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Research Article

Lethal and Sublethal Effects of Synthetic Insecticides on Feeding and Nutritional Physiology of *Spodoptera frugiperda* Larvae

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ABSTRACT

Fall armyworm, *Spodoptera frugiperda*, is a major pest of maize, and insecticide-induced effects on larval feeding and nutritional physiology are important considerations for its management. The present study evaluated the lethal and sublethal effects of chlorantraniliprole + thiamethoxam, chlorantraniliprole, flubendiamide, and spirotetramat against 3rd- and 4th-instar *S. frugiperda* larvae under laboratory conditions. Each insecticide was tested at 5, 10, and 15 ppm, corresponding to 25, 50, and 100% of the recommended dose, respectively. Larval mortality, consumption index (CI), growth rate (GR), efficiency of conversion of ingested food (ECI), efficiency of conversion of digested food (ECD), and approximate digestibility index (ADI) were assessed after 48 h. All insecticides and concentrations significantly affected larval mortality, with mortality generally increasing with concentration. Chlorantraniliprole + thiamethoxam was the most effective treatment, causing 97.96% and 88.51% mortality in 3rd- and 4th-instar larvae, respectively, at 15 ppm, whereas spirotetramat produced the lowest mortality (63.87% and 38.15%, respectively). Insecticide exposure also significantly reduced feeding and growth, with chlorantraniliprole + thiamethoxam generally producing the lowest CI and GR. ECI, ECD, and ADI were significantly lower in treated larvae than in the control, particularly following chlorantraniliprole + thiamethoxam exposure. Third-instar larvae generally exhibited greater susceptibility than 4th-instar larvae. Overall, chlorantraniliprole + thiamethoxam and chlorantraniliprole showed the greatest lethal and sublethal effects, indicating their potential for *S. frugiperda* management. Field studies are warranted to validate these laboratory findings and optimize application rates within integrated pest management programs.

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Introduction

Maize (*Zea mays* L.), a member of the Poaceae family, is an important cereal and fodder crop in Pakistan and serves as a major source of food for humans, poultry, and livestock (Waris et al., 2023). Maize cultivation has expanded considerably in recent years, covering approximately 1.418 million ha and producing 8.465 million tons (Ahmad et al., 2024). Owing to its nutritional value and importance as livestock feed, maize plays a critical role in Pakistan's agricultural economy. However, its productivity is constrained by various biotic and abiotic factors, including inadequate nutrient availability and water stress. Maize grain contains approximately 66.2% carbohydrates, 11.1% protein, 3.6% lipids, 3.6% vitamins and minerals, and 2.7% dietary fiber, with an energy value of approximately 365 kcal per 100 g (Kaushal et al., 2023). The production potential of maize is increasingly threatened by a range of biotic and abiotic stresses (Gondal et al., 2022). Among biotic constraints, insect pests and pathogens are major threats to maize production worldwide, including in Pakistan. Important insect pests include the maize stem borer, maize shoot fly, and cutworms (Kim et al., 2020; George et al., 2003). Insect infestations can occur throughout the maize growing season, with particularly severe damage during the vegetative and reproductive stages (Khan et al., 2016).

The fall armyworm (FAW), *Spodoptera frugiperda* (J.E. Smith), is an invasive lepidopteran pest that has emerged as a major threat to maize production in many parts of the world, including Pakistan (Nagoshi et al., 2019; Bhatti et al., 2021). Native to tropical and subtropical regions of the Americas, *S. frugiperda* has subsequently invaded the Canary Islands and rapidly spread across Africa, Asia, and Australia (Kenis et al., 2022). The species is highly polyphagous and has been reported to feed on more than 350 plant species belonging to approximately 76 families, including Poaceae, Asteraceae, and Fabaceae (Montezano et al., 2018; Kenis et al., 2022). Its successful invasion and establishment have been attributed to its broad host range, high fecundity, voracious feeding behavior, strong migratory capacity, rapid development, and ability to develop resistance to insecticides (Koffi et al., 2020; Wu et al., 2021).

S. frugiperda is multivoltine and completes its life cycle through the egg, six larval instars, pupa, and adult stages (IITA, 2016). Females generally deposit eggs in masses on the upper surface of leaves, with each mass commonly containing 100–200 eggs, while an individual female can lay up to approximately 1,000 eggs during her lifetime (Marri et al., 2023). Under favorable environmental conditions, its high reproductive capacity can facilitate rapid population growth and severe crop damage (Goergen et al., 2016). Larvae are the primary damaging stage, and feeding intensity increases progressively with larval development. Early instars typically scrape the leaf surface and produce characteristic window-like damage, whereas later instars consume substantial portions of the leaf tissue and may cause extensive defoliation (Bannor et al., 2022). Larvae consume a large proportion of their total food requirement during the later instars, with individual larvae reportedly responsible for approximately 90% of total foliage consumption during the larval period. Pupation generally occurs in the soil, although pupae may also be found in leaf litter. The pupal period typically lasts 8–9 days during summer but may extend to 20–30 days under cooler winter conditions. Adult longevity is generally about 10 days but may extend to approximately 21 days depending on environmental conditions (Ramzan et al., 2021). Maize is one of the principal hosts of *S. frugiperda*, and infestations have been associated with yield losses ranging from 15% to 73% (Day et al., 2017; Bannor et al., 2022).

Management of *S. frugiperda* has traditionally relied on an integrated combination of cultural practices, host-plant resistance, biological control, botanical insecticides, and synthetic insecticides (Womack et al., 2018; Tambo et al., 2020). Among these approaches, chemical insecticides remain the most widely adopted means of FAW management because of their rapid action and relatively consistent efficacy (Fatoretto et al., 2017; Jatoi and Sahito, 2023). However, insecticide exposure can produce both lethal and sublethal effects that extend beyond direct mortality, influencing insect physiology, behavior, development, fecundity, and population dynamics. For example, sublethal exposure to farnesyl acetate, methylthio-diafenthiuron, and chlorantraniliprole has been reported to delay larval and pupal development in *Plutella xylostella* and *S. frugiperda* (Guo et al., 2013; Su et al., 2020). Similarly, exposure to thiamethoxam seed treatments adversely affected larval development, adult reproduction, survival, pupation, and adult emergence of *S. frugiperda*, with effects becoming more pronounced at higher concentrations (Batuxi et al., 2020). Sublethal exposure to spinetoram has also been shown to delay development and reduce larval weight in *S. frugiperda* (Gao et al., 2021).

Although insecticide efficacy is commonly evaluated based on mortality, sublethal effects may have important implications for the population dynamics and long-term management of *S. frugiperda*. Understanding these effects is therefore essential for optimizing insecticide use and developing sustainable integrated pest management strategies. Accordingly, the present study evaluated the sublethal effects of selected synthetic insecticides on the biological and developmental parameters of *S. frugiperda* under laboratory conditions.

Materials and Methods

The experiment was conducted at the Insecticide Toxicology Laboratory, Department of Entomology, Faculty of Crop Protection, Sindh Agriculture University, Tandojam, Pakistan, in 2024.

Insect collection and rearing

Larvae of *S. frugiperda* were collected from a maize field near Tandojam and reared under controlled laboratory conditions at 27 ± 2°C and 60–70% relative humidity. Larvae of different instars were maintained separately in small plastic containers to minimize cannibalism. The larvae were provided with fresh maize leaves, which were replaced daily until pupation. The pupae were subsequently transferred to plastic containers containing sawdust and maintained until adult emergence.

Newly emerged adults were paired and maintained in plastic cages (2 ft^3) and provided with a 20% honey solution as a food source. Fresh maize leaves were also placed inside the cages as an oviposition substrate. Newly laid eggs were carefully collected using a camel-hair brush and scissors and transferred to Petri plates for hatching. Individuals of the F_1 generation were used in the subsequent bioassays.

Experimental setup and data collection

Third- and fourth-instar larvae of *S. frugiperda* were used for the bioassays. Four synthetic insecticides, Belt (flubendiamide 480 SC), Alkan (chlorantraniliprole 20 EC), Virtako (chlorantraniliprole + thiamethoxam 0.69 GR), and Movento (spirotetramat 240 SC), were evaluated alongside an untreated control (water only). Each insecticide was tested at three concentrations corresponding to 100% (recommended field dose), 50% (10 ppm), and 25% (5 ppm) of the recommended dose, depending on the respective field application rate (Table 1). The experimental design therefore comprised two factors: four insecticides and three concentrations, resulting in 12 insecticide × concentration treatment combinations, in addition to the untreated control. The required concentrations were prepared separately in 100 mL of distilled water. For each treatment, fresh maize leaves were immersed in the respective insecticide solution for 2 min and then allowed to air-dry for 30 min. The treated leaves were subsequently provided to *S. frugiperda* larvae as a food source. Appropriate untreated leaves were provided to larvae in the control treatment. Mortality and other relevant biological responses were recorded at the predetermined observation intervals.

