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RESEARCH

Biometric Traits and Biology of Fall Armyworm (*Spodoptera frugiperda*) Reared on Maize (*Zea mays*) under Laboratory Conditions

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Abstract

The Fall armyworm (*Spodoptera frugiperda*), a formidable pest now widespread in Nepal, poses a major threat to maize production, underscoring the need for detailed biological insights to inform effective management strategies. Therefore, this study was conducted under laboratory conditions to assess the morphometric and biological parameters of FAW on *Zea mays*. Morphometric traits such as head capsule width and length, body length and width, adult body length, and wingspan were examined. Biological parameters, including the duration of larval instars, pre-pupal, pupal, and adult stages, were also analyzed. In larvae, the smallest measurements were recorded in the first instar (head capsule length 0.20±0.02 mm; width 0.31±0.02 mm; body length 1.48±0.08 mm; body width 0.30±0.03 mm), while the largest were observed in the sixth instar (2.85±0.09 mm, 2.86±0.06 mm, 33.23±0.43 mm, and 5.15±0.14 mm, respectively). Pupae measured 14.45±0.78 mm in length and 4.40±0.04 mm in width. The adult forewing span (30.5±1.6 mm) was larger than the hindwing span (22.6±1.0 mm), with a body length of 13.8±0.9 mm. Egg hatching took 2 days, and the larval stage lasted between 9 and 14.3 days. The pre-pupal and pupal stages lasted 2.20±0.47 and 9.08±0.90 days, respectively, followed by an adult lifespan of 5.50±1.50 days. The results shed light on the lifecycle, essential for understanding its population dynamics and guiding ecologically based management.

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Statement of Sustainability: This study provides crucial insights into growth patterns and developmental stages of *Spodoptera frugiperda* as a major pest of maize. The study helps build sustainable and targeted pest management strategies that reduce crop losses and increase maize output. These observations enable farmers and other agricultural stakeholders to put into practice practical strategies that improve food security and foster resilient maize production systems, which is in line with SDG 2-Zero Hunger.

1. Introduction

Maize (*Zea mays* L.) ranks among the world's most important cereal crops, widely cultivated under various soil types and agroclimatic conditions. It is among the three most extensively grown crops, following rice and wheat, in developing countries (Sairam et al., 2025). In Nepal, maize ranks after rice as a major cereal and plays a crucial role in food security. It is a dietary staple in the hilly regions and plays a major role in the Terai's animal feed industry (Atreya et al., 2025; Bogati et al., 2021; Thapa et al., 2022; Shrestha et al., 2023; Gautam et al., 2022).

Despite its importance, maize production suffers significant yield losses due to insect pests. At least 12 insect species are recognized as major pests that attack maize throughout its growth stages, from planting to storage (Bhatti et al., 2021). Even with continuous management efforts, insect-related losses remain a persistent challenge (Dessie et al., 2024). Among these pests, the fall armyworm (*Spodoptera frugiperda* J. E. Smith), has emerged as one of the most destructive pests to maize worldwide (Fan et al., 2024; Firake and Behere, 2020). Fall armyworm (FAW) is a highly polyphagous pest, reported to feed on 353 host plants across 76 families, including many important crops such as rice, sugarcane, sorghum, tomato, potato, cotton, and beet. However, maize is the most severely affected host. The pest's exceptional ability to migrate allows adults to travel several hundred kilometers in a single night (Wang et al., 2025). Infestation is further accelerated by high fecundity, wide host range, and the ability to evolve resistance to pesticides. Females are capable of laying up to 1,500 eggs during their short lifespan (Khatun et al., 2023; Chisonga et al., 2023). As a consequence of these biological traits, FAW larvae cause severe damage by feeding on leaves, stems, tassels, silks, and ears, leading to symptoms such as "dead heart," ragged leaf edges, and papery windows. Older larvae often conceal themselves within maize funnels, reducing the efficacy of foliar insecticide applications, whereas younger larvae hide during the day and feed nocturnally (Mahat et al., 2021; Chen et al., 2025; Kusi et al., 2024; Mohamed et al., 2023; G. C. et al., 2020).

