecticides to control the black cutworm [3]. Recent research has significantly expanded our understanding of A. ipsilon's relationship with host plants. Scientists conducted comprehensive studies demonstrating that host plant selection is influenced by multiple factors: Chemical defenses in host plants, Nutritional quality of plant tissues, Physical characteristics of plant surfaces and Plant growth stages and seasonal variations [4].

Potato (Solanum tuberosum) is the most important edible and nutritious vegetable crop in Africa especially Egypt [5]. In addition, potato plants are a good source of proteins, energy, minerals, vitamins, and fats [6-8]. Moreover, potato plants have an unusual high range of utilization possibilities, so their production is more attractive [9]. The selection of host plants in this study was based on multiple scientific criteria to ensure comprehensive understanding of A. ipsilon larval development and management. The chosen plants represent major economic crops in the region that are significantly impacted by A. ipsilon infestations. The selection process considered both the geographical distribution and seasonal availability of these plants during the pest's active periods, ensuring their compatibility with local environmental conditions including temperature and humidity parameters. The selected host plants are widely cultivated in the region, making them economically significant targets for pest management strategies. Their prevalence in local farming systems ensures that any findings regarding eco-safe management tools will have practical applications for regional agricultural practices. Furthermore, this selection reflects the natural host plant diversity encountered by A. ipsilon in field conditions, providing insights into the pest's adaptability and host plant preferences under different ecological scenarios.

The level of plant nutrients and allelochemicals usually affects plant suitability and resistance to herbivores and insects Feeding behavior and development Studies by showed significant variations in Consumption rates, Food conversion efficiency and Growth rates across different host plants [10]. Many reports discussed the effects of the nutritional composition of various crops by making consumption analysis of different food sources and host preferences of FAW [11]. Chemical control strategies have been the main method for pests' control, but this strategy become less effective due to insecticide-resistant population [12]. Many essential oils with insecticidal properties have been estimated on the biological behavioral and physiological parameters of moths [13]. Among the essential oils explored, the clove oil, *Syzygium aromaticum* (Myrtaceae), which mainly consists of Eugenol, an aromatic compound that exhibits antioxidant, antibacterial, anti-inflammatory for humans; repellent, and insecticide activities for insect pests [14, 15].

Scientists proved that there is an accumulation of heavy metals in insects that feed on plants containing one, two, or all three of the heavy metals: zinc (Zn), cadmium (Cd), and copper (Cu) which are essential, such as copper and zinc [16-18]. Heavy metals in insects have a clear effect on mortality [19], growth [20], and physiology [21] Zinc and copper make a connection to the cytosol metalothionein inside the midgut of many organisms which are essential elements, but high concentrations may be toxic.

The use of a magnetic field seems to promote new tactics in pest control as a physical method [22]. Latterly, we should shed light on magnetism and the performance of electromagnetic waves on various biological aspects of insects. Mechanisms of action of magnetic field application in pest control were represented by direct physiological effects that caused cellular level impact via disruption of membrane potential and ion channels, alteration of cellular metabolism, interference with enzymatic activities and modification of protein synthesis pathways. Also, they caused behavioral modifications via navigation disruption that caused interference with magnetoreception, disorientation in movement patterns, altered feeding behavior. In addition, they caused developmental effects that concerned with growth regulation, impact on molting hormone production, changes in developmental timing and reduced pupation success rate.

This study aims to evaluate the impact of different host plants on the growth, development, and nutritional indices of *A. ipsilon* larvae and assess the effectiveness of eco-friendly management strategies, including innovative magnetic field applications.

2. Material and Methods:

- Rearing of Agrotis ipsilon:

Initial A. *ipsilon* colony was established from fieldcollected eggs. Larvae were reared under controlled laboratory conditions, Temperature: $26\pm1^{\circ}$ C, Relative humidity: $70\pm5\%$ and Photoperiod: 14:10 (L:D) hours. Colony was maintained for two generations before experimentation to eliminate field effects.

In the laboratory, the culture of *Agrotis ipsilon* was reared according to [23, 24] Larvae of *A. ipsilon* were obtained from the Plant Protection Research Institute, Agricultural Research Center. The larvae were incubated under constant conditions which were 25 ± 2 °C, 70 \pm 5% RH, and 12:12 h (L:D) photoperiod. To evade larval cannibalism, larvae were reared individually in small cups, (7.0 cm in diameter, 3.5 cm in height), and provided with sawdust to reduce moisture [25]. The larvae fed daily on fresh castor oil bean leaves, Ricinus communis L., until pupation then moths were transferred to glass jars (5L) that fed on 10% sucrose solution [27].

