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Life cycle and larval performance of the endangered pea blue butterfly, *Lampides boeticus* (Linneaus, 1767) (Lepidoptera: Lycaenidae) from Visakhapatnam, east coast of south India

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Abstract
This research presents the first in-depth exploration of the life cycle of the endangered pea blue butterfly, *Lampides boeticus* L., and examines larval performance, focusing on food intake, utilization, and the duration of the life cycle on the host plant *Crotalaria laburnifolia* L. The study was conducted in 2023 in Visakhapatnam, South India (17° 42' N and 82° 18' E). The findings indicate that the butterfly's life cycle spans 17 to 22 days, with an average of 20.20 ± 0.84 days. The life stages include an egg stage lasting 3 days, a larval stage of 7-9 days, and a pupal stage of 7-10 days. Nutritional indices were assessed across different instars, revealing Approximate Digestibility (AD) between 70.98% and 97.05%, Efficiency of Conversion of Digested food (ECD) from 4.84% to 22.10%, and Efficiency of Conversion of Ingested food (ECI) from 4.70% to 15.69% under controlled laboratory conditions of 28 ± 2°C and 80 ± 10% relative humidity. The high ECD and ECI values suggest that *Lampides boeticus* is well adapted to urban environments, contributing to its ecological success in Visakhapatnam.

Keywords: Life history, *Lampides boeticus*, captive rearing, immature stages, food utilization indices

Introduction

The genus *Lampides* Huebner is distinguished by having only one species, *Lampides boeticus*, commonly known as the pea blue in India and the long-tailed blue in Britain and Europe. This butterfly is a well-known migrant with larvae that primarily feed on a variety of plants within the Leguminosae family, particularly the Papilionoideae subfamily (David *et al.*, 2008) [8]. These larvae can become agricultural pests by feeding on the flowers and unripe seeds of green pods (Khalid, 2024) [15]. *Lampides boeticus* is recognized for its extensive distribution across the globe, being present in the Palaearctic region from Britain to Japan and occupying suitable habitats throughout Africa, Madagascar, Southeast Asia, and Australia. Its range also extends eastward to parts of Oceania, including Hawaii. The species thrives across various biomes, including temperate, subtropical, and tropical regions, both in lowland and mountainous areas, typically favoring open or disturbed environments (David *et al.*, 2008) [8].

In India, although *Lampides boeticus* is frequently cited as prevalent, its distribution is somewhat sporadic (Larsen, 1987) [16]. Furthermore, the species is listed as endangered under Schedule II of the India Wildlife Protection Act of 1972. To support effective breeding and rearing programs, as well as conservation efforts, it is crucial to have detailed knowledge of the butterfly's life history, immature stages, and specific habitat requirements (New *et al.*, 1995; Dennis *et al.*, 2003 [11], 2006 [10]; Dennis, 2010) [9]. This study seeks to contribute valuable information regarding the immature stages, larval performance on the host plant *Crotalaria laburnifolia* L., and the complete life cycle from egg to adult emergence for *Lampides boeticus* L.

Materials and methods

The present research was carried out in Visakhapatnam during the year 2023. Visakhapatnam, positioned at 17° 42' N latitude and 82° 18' E longitude on the eastern coastline of India, is part of the state of Andhra Pradesh. The reproductive behaviors of the pea blue butterfly (*Lampides boeticus*) were closely observed between 0800 and 1500 hours at two specific sites: the Andhra University campus and an adjacent Zoo Park area located 5 kilometers from the campus. When adult butterflies were spotted, detailed observations were made to document the duration of copulation and oviposition activities. Post-oviposition, leaves containing eggs were carefully collected and placed in Petri dishes (15 cm × 2.5 cm) for transport to the laboratory.

Once in the laboratory, these leaves were transferred to smaller Petri dishes (10 cm × 1.5 cm) lined with moistened blotting paper to maintain humidity and prevent desiccation. These dishes were then placed inside a spacious, clean cage covered with wire gauze. No additional protection against egg predation was necessary, as no ants were detected during the study. Egg hatching times were documented every six hours. Upon hatching, the larvae were transferred with care using a camel hairbrush to clean Petri dishes, also lined with moistened blotting paper. The larvae were provided daily with accurately measured portions of tender host plant leaves, and any leftover food along with feces was collected and weighed every 24 hours.

Larval development was closely monitored, recording changes in instar stages and corresponding characteristics such as length, width, and weight. The indices related to food utilization by the larvae were calculated following the methodology outlined by Waldbauer (1968) [34]. All parameters were measured using fresh weights and with five replications to ensure accuracy.