Nepal provides a favorable temperature range for the establishment of FAW, making the country particularly susceptible to its invasion (Bista et al., 2020). In this context, developing sustainable management strategies requires a comprehensive understanding of the biology, morphology, damage symptoms, life cycle, and migration of FAW (Sharma et al., 2022; Hailu et al., 2024; EL-Lebody et al., 2024). Knowledge of FAW's biological characteristics and the host range it targets is crucial to comprehending its invasion patterns, predicting population trends, and identifying the most vulnerable life stage for control. This understanding also lays a foundation for future research on how environmental changes, such as global warming, may influence pest dynamics (Youssef et al., 2024; Chaudhary et al., 2023; Reddy et al., 2021; Kalyan et al., 2020). Additionally, morphometric studies provide valuable insights into growth patterns and their interactions with various maize cultivars. Such research can identify crop varieties that either promote or impede pest development, thereby enhancing predictive models of population dynamics and supporting sustainable farming practice (Choudhari et al., 2024).

In this study, we concentrated on understanding *S. frugiperda*'s developmental biology under controlled settings, intending to provide insights into developmental stages and critical morphological features. We meticulously observed larval instars, pupal features, and adult morphometrics to create baseline data that can aid in accurate identification and stage-specific management. In a larger sense, our findings help to integrate pest management tactics by accurately distinguishing between FAW and related species, providing a foundation for building long-term management strategies.

2. Materials and Methods

2.1. Description of Study Site

The egg mass of the fall armyworm was collected from a maize field at the College of Natural Resource and Management, Agriculture and Forestry University in Rolpa, Nepal, located at 28°34'41.8'' N latitude, 82°59'90.7'' E, and at an altitude of 996.6 meters above sea level. The egg mass was then transferred to the entomology laboratory for maintenance under controlled conditions. In the lab, the larvae were raised with a relative humidity of 70±10% and a temperature of 26±2°C. A 14-hour light and 8-hour dark photoperiod was maintained, creating an ideal environment for consistent growth, physiological development, and overall larval performance.

2.2. Rearing of Fall Armyworm under Laboratory Conditions

The collected egg mass was kept in a petri dish (12 cm in diameter and 8.5 cm in height) lined with filter paper and a moistened cotton ball to create a microenvironment conducive to hatching. After hatching, the neonates were carefully removed from the remaining flocculent coating using a camel-hair brush and transferred to a different petri dish to avoid cannibalism. All larval stages were reared in petri dishes and fed only fresh maize leaves. Additionally, the filter paper lining the dishes was changed daily to maintain sanitation and prevent contamination. Growth, development, and survival were closely monitored until pupation. Once in the pupal stage, the larvae were transferred to oviposition cages measuring 50 × 50 × 50 cm, containing a maize plant as the natural substrate. A 10% honey solution was provided to adult moths to support their nutrition and reproductive capacity (Hong et al., 2021). The newly laid egg masses were then collected and studied for their biology and morphometric traits during development.

2.3. Host Plant

A 2 m^2 plot was established inside a net house, where maize plants of the variety Manakamana-3 were grown about 20 days before the expected hatching of first-instar larvae. To ensure consistent nutrition during larval development, fresh maize leaves were carefully collected and fed to the larvae. The leaves were replaced regularly until pupation. The same maize variety was cultivated in plastic pots (10 cm diameter and 15 cm height) for the adult stage, and they were planted 15 days before the adults emerged. These 15-day-old maize plants served as substrates for oviposition, providing suitable host material for egg laying.

2.4. Biology of Different Life Stages

The biological variables were meticulously assessed from 30 individuals across all developmental stages, including the entire life cycle from egg to the end of the adult stage, as well as the duration of egg incubation, the chronological age of successive larval instars (I-VI), pre-pupae and pupae, adult lifespan, and fertility, determined by the number of eggs laid per female (Maharani et al., 2021). Adult lifespan was assessed under non-feeding conditions, where individuals were kept without access to any food (nectar) during their entire lifespan. The transition between larval instars was marked by the shedding of the old cuticle at each larval ecdysis (Hong et al., 2021). To count the number of eggs per cluster, the protective scales on adult moths were gently removed using a camel-hair brush. The exposed eggs were counted with a hand lens (10×18 mm) to accurately measure reproductive output (Sharma et al., 2022).