Table 1. Details of insecticides and their concentrations used in the study

Insecticides	Recommended dose/acre	Active ingredient (%)	Insecticide required for dilution (micro liter / 100 ml)		
			15 ppm	10 ppm	5 ppm
Chlorantraniliprole 20% EC	50 ml	20	7.5	5	2.5
Spirotetramat 240SC	125 ml	240	0.63	0.42	0.21
Flubendiamide 480SC	50 ml	480	0.31	0.21	0.1
Chlorantraniliprole + Thiamethoxam 0.69GR	80 g	0.69	217.39	144.93	72.46

An analytical balance (Model ADV220) was used to determine the fresh weight of the larvae. Ten larvae were used for each insecticide concentration, with each larva placed individually in a separate plastic cup to prevent cannibalism. Maize leaves were weighed before treatment and then dipped in the respective insecticide concentrations. The treated leaves were allowed to air-dry for 30 min before being provided to the *S. frugiperda* larvae. A single larva was maintained in each cup throughout the experiment. For the control treatment, maize leaves were dipped in distilled water following the same procedure used for the insecticide treatments.

After 48 h, larval mortality, amount of food consumed, larval weight, fecal weight, and dry weight of the remaining food were recorded. The following nutritional indices were calculated according to the methods described by Waldbauer (1968):

$$\text{Consumption Index (CI)} = \frac{F}{T \times A}$$

$$\text{Growth Rate (GR)} = \frac{G}{T \times A}$$

Where F is the fresh weight of food consumed (g), G is the fresh weight gained by the larva (g), T is the feeding period (days), and A is the mean fresh weight of the larva (g) before feeding.

$$\text{Efficiency of Conversion of Ingested Food (ECI)} = \frac{G}{F} \times 100$$

$$\text{Efficiency of Conversion of Digested Food (ECD)} = \frac{G}{F - E} \times 100$$

$$\text{Approximate Digestibility (ADI)} = \frac{F - E}{F} \times 100$$

Where G is the fresh weight gained by the larva during the feeding period (g), F is the fresh weight of food consumed (g), and E is the weight of feces produced (g) during the feeding period.

Fresh weight measurements of maize leaves were used to calculate the nutritional indices. This approach was adopted following Waldbauer (1968), who indicated that either fresh or dry weights may be used for calculating digestibility indices. However, values obtained using fresh- and dry-weight measurements are not directly comparable because the proportions of dry matter in the food, feces, and insects may differ substantially.

Experimental design and statistical analysis

The experiment was arranged in a completely randomized design (CRD) comprising 12 treatment combinations derived from four insecticides and three concentrations, in addition to an untreated control in which only distilled water was applied. Each treatment was replicated 10 times. The collected data were subjected to analysis of variance (ANOVA), and treatment means were compared using the least significant difference (LSD) test. All statistical analyses were performed using STATISTIX 8.1 software.

Results

Effect of different synthetic insecticides at various concentrations on the mortality of 3rd- and 4th-instar *S. frugiperda* larvae after 48 h under laboratory conditions

Results on the efficacy of different insecticides applied at various concentrations against third-instar *S. frugiperda* larvae are presented in Figure 1. All tested insecticides significantly affected larval mortality, with significant differences observed among insecticides (F = 17.90, $p < 0.001$) and concentrations (F = 11.00, $p < 0.0001$). However, the interaction between insecticide and concentration was not significant (F = 0.67, $p > 0.05$). Larval mortality generally increased with increasing insecticide concentration.

After 48 h of exposure, at 25% (5 ppm) of the recommended dose, the highest mortality of third-instar *S. frugiperda* larvae was recorded with chlorantraniliprole + thiamethoxam (78.86 ± 6.30%), followed by chlorantraniliprole (70.18 ± 4.28%), flubendiamide (55.23 ± 5.22%), and spirotetramat (33.97 ± 1.47%). At 50% (10 ppm) of the recommended dose, chlorantraniliprole + thiamethoxam again produced the highest mortality (85.24 ± 8.72%), followed by chlorantraniliprole (79.00 ± 8.56%), flubendiamide (69.24 ± 5.29%), and spirotetramat (51.96 ± 5.93%). At the recommended dose (15 ppm), chlorantraniliprole + thiamethoxam resulted in the highest larval mortality (97.96 ± 4.03%), followed by chlorantraniliprole (87.97 ± 10.86%), flubendiamide (76.60 ± 9.20%), and spirotetramat (63.87 ± 3.22%).

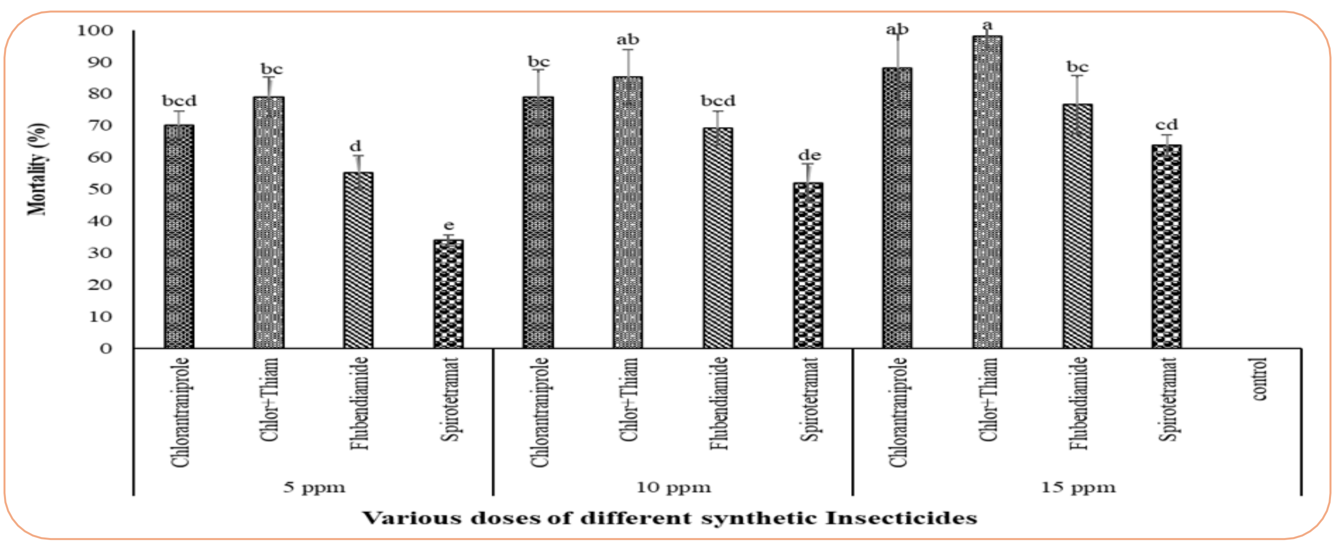

Figure 1. Impact of different synthetic insecticides at various concentrations on mortality of 4th instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 13.724, $p < 0.05$).

Figure 2 illustrates the mortality of 4th-instar *S. frugiperda* larvae following exposure to different insecticides at three concentrations for 48 h. All insecticides significantly affected larval mortality ($F = 40.68$, $p < 0.001$), and a significant effect of concentration was also observed ($F = 13.02$, $p < 0.001$). In contrast, the interaction between insecticide and concentration was not significant ($F = 0.60$, $p > 0.05$), indicating that the response to increasing insecticide concentrations did not differ significantly among the tested insecticides.

At 25% of the recommended concentration (5 ppm), the highest larval mortality was recorded for chlorantraniliprole + thiamethoxam (63.58 ± 7.74%), followed by chlorantraniliprole (55.02 ± 5.23%), flubendiamide (45.02 ± 2.81%), and spirotetramat (29.54 ± 1.68%). At 50% of the recommended concentration (10 ppm), chlorantraniliprole + thiamethoxam produced the highest mortality (75.39 ± 6.81%), followed by chlorantraniliprole (64.78 ± 2.98%), flubendiamide (59.55 ± 4.63%), and spirotetramat (33.61 ± 2.55%). At the full recommended concentration (100%; 15 ppm), mortality was highest with chlorantraniliprole + thiamethoxam (88.51 ± 4.93%), followed by chlorantraniliprole (73.01 ± 2.71%), flubendiamide (62.86 ± 6.46%), and spirotetramat (38.15 ± 4.95%). Overall, chlorantraniliprole + thiamethoxam exhibited the greatest larvicidal activity at all tested concentrations, whereas spirotetramat consistently produced the lowest mortality.