2.1. 1st experiment: Nutritional indices:

Used plants in nutrient:

Potato leaves that were provided to the larvae were initiated from a farm known as being free from insecticides. This farm was at New Brembal farm, Mansoura city, Dakahlia governorate, Egypt. This farm is known as free of insecticides. Host plant were carefully selected based on their local agricultural significance. Plants were cultivated under controlled greenhouse conditions (25±2°C, 65±5% RH).Fresh leaves were collected at similar growth stages (45 days after planting). Leaves were thoroughly washed with distilled water and airdried before use. Castor bean leaves (as control) were provided from castor plants next to canals and drains, as soon as castor plants are wild plants.

Experiment:

Three groups of newly molted 4th-instar larvae that had a known weight were reared in the laboratory on potato leaves. However, at the same time, larvae fed on castor bean leaves (as standard), till larvae died or pupated. After that, the fresh mass of surviving instar larvae of (4th, 5th, and 6th - instars), consumed leaves and feces in each rearing cup were reported daily. For estimating the actual loss of moisture, that was used in calculating the corrected weight of consumed leaves, the fresh leaves were kept in a similar rearing cup under the same experimental conditions.

Growth Parameters:

- Larval weight recorded every 48 hours using analytical balance (± 0.001 g).
- Larval duration monitored daily
- Survival rates calculated at each instar
- Pupal weight and duration documented

Food utilization and consumption were calculated according to the equations of [28] as follows:

- Consumption index (CI) =E/TA
- Relative growth rate (RGR) =P/EA
- Approximate digestibility $(AD) = (E-F/A) \times 100$
- Efficiency of conversion of ingested food to biomass (ECI) =
 - (RGR/CI) X 100
- Efficiency of conversion of digested food to biomass (ECD) =(P/E-F) X 100

Where:

- A = Fresh mean weight of larvae during the feeding period (mg)
- F = Fresh mass of feces (mg)
- E = Fresh weight of food ingested (mg)
- P = Fresh weight gain of larvae at the end of the feeding period (mg)
- T = Duration of the feeding period (day).

2.2. 2nd experiment: Control of A. ipsilon:

Natural products

Clove oil and Zinc elements were used in the current study. Zinc element was bought from Gomhoria Company, Mansoura, Dakahlia Governorate. However, clove oil is being prepared in the laboratory by steam distillation apparatus as described by [29, 30].

Extraction of Clove Oil:

Clove oil was extracted from the buds of dried clove, (25 g) and was used, in a steam distillation apparatus that was found in the Plant Protection Institute. Mansoura, Egypt. Furthermore, the separated oil was dried over anhydrous sodium sulfate and then stored in dark glass bottles at 4° C in the refrigerator till used. The clove oil and zinc element were tested with magnetic and non-magnetic. The magnetic flux was measured with a Magnetizing Battery apparatus which was 180 ml tesla.

Preparing the Stock Solution of the Tested Materials:

Concentrations of clove oil and zinc element were prepared based on the tested weight and volume of distilled water (w/v), in the presence of tween 80 (0.1%) as an emulsifier for clove oil. Four diluted concentrations were prepared for each material and used to draw the LC-P Lines and four replicates for each concentration.

Method of application:

Poisonous baits method:

Poisonous baits were prepared in line with [31] but with some changes by mixing 1.5 kg of wheat bran with 0.5 g molasses (that is black honey, which is used as an attractive substance) and about 0.5 liters of water. All of these constituents mix well together and are left overnight in a warm and dark place until fermentation. After that, mix 10 gm of the prepared poisonous baits with 10 ml of different concentrations of the tested materials. Later, the paste was divided equally in each treatment and was supplied to the larvae. Applications performed during early morning hours and control treatments maintained under identical conditions Observations recorded at 24, 48, and 72 hours post-treatment

Ten individuals of larvae for each replicate were used to estimate the mortality line, four concentrations and four replicates for each concentration were used. The concentrations were 500, 1000, 5000 &10000 ppm for magnetized and un-magnetized clove oil. While, the concentrations were 0.5, 1, 1.5& 2 ppm for magnetized and un-magnetized zinc elements. The percentage of mortality was recorded after one, three, five, and seven days and the data was corrected relatively to control mortality [32]. After that, LC₅₀ values were determined using the probit analysis statistical method of [33].