2.5. Morphometrics of Different Life Stages

Morphometric parameters, including head capsule dimensions, wings, body lengths, and pupal measurements, were carefully gathered from 30 individuals at each developmental stage. For the assessment, exuviae were examined under a microscope. Immediately after each larval molt, measurements were taken for head capsule width and length, and also for body length and width, using calibrated stage and ocular micrometers (AM Scope stage micrometer and Olympus ocular micrometer) for first and second-instar larvae (Bankar et al., 2025). The third to sixth larval instars, pupae, and adults were measured precisely using a digital Vernier caliper (0–150 mm). While pupal measurements were taken from the anterior tip of the head to the posterior end of the abdomen, along with the width of the broadest segment, larval measurements included body length and width as well as head capsule width (Neupane et al., 2023). The wings of the adult males and females were thoroughly spread and dried before being pinned. Body length was measured from the anterior tip of the head to the posterior tip of the abdomen, and forewing expansion was measured (Khatun et al., 2022). A digital Vernier caliper was used for all measurements to guarantee accuracy.

2.6. Statistical Analysis

The experimental data were collected in the laboratory and then entered into Microsoft Excel 2019 for organizing and preprocessing. All statistical analyses, including data visualization and descriptive statistics, were performed using the statistical software R (version 4.4.3).

3. Result and Discussion

3.1. Morphometric Traits and Measurements

The clusters of eggs laid by females typically ranged from one to two overlapping layers, naked or covered by a thin grayish scale. A similar cluster was reported by Kasige et al. (2022), where eggs were found in both single and multiple layers. In our study, the freshly laid eggs by gravid females were spherical and pale green, but just before hatching (Figure 1a-i), they turned golden yellow and eventually black, which is consistent with the results reported by Marri et al. (2023) and Sharma et al. (2022).

The 1st instar larva was small and semi-transparent, with a darkening prothoracic shield, a contrasting black head capsule, and a pale, creamy body, similar to the descriptions observed by Madali et al. (2025). Larvae in their 2nd instar were greenish-yellow, but those in their 3rd instar displayed three developing white stripes along their dorsal and lateral areas while maintaining the same color, consistent with the observation of Manjula et al. (2019). In contrast to the medium-sized, fairly developed head capsule of 3rd instar larvae, 4th instar larvae showed growth, as seen in a somewhat larger head capsule in length and width, along with more pronounced abdominal spot patterns and clearer body segmentation. The larvae became robust and cylindrical in later instars, especially the 5th and 6th, with dark brown bodies, prominent mandibles, an inverted "Y" mark on the head, white dorsal streaks, and rectangular spot patterns on the abdominal segments, aligning with the findings of Bhatti et al. (2021) and Sharma et al. (2022). After the last molt, pupae gradually changed from yellowish to bright green, eventually turning glossy reddish-brown (Figure 2j-l), which corroborates previous findings by Neupane et al. (2023). In females, sexual dimorphism is evident by a comparatively greater distance between the genital and anal openings, the pattern that aligns with that of Siddhapara et al. (2021). The head capsule length, head capsule width, and larval length for each of the six larval instars are summarized in Table

1. The average head capsule length of the 1st instar was 0. 20±0. 02 mm, increasing progressively through the 2nd instar (0. 39±0. 04 mm), 3rd instar (0. 80±0.06mm), 4th instar (1. 05±0. 05 mm), 5th instar (1. 88±0. 09 mm), and 6th instar (2. 85±0. 09 mm) instars. Similarly, the head capsule width of the 1st instar was 0.31±0.02 mm and increased gradually in subsequent instars, reaching a maximum of 2.86±0.06 mm in the 6th instar. Correspondingly, the body length of the 1st instar larvae measured 1.48±0.08 mm, increasing in later instars to 33.23±0.43mm in the 6th instar. Finally, the larva's breadth measured 0. 30±0. 03 mm in the 1st instar, then increased progressively in the 2nd (0. 76±0.05mm), 3rd (1. 78±0. 09 mm), 4th (2. 03±0. 07 mm), 5th (3. 30±0. 31 mm), and reached 5.15±0.14 mm in the 6th instar. In line with Montezano et al. (2019), Bhatti et al. (2021), and Khatun et al. (2022), this study demonstrated that the measurements of the fall armyworm's head capsule, body width, and body length vary with instar, being smaller in the early instars and growing in size as development progresses. The pupal stage had a body length of 14. 45±0.78 mm and a breadth of 4.40±0.04 mm. Ojumoola and Omoloyea (2022) reported slightly smaller dimensions compared to those seen in our work, which may have been due to discrepancies in the experimental setting. Overall, the steady increase in all morphometric features from the first instar to the pupa demonstrated the gradual growth of *S. frugiperda*.