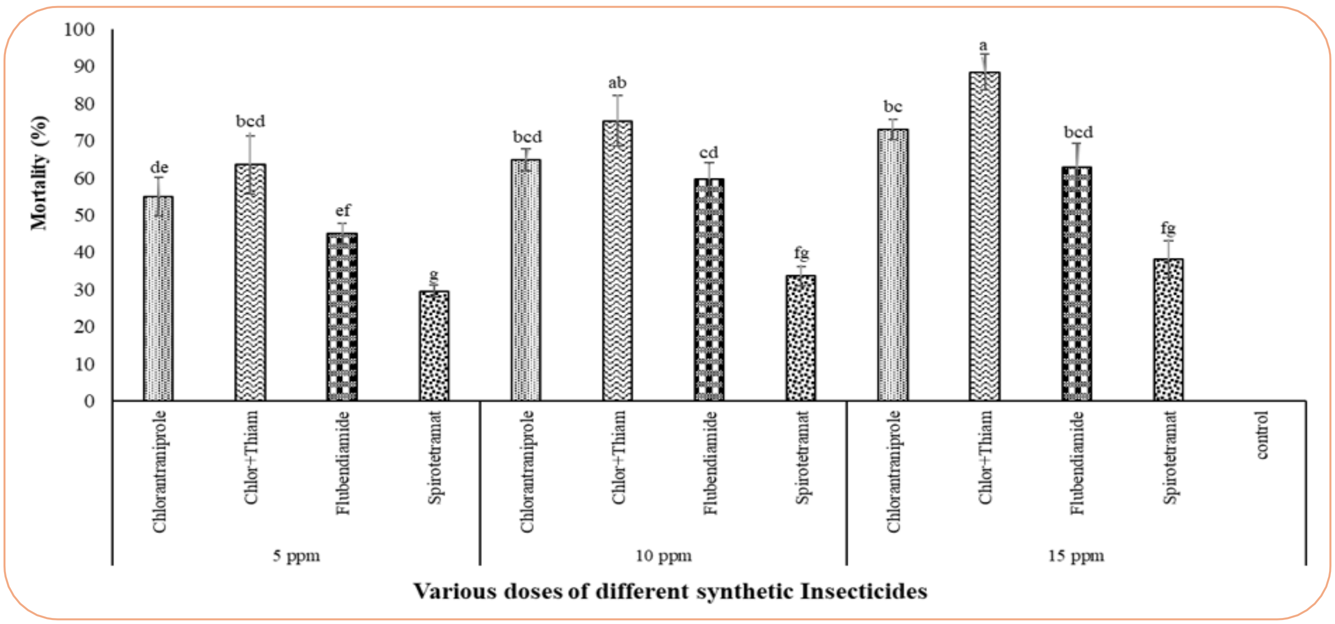

Figure 2. Impact of different synthetic insecticides at various concentrations on mortality of 3rd instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 18.911, $p < 0.05$).

Effect of different synthetic insecticides at various concentrations on the digestibility indices of 3rd-instar *S. frugiperda* larvae after 48 hours of application under laboratory conditions

Figure 3 presents the effects of different concentrations of synthetic insecticides on the consumption index (CI) of 3rd-instar *S. frugiperda* larvae 48 h after treatment. Significant differences were observed among the insecticides ($F = 10.94$, $p < 0.001$) and their concentrations ($F = 10.02$, $p < 0.001$), whereas the interaction between insecticides and concentrations was not significant ($F = 0.29$, $p > 0.05$). The highest CI was recorded in the control group (2.502 ± 0.21 mg), indicating the greatest food consumption in the absence of insecticide exposure.

Among the insecticide treatments, the CI generally decreased with increasing insecticide concentration. At 15 ppm, the lowest CI values were recorded for chlorantraniliprole + thiamethoxam (0.985 ± 0.04 mg) and chlorantraniliprole (1.208 ± 0.05 mg), followed by flubendiamide (1.476 ± 0.12 mg) and spirotetramat (1.552 ± 0.09 mg). At 10 ppm, the highest CI was observed with spirotetramat (1.841 ± 0.23 mg), followed by flubendiamide (1.611 ± 0.16 mg), chlorantraniliprole (1.514 ± 0.03 mg), and chlorantraniliprole + thiamethoxam (1.179 ± 0.07 mg). At 5 ppm, spirotetramat resulted in the highest CI (2.098 ± 0.10

mg), followed by flubendiamide (1.728 ± 0.09 mg), chlorantraniliprole (1.635 ± 0.13 mg), and chlorantraniliprole + thiamethoxam (1.478 ± 0.24 mg).

The reduced CI observed in the treated larvae, particularly at higher insecticide concentrations, indicated a marked reduction in food consumption following insecticide exposure. The pronounced reduction associated with chlorantraniliprole + thiamethoxam and chlorantraniliprole was consistent with the higher larval mortality observed in these treatments.

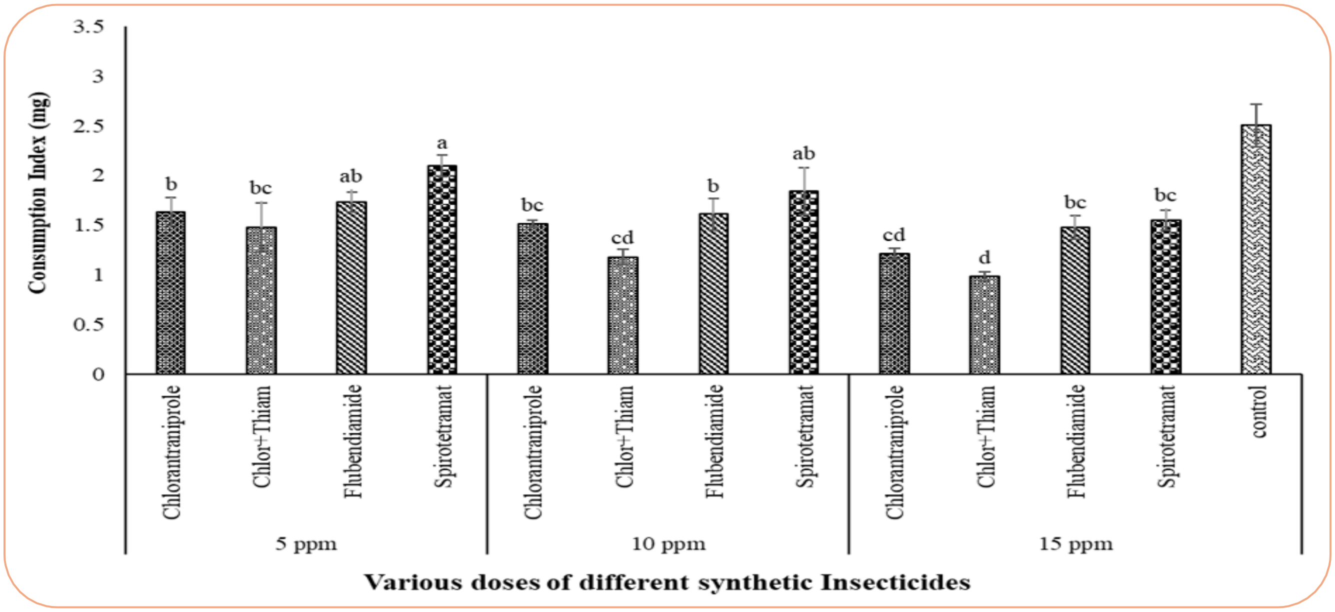

Figure 3. Impact of different synthetic insecticides at various concentrations on CI of 3rd instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 0.3860, $p < 0.05$).

Due to reduced feeding by the 3rd-instar larvae of *S. frugiperda* after 48 h of insecticide application, a relatively lower growth rate (GR) was observed in larvae exposed to different concentrations of the synthetic insecticides (Figure 4). The effects of insecticides, concentrations, and their interaction on larval GR were significant (insecticide: $F = 29.21$, $p < 0.001$; concentration: $F = 39.01$, $p < 0.001$; insecticide × concentration: $F = 3.24$, $p < 0.05$). The highest GR was recorded in the untreated control (0.345 ± 0.01 g). At 5 ppm, Spirotetramat resulted in the highest GR (0.285 ± 0.02 g), followed by Flubendiamide (0.131 ± 0.007 g), Chlorantraniliprole (0.118 ± 0.005 g), and Chlorantraniliprole + Thiamethoxam (0.092 ± 0.008 g). At 10 ppm, Spirotetramat again produced the highest GR (0.181 ± 0.01 g), followed by Flubendiamide (0.137 ± 0.01 g), Chlorantraniliprole (0.137 ± 0.01 g), and Chlorantraniliprole + Thiamethoxam (0.120 ± 0.01 g). At 15 ppm, Spirotetramat recorded the highest GR (0.285 ± 0.02 g), followed by Flubendiamide (0.212 ± 0.01 g), Chlorantraniliprole (0.208 ± 0.008 g), and Chlorantraniliprole + Thiamethoxam (0.120 ± 0.01 g).