Equation: [34] (to determine LC₅₀ index)

- Toxicity index for LC₅₀
- $\frac{LC_{50} \text{ of the most effective compound}}{LC_{50} \text{ of the least effective compound}} x \ 100$

3. Results and Discussion 3.1. <u>1st experiment:</u>

1. Consumption index:

Data in **Table (1)** illustrated that feeding of *A. ipsilon* larvae on potato leaves increased CI at 4th instar larvae in 1st & 2nd days old (0.79& 0.43), compared with larvae fed on castor leaves as control (0.59& 0.35), respectively. Also, CI increased in 5th in 1st day old in the case of potato leaves (1.5) compared with those fed on castor bean leaves (0.55), while at 2nd day old, CI value decreased than larvae fed on castor bean leaves which were 0.44& 0.46, respectively. However, feeding of *A. ipsilon* larvae on potato leaves caused increasing Cl in 1, 2, and 3 days old of 6th instar larvae compared to those that fed on castor leaves which were (1.29, 0.43& 0.49) for potato but were (0.80, 0.34& 0.34) for castor leaves, respectively at (**Table 1**).

Increased CI of *A. ipsilon* larvae on potato leaves may be due to the presence of glycoalkaloids [34-36] illustrated that *Solanum tuberosum* L. contains glycoalkaloids – natural toxic substances present in plants of the Solanaceae family.

In insects, scientists found that poor nutrition during development leads to undersized adults; frequently, lower egg production might be predicted [37, 38]. Also, a decrease in food consumption as a result of the treatment of *S. littoralis* larvae with different allele chemicals has been detected [39].

Table (1): CI of A. *ipsilon* larvae fed as 4th instars on castor bean and potato leaves: CI (consumption index).

Instar	Age of instar (days)	Standard (Castor bean leaves)	Potato leaves
4 th	1	0.59 ± 0.01	0.79 ± 0.01
4	2	0.35 ± 0.01	0.43 ± 0.01
5 th	1	0.55 ± 0.01	1.5 ± 0.01
	2	0.46 ± 0.01	0.44 ± 0.01
6 th	1	0.80 ± 0.01	1.29 ± 0.01
	2	0.34 ± 0.01	0.43 ± 0.01
	3	0.34 ± 0.01	0.49 ± 0.01

Also, environmental factors can effect on food consumption and modify host plant interactions [40] research highlighted how environmental factors such as temperature effects that caused feeding behavior changes, development rate variations and host preference shifts. Besides humidity Impacts that caused effect on plant tissue accessibility, feeding efficiency and survival rate.

2. Relative growth rate:

Table (2) demonstrated that the RGR of cutworm larvae that fed on potato leaves was significantly decreased in 1-day-old of 4^{th} instar larvae, 1- and 2-days-old 5^{th} and the entire 6^{th} instar larvae compared with

those larvae which fed on castor bean leaves. As soon as the RGR increased in 2-day-old in 4th instar larvae.

The decrease in the RGR of *A. ipsilon* larvae on potato leaves, especially during the last-instar larvae might be due to a decrease in food consumption and food utilization efficiencies. As these results were obtained by [41].

Recent investigations have revealed complex relationships between host plant chemistry and larval development: Protein-carbohydrate balance affects larval growth rates [42]

3. Approximate digestibility:

Compared with castor bean leaves, **Table (3)** demonstrated that feeding larvae of *A. ipsilon* on potato leaves caused a significantly increase in the AD of 4th, 5th, and 6th instar larvae. Scientists found that an increase in digestion may be due to a lack of the muscle tone and longer exposure to digestive enzymes [43]. These results agreed with some results that showed a rise in AD, such as Glyphodes pyloalis 4th instar larvae treated with Thymus vulgaris and Origanum vulgare [44] and *Plutella xylustella* 3rd instar larvae treated with *O. vulgare* [45].

On the contrary, [46] showed an inhibition in AD of various insects by some botanical extracts, for instance, *Pieris rapae* larvae that were treated with methanol extract of *Silybium marianum*. Also, [47] showed a non-significant reduction of AD in all tested botanical oils (BOs), which may be due to the low percentage of excretion of consumed food by larvae because of the inhibitory effect of these tested BOs as compared to control.