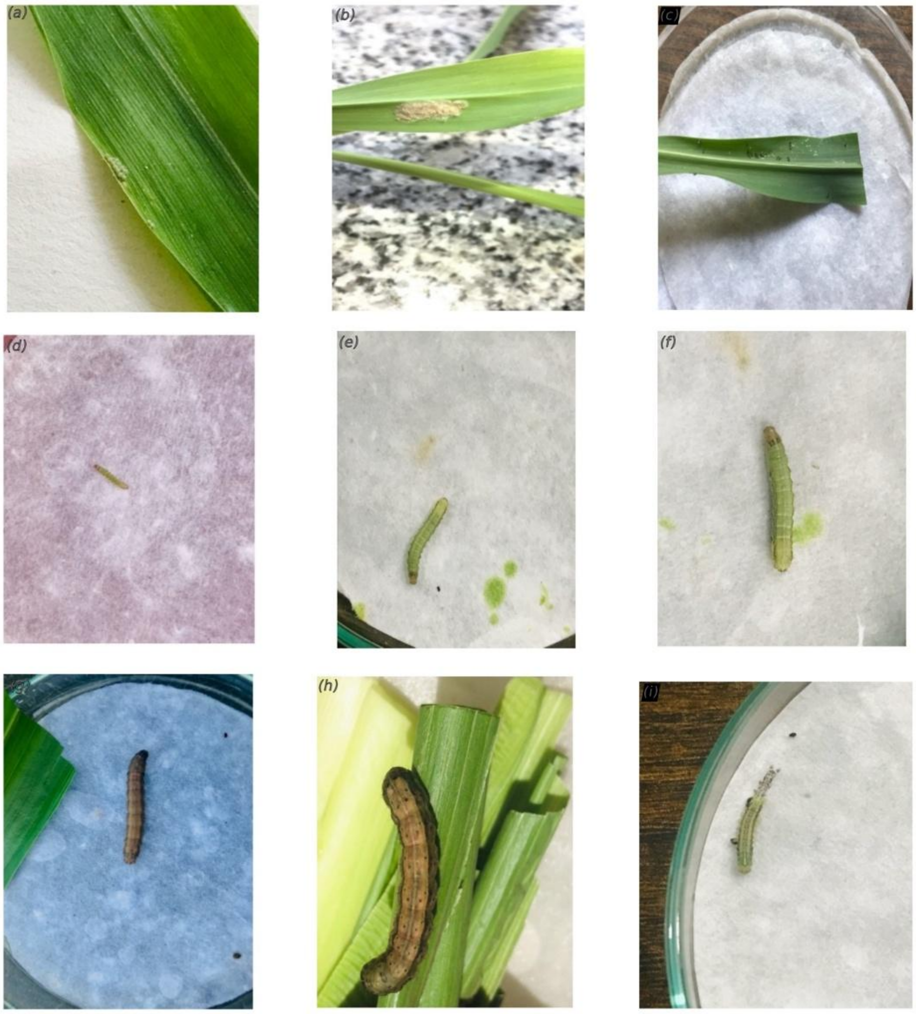

Figure 1. Egg mass and larvae of *S. frugiperda*: Eggs are laid in clusters, firmly attached to the substrate and arranged in close-packed rows resembling a beaded chain. Individual eggs are round, small, and uniform in color, with smooth, glossy, pigmentless chorions ranging from creamy white to pearl white. Dorsally, the eggs appear symmetrical and circular. (a) Egg masses are laid in a single layer, with clutches exposed. (b) Egg masses are deposited in multiple layers and fully covered with scales and hairs derived from the female's body. (c) First-instar larvae have a prominent black head capsule on a light green body. (d) Second-instar larvae have gradually darker and more robust bodies. (e) Third-instar larvae show emerging white longitudinal stripes along the body. (f) Fourth-instar larvae display a prominent white longitudinal stripe along the body. (g) Fifth-instar larvae have a reddish-brown head, a brownish body, a noticeable inverted "Y" mark between the eyes, and fully formed mouthparts. (h) Sixth-instar larvae have a brownish body, a distinct midline extending from the base of the head's inverted "Y" mark to the final abdominal segment, and four conspicuous black dorsal spots on the penultimate segment arranged in a square. (i) depicts a larva during molting as it sheds its outer cuticle to allow growth and transition to the next instar.