Figure 5 presents the effects of different insecticides and their concentrations on the efficiency of conversion of ingested food (ECI) in 3rd-instar *S. frugiperda* larvae 48 h after treatment. A highly significant difference was observed among the insecticides ($F = 15.28$, $p < 0.001$), whereas neither insecticide concentration ($F = 2.99$, $p > 0.05$) nor the interaction between insecticide and concentration ($F = 0.67$, $p > 0.05$) had a significant effect on ECI. The highest ECI was recorded in the control (13.41 ± 1.26%). Among the insecticide treatments, at 5 ppm, the highest ECI was observed with spirotetramat (12.485 ± 0.39%), followed by flubendiamide (10.144 ± 0.68%), chlorantraniliprole (9.136 ± 1.14%), and chlorantraniliprole + thiamethoxam (8.951 ± 1.14%). At 10 ppm, spirotetramat again resulted in the highest ECI (11.870 ± 0.84%), followed by flubendiamide (10.167 ± 0.55%), chlorantraniliprole (9.205 ± 0.51%), and chlorantraniliprole + thiamethoxam (7.466 ± 0.87%). Similarly, at 15 ppm, the highest ECI was recorded with spirotetramat (10.296 ± 0.26%), followed by flubendiamide (9.885 ± 0.50%), chlorantraniliprole (8.821 ± 0.29%), and chlorantraniliprole + thiamethoxam

(6.811 ± 0.64%). Overall, ECI tended to decline with increasing insecticide concentration, with the lowest values generally observed at 15 ppm, particularly under the chlorantraniliprole + thiamethoxam treatment. These findings indicate that insecticide type had a significant influence on the efficiency with which ingested food was converted into larval biomass, whereas the effects of concentration and the insecticide × concentration interaction were statistically non-significant.

Figure 6 illustrates the effects of different concentrations of synthetic insecticides on the Efficiency of Conversion of Digested Food (ECD) of 3rd-instar *S. frugiperda* larvae 48 h after treatment. A significant difference was observed among the insecticides (F = 3.00, $p < 0.05$), whereas the effects of insecticide concentration (F = 1.92, $p > 0.05$) and the interaction between insecticide and concentration (F = 0.06, $p > 0.05$) were non-significant. The highest ECD was recorded in the control (19.446 ± 2.84%). Among the insecticide treatments, the highest ECD at 5 ppm was recorded for spirotetramat (16.488 ± 1.14%), followed by flubendiamide (15.317 ± 0.64%), chlorantraniliprole (13.986 ± 1.04%), and chlorantraniliprole + thiamethoxam (13.361 ± 1.17%). At 10 ppm, spirotetramat again exhibited the highest ECD (15.173 ± 1.41%), followed by flubendiamide (14.779 ± 0.89%), chlorantraniliprole (13.268 ± 1.33%), and chlorantraniliprole + thiamethoxam (12.707 ± 0.85%). Similarly, at 15 ppm, the highest ECD was recorded for spirotetramat (14.181 ± 2.44%), followed by flubendiamide (13.677 ± 0.71%), chlorantraniliprole (12.965 ± 0.72%), and chlorantraniliprole + thiamethoxam (11.610 ± 1.04%). Overall, ECD was lower in insecticide-treated larvae than in the control, indicating reduced efficiency of nutrient conversion following insecticide exposure.

Figure 7 presents the Approximate Digestibility Index (ADI) of 3rd-instar *S. frugiperda* larvae exposed to different concentrations of the tested insecticides for 48 h. Among the synthetic insecticides, a significant effect was observed for insecticide type ($F = 5.56$, $p < 0.05$) and concentration ($F = 3.58$, $p < 0.05$), whereas their interaction was not significant ($F = 0.11$, $p > 0.05$). The highest ADI was recorded in the control treatment (79.279 ± 5.59%). Among the insecticide treatments, the highest ADI at 5 ppm was observed with spirotetramat (71.486 ± 6.87%), followed by flubendiamide (65.052 ± 2.98%), chlorantraniliprole (62.909 ± 3.08%), and chlorantraniliprole + thiamethoxam (52.397 ± 4.01%). At 10 ppm, spirotetramat again produced the highest ADI (67.593 ± 6.05%), followed by flubendiamide (59.338 ± 3.33%), chlorantraniliprole (56.080 ± 6.10%), and chlorantraniliprole + thiamethoxam (48.604 ± 8.54%). At 15 ppm, the highest ADI was recorded for spirotetramat (63.127 ± 8.88%), followed by flubendiamide (53.769 ± 3.98%), chlorantraniliprole (46.660 ± 4.97%), and chlorantraniliprole + thiamethoxam (44.087 ± 7.16%). Overall, ADI declined progressively with increasing insecticide concentration, with spirotetramat consistently exhibiting the highest ADI among the tested insecticides.

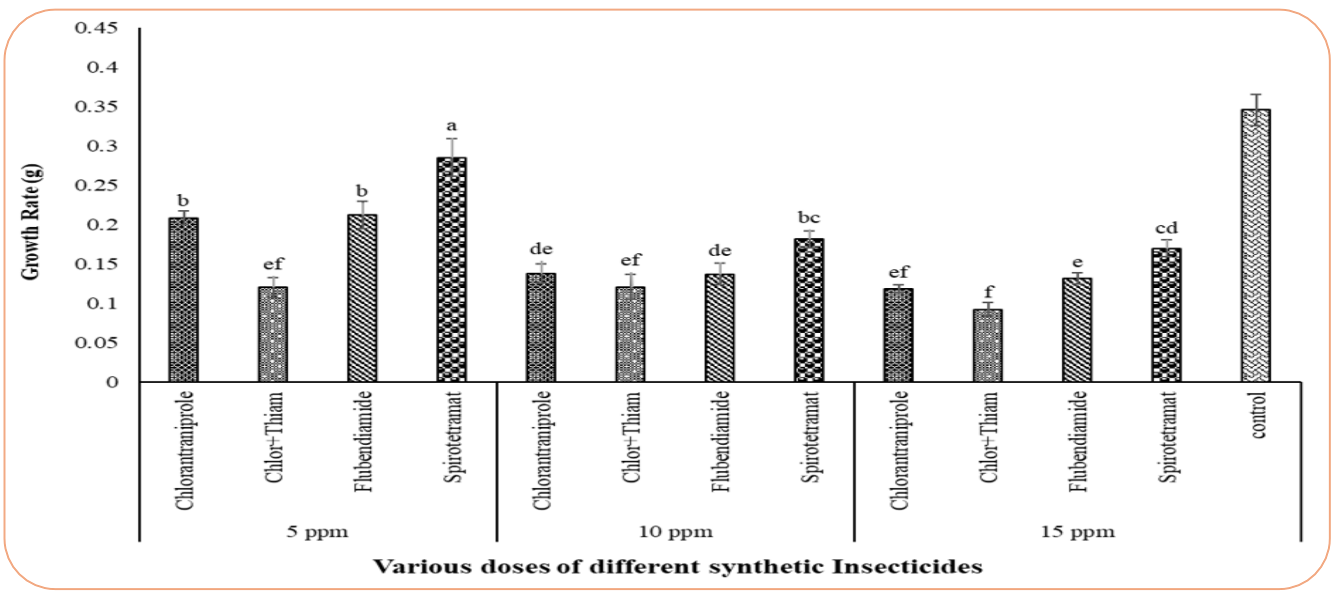

Figure 4. Impact of different synthetic insecticides at various concentrations on the growth rate of 3rd instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 0.0378, $p < 0.05$).