Table (2): RGR of *A. ipsilon* larvae fed as 4th instars on castor bean and potato leaves:

Instar	Age of in-	Castor bean -	Potato leaves	
	star (days)	leaves (Standard)		
4 th	1	1.62 ± 0.01	1.04 ± 0.02	
	2	0.58 ± 0.01	0.63 ± 0.02	
5 th	1	1.62 ± 0.01	0.75 ± 0.3	
	2	0.48 ± 0.01	$0.40\pm\!\!0.03$	
6 th	1	1.08 ± 0.02	0.76 ± 0.03	
	2	0.53 ± 0.02	$0.49\pm\!\!0.03$	
	3	0.28 ± 0.03	0.21 ± 0.03	

Table (3): AD% of *A. ipsilon* larvae fed as 4th instars on castor bean and potato leaves:

Instar	Age of in- Castor bean leaves		Potato	
mstar	star (days)	(Standard)	leaves	
⊿ th	1	4.84 ± 0.02	8.31 ± 0.02	
4	2	5.93 ± 0.02	$16.36\pm\!\!0.02$	
5 th	1	4.36 ± 0.02	$17.93\pm\!\!0.02$	
	2	8.57 ± 0.02	10.55 ± 0.03	
	1	7.66 ± 0.03	$16.98\pm\!\!0.03$	
6 th	2	5.46 ± 0.03	$11.02\pm\!\!0.03$	
	3	9.11 ±0.03	24.01 ± 0.03	

AD :Approximate digestibility

4. Efficiency of conversion of ingested (ECI) and digested food to biomass (ECD) :

Data in **Table (4)** showed that feeding of *A. ipsilon* larvae on potato leaves decreased the in $4^{th} 5^{th}$, and 6^{th} instar larvae in 1-day-old and 6^{th} instar larvae in the 3-day- old compared with larvae fed on castor bean leaves. However, ECI increased in $4^{th} 5^{th}$, and 6^{th} instar larvae in 2-day-old.

Our results were in agreement with the results of [48] who demonstrated that the methanol extract of *Cleone arabica* seeds caused increasing in food utilization while decreasing food consumption in *S. littoralis.* Perhaps, this action occurred by the larvae as an attempt to indemnify the reduced food consumption.

Research illustrated the cause of the reduction in food utilization and mentioned that this might be due to the diversion of energy from biomass production to the induction of enzymes which are involved in the detoxification of candidate compounds [49]. Furthermore, [50] described the reduction in efficiency of food utilization that increased energetic costs arising from a reduced ability to utilize nitrogen, and this might not be necessary with absorption from the gut.

However, **Table (5)** showed that feeding of *A. ipsilon* larvae on potato leaves caused increasing in the ECD in 4th, and 5th instar larvae in 1- and 2- days old and 6th instar larvae in 2- and 3- days old comparing with castor bean leaves. However, ECD deceased in 6th instar larvae in 1- day old.

Table (4): ECI (%) of *A. ipsilon* larvae fed as 4th instars on castor bean and potato leaves

Instar	Age of instar (days)	Castor bean leaves (Standard)	Potato leaves
4 th	1	27.46 ± 0.02	14.63 ± 0.01
4	2	16.57 ± 0.02	18.31 ± 0.01
5 th	1	29.45 ± 0.02	6.25 ± 0.02
	2	10.43 ± 0.02	11.36 ± 0.02
	1	13.5 ±0.03	8.42 ± 0.02
6 th	2	15.59 ± 0.03	16.28 ± 0.03
	3	8.24 ± 0.03	7.14 ±0.03

ECI: Efficiency of conversion of ingested food to biomass.

The current results were in agreement with [51] illustrated that the reduction in ECI indicated that most food is converted into energy while less food is converted to body tissue growth. Furthermore, ECD diminished as the proportion of digested food that converted into energy increased and it exhibited a post-ingestion toxic effect, that can be considered secondary phagodeterrence responsible for the reduced RGR; the previous studies showed a reduction in ECI and ECD of some larvae of Lepidoptera which treated with different botanical extracts [52, 53].

Table (5): ECD (%) of A. ipsilon larvae fed as
4 th instars on castor bean and potato leaves

Age of instar (days)	Castor bean leaves (Standard)	Potato leaves
1	8.06 ± 0.03	10.60 ± 0.03
2	3.83 ± 0.03	15.30 ± 0.03
1	7 ± 0.03	7.89 ± 0.03
2	5.3 ± 0.04	9.06 ± 0.04
1	8.98 ± 0.04	8.67 ± 0.04
2	0.3 ± 0.04	12.07 ± 0.04
3	4.53 ± 0.04	$9.55\pm\!\!0.04$
	instar (days) 1 2 1 2 1 2 2 1 2	$\begin{array}{c c c c c c c c c c c c c c c c c c c $

ECD: Efficiency of conversion of digested food to biomass.