The wing patterns of adult moths showed sexual dimorphism (Figure 2m-n); males had dark brown to grey forewings with characteristic kidney-shaped patterns, while females had uniformly grey forewings. Both sexes had silvery-white hindwings with brownish apical margins, which aligns with the previous findings by Navasero and Navasero (2020). The forewing span averaged 30.5±1.6 mm, whereas the hindwing spanned slightly less at 22.6±1.0 mm, and the body length of the adult was 13.8±0.9 mm, reflecting a comparatively smaller size relative to the wings as reported by Khatun et al. (2022).

Figure 2. Developmental stages from pre-pupae to adult of *S. frugiperda*: j) shows a prepupae. (k) recently emerged pupa, which is initially green, but progressively changes to brown as the pupa matures. (l) illustrates a fully grown pupa with a reddish-brown coloration, signifying complete cuticle sclerotization. (m) Adult males have a straw-colored discal cell and triangular white dots near the tip and in the middle of the wing; the forewing is mottled in light brown, grey, and straw colors. (n) Females have less pronounced patterns on their forewings, with subtle brown and grey mottling.

Table 1. Morphometric trait measurements spanning the developmental phase of *S. frugiperda*.

Larval instar	**Head Capsule Length (mm)±SD**	**Head Capsule Width (mm)±SD**	**Length of larva (mm)±SD**	**Breadth of larva (mm)±SD**
1st Instar	0.20±0.02	0.31±0.02	1.48±0.08	0.30±0.03
2nd Instar	0.39±0.04	0.58±0.06	3.46±0.25	0.76±0.05
3rd instar	0.80±0.06	0.89±0.06	7.75±0.61	1.78±0.09
4th Instar	1.05±0.05	1.53±0.07	11.58±0.44	2.03±0.07
5th Instar	1.88±0.09	2.07±0.06	18.45±0.44	3.30±0.31
6th Instar	2.85±0.09	2.86±0.06	33.23±0.43	5.15±0.14
Pupae	-	-	14.45±0.78	4.40±0.04
Adult	-	Forewing Span (mm)±SD	Hindwing Span (mm)±SD	Length of Body (mm)±SD
		30.5±1.6	22.6±1.0	13.8±0.9

Note: Length measurements are recorded in millimeters (mm), and the standard deviation (SD) represents the variability of measured values around the mean.

3.2. Biological Parameters and Measurements

The duration of each development stage in *S. frugiperda* showed clear differences (Table 2). The egg stage lasted for 2±00 days. The 1st instar persisted for 1.96±0.08 days (range 1.92–2.21), while the 2nd and 3rd instars lasted 1.82±0.07 days (1.79–2.00) and 1.11±0.31 days (1.00–2.00), respectively. The 4th instar took 1.72±0.33 days (1.00–2.00), and the 5th and 6th instars extended longer, at 2.18±0.38 days (1.29–3.00) and 2.34±0.45 days (2.04–3.08), respectively. The developmental period of the fall armyworm from the first to sixth instars studied in this study showed tendencies comparable to those described in previous studies by Pathak et al. (2024) and Neupane et al. (2023). However, the observed variability in developmental length may be due to differences in nutritional quality and laboratory methods. The pre-pupae stage was relatively short, lasting 2.20±0.47 days (1.50–2.50). Pupation took a significant amount of time, 9.08±0.90 days (7.00–10.00), contributing substantially to the total development period, which is consistent with the study by Rajisha et al. (2022).

Table 2. Developmental duration of the various life stages of *S. frugiperda*

Stages	Mean (days)±SD	Minimum and maximum range
Eggs	2±00	2
Instar 1	1.96±0.08	1.92–2.21
Instar 2	1.82±0.07	1.79–2.00
Instar 3	1.11±0.31	1.00–2.00
Instar 4	1.72±0.33	1.00–2.00
Instar 5	2.18±0.38	1.29–3.00
Instar 6	2.34±0.45	2.04–3.08
Prepupa	2.20±0.47	1.50–2.50
Pupae	9.08±0.90	7.00–10.00
Adult	5.50±1.50	4.00-7.00
Other biological parameters		
Male: Female sex ratio	1:1.13	

Note: the standard deviation (SD) represents the variability of measured values around the mean.