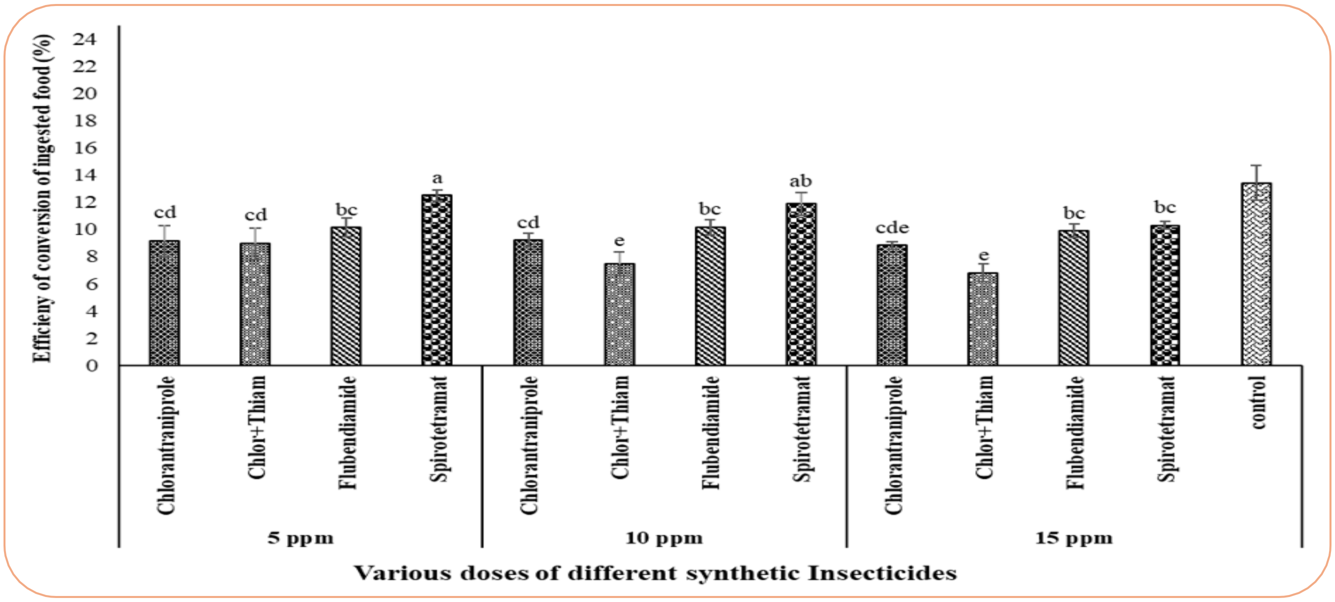

Figure 5. Impact of different synthetic insecticides at various concentrations on the efficiency of conversion of ingested food by 3rd instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 2.0273, $p < 0.05$).

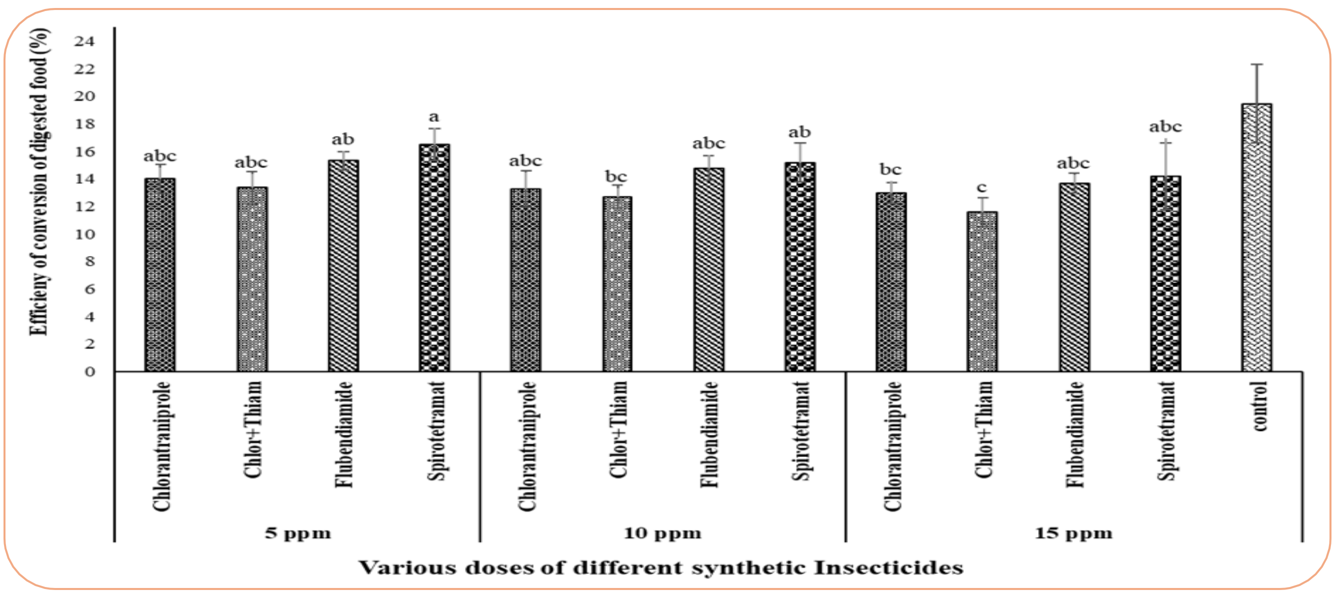

Figure 6. Impact of different synthetic insecticides at various concentrations on the efficiency of conversion of digested food by 3rd instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 3.4451, $p < 0.05$).

Effect of different synthetic insecticides at various concentrations on the digestibility indices of 4th instar *S. frugiperda* larvae after 48 h of application under laboratory conditions

Figure 8 presents the effects of different concentrations of synthetic insecticides on the consumption index (CI) of 4th-instar *S. frugiperda* larvae 48 h after treatment. The analysis revealed highly significant differences among insecticides ($F = 8.76$, $p < 0.001$) and among insecticide concentrations ($F = 14.61$, $p < 0.001$), whereas the interaction between insecticides and concentrations was not significant ($F = 0.42$, $p > 0.05$). The highest CI was recorded in the untreated control (2.780 ± 0.18 mg), indicating the greatest food consumption by larvae in the absence of insecticide exposure.

At 5 ppm, Spirotetramat and Flubendiamide resulted in the highest CI values (2.331 ± 0.15 and 1.927 ± 0.26 mg, respectively), followed by Chlorantraniliprole (1.809 ± 0.07 mg) and Chlorantraniliprole + Thiamethoxam (1.753 ± 0.18 mg). At 10 ppm, the lowest CI was recorded for Chlorantraniliprole + Thiamethoxam (1.232 ± 0.05 mg), followed by Chlorantraniliprole (1.567 ± 0.10 mg), Flubendiamide (1.715 ± 0.18 mg), and Spirotetramat (1.901 ± 0.24 mg). At 15 ppm, Spirotetramat and Flubendiamide recorded the highest CI values (1.614 ± 0.06 and 1.565 ± 0.17 mg, respectively), followed by Chlorantraniliprole (1.307 ± 0.08 mg) and Chlorantraniliprole + Thiamethoxam (1.006 ± 0.03 mg). Overall, the CI generally declined with increasing insecticide concentration, indicating reduced food consumption by *S. frugiperda* larvae following insecticide exposure.

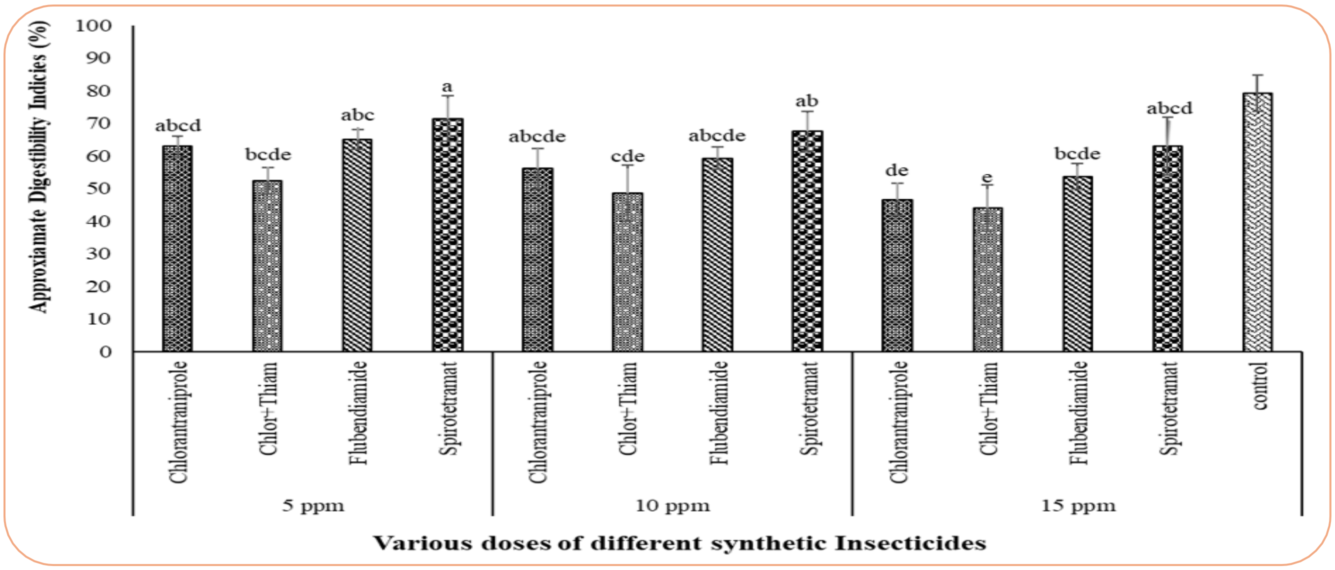

Figure 7. Impact of different synthetic insecticides at various concentrations on the approximate digestibility indices of 3rd instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 16.631, $p < 0.05$).