3.2. 2nd experiment:

Efficacy of the tested materials against 4th instar larvae of *A. ipsilon*:

Data was determined in **Table (6) and Fig. (1)** which represents that, the corrected mortality of larvae was high with non-magnetized materials compared with the magnetized ones. However, the effect of the Zn element (magnetized or not) on the mortality of larvae was more than the effect of magnetized and unmagnetized clove oil. The lowest LC_{50} was 0.917 ppm for unmagnetized Zn element followed by magnetized Zn element, un-magnetized clove oil, and magnetized clove oil that recorded 1.346, 650.135& 1396.972 ppm, respectively. However, LC_{90} was 4.295, 8.231, 72457.382& 130688.298 ppm, respectively.

The toxicity index was 100% for unmagnetized Zn element, followed by the magnetized Zn element then unmagnetized clove oil and magnetized clove oil, respectively.

The slope values indicated that unmagnetized Zn element had a higher value which was 1.911 while unmagnetized clove oil had a lower slope value which was 0.626. LC_{90}/LC_{50} value confirmed that unumagnetized Zn element had a lower value which recorded 4.684 while unumagnetized clove oil had the highest value which was 111.449. Therefore, the highest slope value or the lowest ratio of LC_{90}/LC_{50} means the steepest toxicity line.

Scientists found that the effectiveness of Zn element and other metal elements on the nutritional indices and life table of *Helicoverpa* armigera [54]. Research demonstrated that the use of clove essential oil affects the spermatogenesis and histochemistry of *S. frugiperda* ovarioles, and this is a promising option in controlling it [55]. In contrast with our results, [56] proved that magnetism improved the properties of cinnamic essential oil in controlling *S. littoralis*.

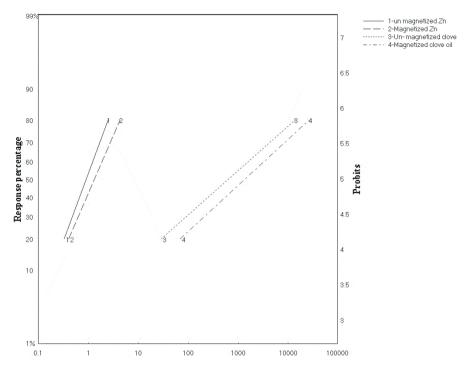


Fig. (1): LC-P line for tested materials of A. ipsilon

Table (6): Efficiency of magnetized and unmagnetized Zn element and clove oil against *Agrotis ipsilon* under laboratory conditions 27 ± 2 °C and $65 \pm 5\%$ RH.

Treatments	Conc.	Corrected mor-	LC ₅₀	LC90	Slope±	Toxicity in-	
	ppm	tality%			S.D.	dex LC50	LC50
	0.5	33.33					
Un- magnetized Zn	1	50	0.917	4.295	1.911± 0.293	100	4.684
element	1.5	60	0.917				
	2	80					
Magnetized Zn el- ement	0.5	23.33	1.346	8.231	1.629± 0.294	68.128	6.115
	1	43.33					
	1.5	53.33					
	2	60					
Un- magnetized clove oil	500	46.67	650.135	72457.382	0.626± 0.123	0.141	111.449
	1000	56.67					
	5000	66.67					
	10000	80					
Magnetized	500	36.67	1396.972	130688.298	0.650±	0.066	93.551
Magnetized	1000	50			0.124		
clove oil	5000	60					
	10000	73.33					

4. Conclusion

The study investigated ecological management strategies and nutritional ecology of *Agrotis ipsilon* (Hufnagel) (Lepidoptera: Noctuidae), a globally significant polyphagous pest. Key findings demonstrated that host plant quality significantly influenced larval development parameters and nutritional indices. Development periods ranged from 12-15 days on optimal hosts to 18-22 days on suboptimal hosts. Efficiency of conversion of ingested food (ECI) values were highest (15-18%) on primary host plants, decreasing to 8-12% on secondary hosts. Larval survival rates correlated strongly with host suitability, reaching 75-85% on primary hosts versus 45-60% on secondary hosts. Investigation of novel physical control methods revealed that unmagnetized zinc was more effective than magnetized zinc or clove oil treatments. Magnetic field exposure resulted in significant behavioral modifications, including 45% reduction in feeding activity and altered pupation patterns, suggesting potential for integration into sustainable pest management strategies. These findings provide insights for developing environmentally compatible control methods for *A. ipsilon*, offering alternatives to conventional insecticides while considering the pest's nutritional ecology.

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