The non-fed adult lifespan ranged from 4 to 7 days (Figures 3 and 4), with an average of 5.50±1.50 days, which partially aligns with the result of Ojumoola and Omoloyea (2022). These findings reveal a gradual increase in developmental time from early to late larval instars, leading to longer pupal and adult lifespans. The recorded sex ratio showed a slight dominance of females over males (1.13 females: 1 male), indicating the higher reproductive potential of the population. In line with this, Huang et al. (2021) and Sharma et al. (2022) also observed a greater abundance of females over males. Previously, Lekha et al. (2020) found that the fall armyworm biology is regulated by parameters such as temperature, relative humidity, and host plants.

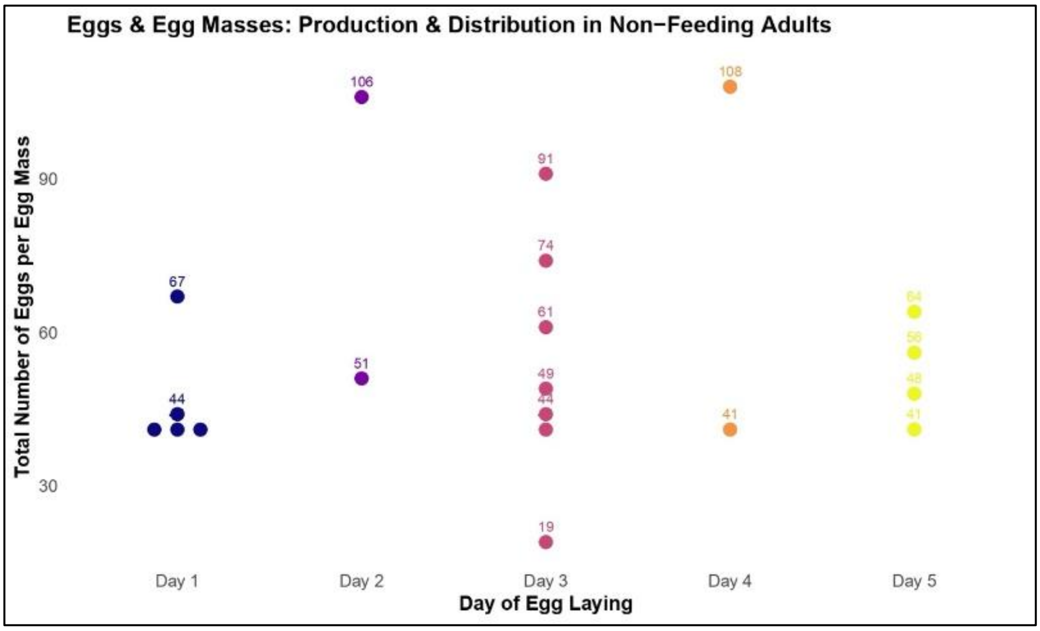

Figure 3. A beeswarm plot depicting egg-laying by a female moth during the oviposition period in non-feeding conditions. Each dot represents a single egg mass, and the number above indicates the total number of eggs in the mass.