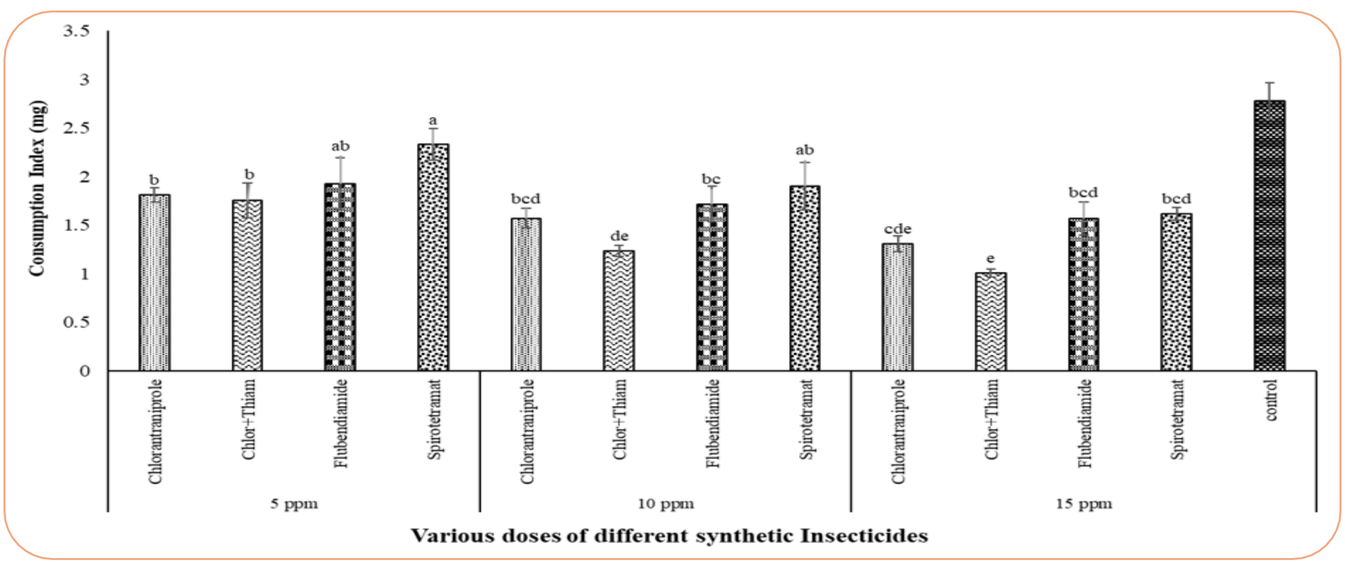

Figure 8. Impact of different synthetic insecticides at various concentrations on the consumption index of 4th instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 0.4364, $p < 0.05$).

The reduced feeding activity of 4th-instar *S. frugiperda* larvae after 48 h of insecticide application resulted in a comparatively lower growth rate (GR) across the tested synthetic insecticides and concentrations (Figure 9). Although the interaction between insecticides and concentrations was non-significant (F = 0.36, $p > 0.05$), significant differences were detected among insecticides (F = 51.41, $p < 0.001$) and concentrations (F = 11.82, $p < 0.001$) in their effects on the GR of 4th-instar *S. frugiperda* larvae. The untreated control exhibited the highest GR (0.392 ± 0.01 g), whereas chlorantraniliprole + thiamethoxam produced the lowest GR (0.133 ± 0.01 g) at 5 ppm, followed by chlorantraniliprole (0.198 ± 0.007 g), flubendiamide (0.241 ± 0.02 g), and spirotetramat (0.324 ± 0.03 g). At 10 ppm, spirotetramat resulted in the highest GR (0.286 ± 0.01 g), followed by flubendiamide (0.208 ± 0.01 g), chlorantraniliprole (0.165 ± 0.01 g), and chlorantraniliprole + thiamethoxam (0.119 ± 0.01 g). At 15 ppm, the lowest GR was recorded for chlorantraniliprole + thiamethoxam (0.098 ± 0.007 g), followed by chlorantraniliprole (0.119 ± 0.005 g), flubendiamide (0.187 ± 0.009 g), and spirotetramat (0.249 ± 0.03 g).

Figure 10 presents the effects of different insecticides and their concentrations on the efficiency of conversion of ingested food (ECI) in 4th-instar *S. frugiperda* larvae 48 h after treatment. A highly significant difference was observed among the insecticides (F = 10.06, $p < 0.001$), whereas neither insecticide concentration (F = 0.52, $p \geq 0.05$) nor the insecticide × concentration interaction (F = 0.26, $p > 0.05$) had a significant effect on ECI. The highest ECI was recorded in the untreated control (17.764 ± 1.25%). At 5 ppm, Spirotetramat resulted in the highest ECI (13.571 ± 1.16%), followed by Flubendiamide (11.026 ± 1.00%), Chlorantraniliprole (9.930 ± 0.23%), and Chlorantraniliprole + Thiamethoxam (9.730 ± 1.09%). At 10 ppm, Spirotetramat again produced the highest ECI (12.903 ± 0.70%), followed by Flubendiamide (11.051 ± 1.11%), Chlorantraniliprole (10.005 ± 0.32%), and Chlorantraniliprole + Thiamethoxam (8.116 ± 1.40%). At 15 ppm, the lowest ECI was recorded for Chlorantraniliprole + Thiamethoxam (7.403 ± 0.91%), followed by Chlorantraniliprole (9.588 ± 0.39%), Flubendiamide (10.745 ± 2.28%), and Spirotetramat (13.366 ± 1.18%). Overall, ECI was substantially lower in insecticide-treated larvae than in the untreated control, with the Chlorantraniliprole + Thiamethoxam combination producing the lowest ECI, particularly at the highest concentration, indicating a pronounced reduction in larval food-conversion efficiency.

Figure 11 presents the effects of different concentrations of synthetic insecticides on the efficiency of conversion of digested food (ECD) in 4th-instar *S. frugiperda* larvae 48 h after treatment. A significant difference was observed among the insecticides (F = 6.37, $p < 0.05$), whereas the effects of insecticide concentration (F = 1.97, $p > 0.05$) and the interaction between insecticide and concentration (F = 0.25, $p > 0.05$) were non-significant. The highest ECD was recorded in the control treatment (20.120 ± 1.47%), whereas the lowest ECD was observed with chlorantraniliprole + thiamethoxam (12.484 ± 0.30%) at 15 ppm, followed by chlorantraniliprole (13.941 ± 1.27%), flubendiamide (15.782 ± 0.85%), and spirotetramat (17.010 ± 0.99%).

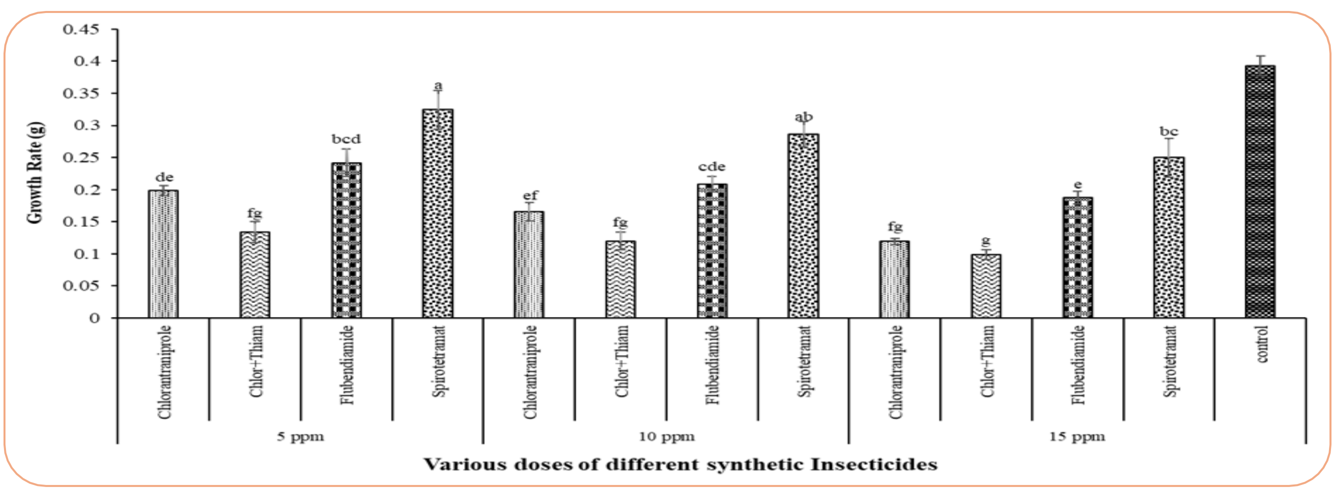

Figure 9. Impact of different synthetic insecticides at various concentrations on the growth rate of 4th instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 0.0500, $p < 0.05$).

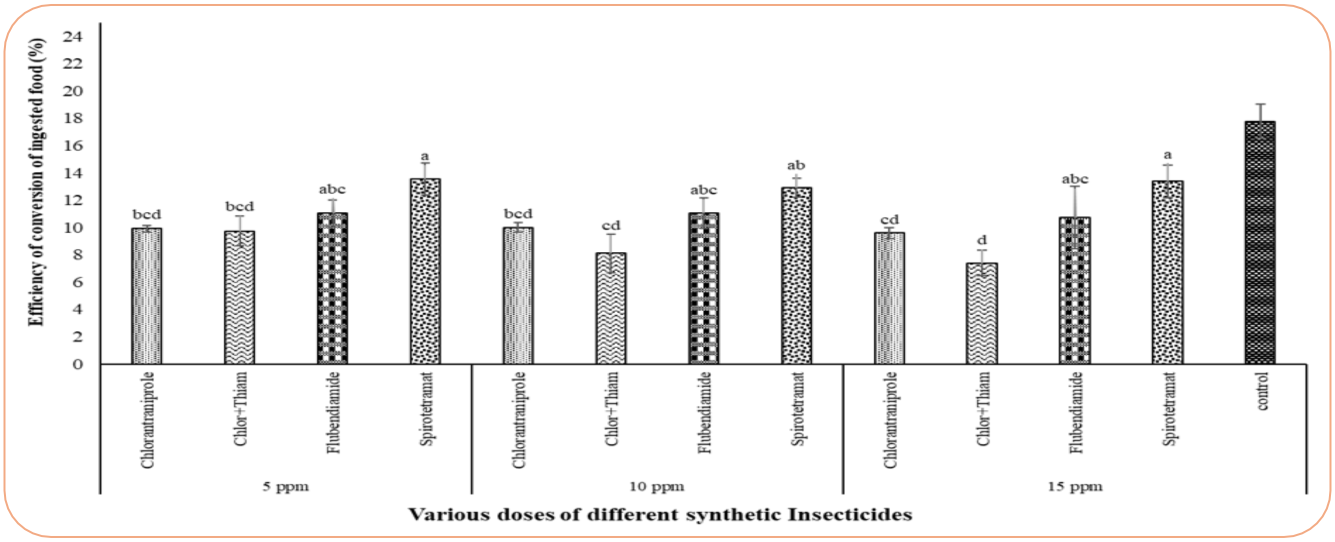

Figure 10. Impact of different synthetic insecticides at various concentrations on the efficiency of conversion of ingested food by 4th instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 3.1868, $p < 0.05$).

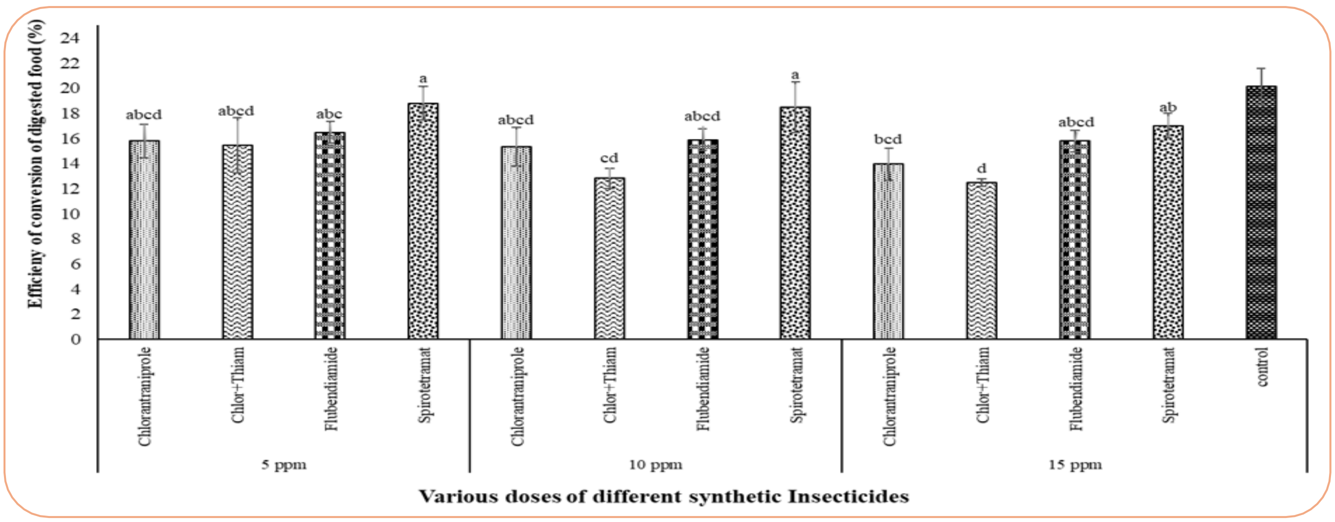

Figure 11. Impact of different synthetic insecticides at various concentrations on the efficiency of conversion of digested food by 4th instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 3.7043, $p < 0.05$).

At 10 ppm, the highest ECD was recorded with spirotetramat (18.466 ± 2.01%), followed by flubendiamide (15.891 ± 0.83%), chlorantraniliprole (15.343 ± 1.55%), and chlorantraniliprole + thiamethoxam (12.804 ± 0.78%). At 5 ppm, the lowest ECD was recorded with chlorantraniliprole + thiamethoxam (15.443 ± 2.18%), followed by chlorantraniliprole (15.792 ± 1.34%), flubendiamide (16.471 ± 0.86%), and spirotetramat (18.805 ± 1.34%).

Results for the Approximate Digestibility Index (ADI) of 4th-instar *S. frugiperda* larvae after 48 h of exposure to different concentrations of the tested insecticides are presented in Figure 12. The results revealed a significant difference among the synthetic insecticides ($F = 5.13$, $p < 0.05$), whereas the effects of insecticide concentration ($F = 1.00$, $p > 0.05$) and the interaction between insecticide and concentration ($F = 0.14$, $p > 0.05$) were non-significant. The untreated control exhibited the highest ADI (82.310 ± 6.46%). At 15 ppm, the lowest ADI was recorded for chlorantraniliprole + thiamethoxam

(52.985 ± 1.47%), followed by chlorantraniliprole (59.844 ± 6.95%), flubendiamide (68.743 ± 3.64%), and spirotetramat (77.919 ± 8.13%). At 10 ppm, spirotetramat recorded the highest ADI (78.882 ± 6.35%), followed by flubendiamide (65.931 ± 10.59%), chlorantraniliprole (62.312 ± 4.84%), and chlorantraniliprole + thiamethoxam (58.004 ± 8.65%). Similarly, at 5 ppm, the lowest ADI was observed with chlorantraniliprole + thiamethoxam (63.219 ± 8.45%), followed by chlorantraniliprole (69.899 ± 4.78%), flubendiamide (72.280 ± 6.82%), and spirotetramat (79.762 ± 4.63%).

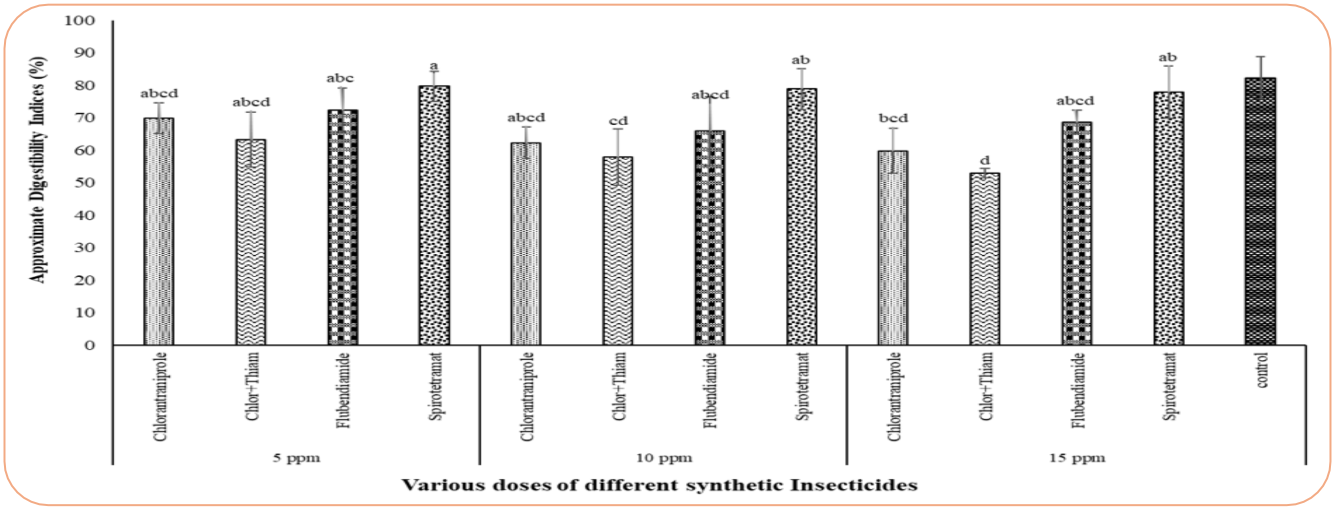

Figure 12. Impact of different synthetic insecticides at various concentrations on the approximate digestibility indices of 4th instar *S. frugiperda* larvae after 48 h. Means followed by same letters are not significantly different from each other (LSD = 19.136, $p < 0.05$).