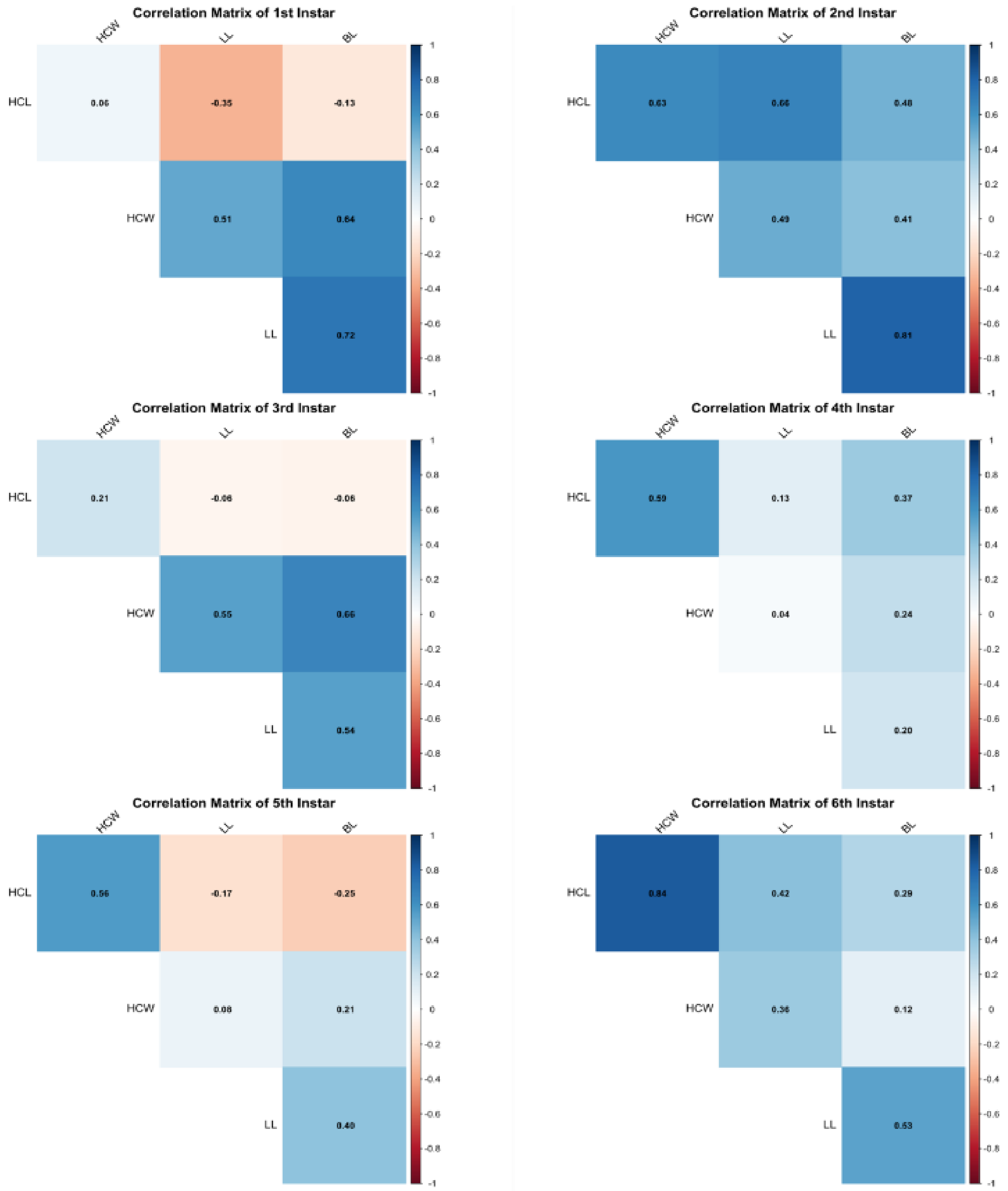

Figure 4. Correlation heatmap among Morphometric traits in larvae of *S. frugiperda* (Note: The negative correlations are depicted in red, while positive correlations appear in blue, with intensity indicating the strength of association. Since data were taken from individual larvae across 30 Petri plates, the matrix provides a descriptive summary of observed relationships. HCL: Head Capsule Length, HCW: Head Capsule Width, LL: length of larva, BL: Breadth of larva).

4. Conclusion

This study provides an analysis of the species' morphometric and biological traits throughout its complete developmental cycle, from egg to adult. Head capsule size, body and wing proportions, and pupal shape all differed, emphasizing the dynamic growth patterns unique to each instar and developmental stage. A correlation study across different instars revealed interrelated growth patterns, suggesting potential predictive links between early and later developmental features. Furthermore, evaluating reproductive traits such as egg production, egg mass distribution under non-feeding situations, and sex ratios provides vital insights into the species' reproductive biology and population dynamics. As a whole, these findings expand on our basic knowledge of the species' lifecycle and offer a strong foundation for further research in the fields of developmental biology, ecological monitoring, and practical pest control. This study emphasizes the importance of a thorough strategy for clarifying insect life-history features and their ecological implications by combining morphometric, biological, and reproductive data.

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Data Availability/Sharing: The datasets used and analyzed during the current study will be made available from the corresponding author upon a reasonable request.

Supplementary Information Availability: Not applicable.

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