Discussion

Given the substantial economic damage caused by fall armyworm worldwide and the importance of insecticides in reducing crop losses, the present study evaluated the efficacy of four synthetic insecticides, chlorantraniliprole + thiamethoxam, chlorantraniliprole, flubendiamide, and spirotetramat, against 3rd- and 4th-instar *S. frugiperda* larvae. Their effects on larval feeding, growth, and nutritional physiology were also assessed using different digestibility and conversion indices. Each insecticide was evaluated at three concentrations corresponding to 25, 50, and 100% of the recommended field dose.

The combination of chlorantraniliprole + thiamethoxam exhibited the highest toxicity, causing mortality of 97.96% and 88.51% in 3rd- and 4th-instar larvae, respectively, at 15 ppm. The greater susceptibility of 3rd-instar larvae suggests that larval age substantially influences insecticide toxicity. The enhanced efficacy of the combination may reflect complementary modes of action of its two active ingredients, although the present results alone do not establish pharmacological synergy. Susanto et al. (2021) similarly identified chlorantraniliprole + thiamethoxam as an effective option for managing *S. frugiperda* in Indonesia. Higher susceptibility of younger larvae has also been reported by Tapsoba et al. (2025) and Patel and Jhala (2020), which may be associated with differences in cuticular properties, detoxification capacity, and physiological development between younger and older instars.

Chlorantraniliprole alone was the second most effective treatment, producing mortality of 87.97% in 3rd-instar and 73.01% in 4th-instar larvae at 15 ppm. Its pronounced activity against younger larvae is consistent with the findings of Idrees et al. (2022), who demonstrated substantial toxicity of chlorantraniliprole against early larval stages of *S. frugiperda*. Flubendiamide exhibited intermediate efficacy, causing 76.60% and 62.86% mortality in 3rd- and 4th-instar larvae, respectively. This response was consistent with the moderate mortality and sublethal effects reported by Deshmukh and Naikwadi (2023). In contrast, spirotetramat produced the lowest mortality, at 63.87% and 38.15% in 3rd- and 4th-instar larvae, respectively. Its comparatively lower acute toxicity, together with its effects on physiological processes, suggests that its contribution to *S. frugiperda* management may extend beyond direct mortality.

In addition to mortality, the insecticides substantially affected larval feeding and growth, as reflected by changes in the consumption index and growth rate. Chlorantraniliprole + thiamethoxam produced the greatest reductions in these parameters, indicating pronounced disruption of feeding and biomass accumulation. In 3rd-instar larvae, CI and GR were 0.985 mg and 0.092 g, respectively, following treatment with the combination. A similar pattern was observed in 4th-instar larvae, demonstrating that the combined treatment impaired both food consumption and subsequent growth in addition to causing mortality. These findings are broadly consistent with Gómez and Pérez (2022), who reported marked reductions in food consumption and growth efficiency following exposure of larvae to chlorantraniliprole-based insecticides. Such reductions in feeding may have important ecological implications because sublethal inhibition of consumption can reduce larval development and consequently limit the probability of successful transition to the pupal and adult stages.

Spirotetramat produced comparatively moderate effects on CI and GR, particularly in 4th-instar larvae, for which the values remained relatively close to those of the control. This response suggests that its effects may involve physiological disruption in addition to direct lethality. Saleh et al. (2024) reported that spirotetramat can induce indirect physiological effects, including alterations in enzyme activity and developmental abnormalities. Consequently, although spirotetramat exhibited lower acute toxicity than the other insecticides tested, its sublethal effects may still contribute to population suppression and could be relevant to integrated pest management (IPM) programs.

The conversion indices, including efficiency of conversion of ingested food (ECI), efficiency of conversion of digested food (ECD), and approximate digestibility index (ADI), further demonstrated substantial physiological disturbances following insecticide exposure. In 3rd-instar larvae treated with chlorantraniliprole + thiamethoxam, the lowest ECI (6.81%) and ECD (11.61%) were recorded at 15 ppm. Similarly, 4th-instar larvae exhibited ECI and ECD values of 7.40% and 12.48%, respectively. These reductions indicate impaired utilization of ingested and digested food for biomass production and may reflect disturbances in nutrient assimilation, digestion, or metabolic allocation. Silva et al. (2020) and Chaudhary and Singh (2021) similarly associated reductions in conversion efficiency with impaired midgut function and compromised nutrient utilization following insecticide exposure. In contrast, spirotetramat produced ECI and ECD values closer to those of the control, particularly at lower concentrations. At 5 ppm, ECI and ECD in 3rd-instar larvae were 12.49% and 16.49%, respectively. Although these relatively small reductions indicate less severe nutritional impairment, they may nevertheless reflect physiological stress with potential consequences for subsequent development, pupation, and adult reproductive performance, as suggested by Sagar et al. (2022).

The ADI was generally higher in untreated larvae, whereas chlorantraniliprole + thiamethoxam produced marked reductions in ADI in both 3rd- and 4th-instar larvae, with values of 44.08% and 52.98%, respectively. Reduced digestibility may indicate alterations in digestive physiology, nutrient assimilation, or enzyme activity. Similar physiological disturbances have been associated with prolonged dietary stress and insecticide exposure, which can compromise larval performance and survival (Silva et al., 2019). Collectively, the reductions in CI, GR, ECI, ECD, and ADI demonstrate that insecticide efficacy cannot be evaluated solely on the basis of mortality. Sublethal effects on feeding and nutrient utilization may substantially contribute to population suppression by delaying larval development and reducing the resources available for subsequent life stages.

Overall, the findings highlight the importance of integrating lethal and sublethal endpoints when evaluating insecticides for *S. frugiperda* management. 'Chlorantraniliprole + thiamethoxam' was the most effective treatment, producing the greatest mortality and the strongest reductions in feeding, growth, and nutritional efficiency. Chlorantraniliprole alone also demonstrated considerable efficacy and may therefore represent a useful component of insecticide rotation programs designed to diversify modes of action and delay resistance development. Flubendiamide showed intermediate efficacy and may serve as an additional management option, whereas spirotetramat exhibited comparatively lower acute toxicity but measurable physiological effects. Nevertheless, recommendations regarding reduced-dose field application should be made cautiously, because laboratory responses may not directly translate to field conditions. Further field-based studies are therefore required to validate the efficacy of these treatments, determine appropriate dose optimization, and assess their effects on resistance development, natural enemies, and other components of the maize agroecosystem.

Conclusions

The present study demonstrated that all four tested insecticides were effective against 3rd- and 4th-instar *S. frugiperda* larvae, with efficacy generally increasing with increasing concentration. Chlorantraniliprole + thiamethoxam and chlorantraniliprole alone exhibited the greatest toxicity, followed by flubendiamide and spirotetramat. Third-instar larvae were generally more susceptible than 4th-instar larvae, indicating a clear influence of larval developmental stage on insecticide susceptibility. In addition to causing mortality, the insecticides substantially affected feeding, growth, digestibility, and food-conversion efficiency, particularly at higher concentrations.

Chlorantraniliprole + thiamethoxam produced the strongest overall effects on both mortality and nutritional physiology, whereas chlorantraniliprole alone also demonstrated substantial efficacy. These findings indicate that both lethal and sublethal responses should be considered when selecting insecticides for *S. frugiperda* management. However, the apparent efficacy of reduced concentrations under laboratory conditions should not be interpreted as sufficient evidence for field application at reduced doses. Field validation is essential to determine effective and economically viable application rates while minimizing selection pressure for insecticide resistance and adverse effects on beneficial organisms. Further investigations should therefore focus on dose optimization, residual efficacy, resistance management, and compatibility with broader IPM strategies.

Author's contribution

AAG, LBR, and GHJ conceived, designed, and supervised the study. MHI and MMK conducted the experiments. GSK performed the statistical analysis. SAL prepared the initial manuscript draft, and AAG critically revised and finalized the manuscript.

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Conflict of Interest

The authors declare no conflict of interest.

Sustainable Development Goals Targeted

SDG 2: Zero Hunger

SDG 12: Responsible Consumption and Production

SDG 15: Life on Land

Policy Addressed

The findings support integrated pest management, rational pesticide use, and insecticide resistance management in maize. They also provide evidence for optimizing insecticide applications against *Spodoptera frugiperda* while minimizing unnecessary pesticide exposure and promoting sustainable maize production.

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