The transition from larva to pupa, along with the characteristics of the pupa—such as color, shape, size, weight, and the time required for adult emergence—was also meticulously recorded. Measurements were taken using millimeter graph paper for precision. Laboratory conditions were controlled to maintain a temperature of 28 ± 2°C and a

relative humidity of 80 ± 10%, with natural indirect sunlight ranging from 12 hours in November/January to 14 hours in June/July.

Both laboratory-emerged butterflies and those captured in the wild were utilized for describing the characteristics of adult butterflies.

Results

Adult Stage (Fig 1a)

Field Characteristics: The adult pea blue butterfly, with a wingspan ranging from 25 to 35 mm, exhibits a slender and short body, measuring 10.0-12.0 mm in length. The dorsal side of the body is dark brown, while the ventral side is white. A notable difference between males and females is in their dorsal coloration: males display a violet-blue color, whereas females are brown with a blue hue at the wing bases. Both sexes possess pale brown to white forewings and hindwings on the ventral side, accented with light to reddish-brown bands and a distinct white band along the outer margin of the hindwings. The hindwings also feature two black spots with an orange tint above and a greenish tint below, along with a delicate black tail tipped with white.

Behavior: Pea blue butterflies are adept fliers, capable of rapid wing beats and long-distance flights. They are often observed foraging for nectar among shrubs and basking in sunlight with wings partially open. Both males and females are frequently seen resting on the walls of university verandas.

Nectar Sources: These butterflies predominantly gather nectar from low-growing herbs and shrubs, but also from the small tree *Gliricidia sepium* during its blooming season, which extends from December to early February. Other nectar sources include *Antigonon leptopus*, *Borreria hispida*, *Corchorus acutangulus*, *Cosmos sulphureus*, *Duranta repens*, *Gisekia pharnaceoides*, *Gomphrena globosa*, *Ixora undulata*, *Justicia procumbens*, *Merremia tridentata*, *Portulaca quadrifida*, *Sida acuta*, *Sida cordifolia*, *Sida veronicaefolia*, *Tephrosia purpurea*, *Tribulus terrestris*, *Tridax procumbens*, *Triumfetta pentandra*, and *Vernonia cinerea*.

Oviposition Host Plants: The pea blue butterfly is known to lay eggs on various Fabaceae plants, with *Crotalaria laburnifolia* being identified as the most commonly used host plant for oviposition in this study.

Egg Stage (Fig 1b): Mating occurs during daylight hours and typically lasts about an hour. Females, after mating, deposit eggs singly on the sepals of flower buds and stalks, usually between 1100 and 1600 hours. Each flower bud may receive between 1 to 5 greenish-blue eggs, which gradually turn pale before hatching. The eggs, measuring 0.6-0.7 mm in diameter, are turban-shaped and hatch within three days. Upon hatching, the larvae do not consume the eggshell but instead bore into the flower buds to feed on the inner contents. The larval stage includes four molts and five instars, lasting 7-9 days.

Larval Stage (Fig 1c-g)

- **Instar I:** This stage lasts for about one day, and the larvae are barely visible to the naked eye.
- **Instar II:** Lasting for another day, larvae grow to 2.0-3.0 mm in length and 0.70-0.90 mm in width. They are brown with a mid-dorsal dark reddish-brown line and have black heads.
- **Instar III:** This stage also lasts a day, with larvae reaching 3.00-3.50 mm in length and 1.00-1.50 mm in width. They become woodlouse-shaped, greenish-brown, hairy, with a mid-dorsal reddish-brown line.
- **Instar IV:** Lasting 1-2 days, larvae grow to 5.00-8.00 mm in length and 1.80-2.50 mm in width, turning lemon yellow to pale green with clear segmentation and black lateral spots.
- **Instar V:** The final instar lasts 3-4 days, during which larvae grow to 8.00-15.00 mm in length and 4.20-5.00 mm in width. Their heads are brown, and their bodies are initially apple green, later turning cream. The lateral black spots also change to a cream color. A mid-dorsal reddish-brown line persists, and reddish-brown crossed lines appear laterally, eventually fading in color. The ventral side of the larvae is cream-colored.

No ant association with the larvae was observed

Pupal Stage (Fig 1h): The pupal stage spans 7-10 days. Pupae measure 10.0-11.0 mm in length and 3.5-5.0 mm in width. They are dorsally brown with dark brown spots and blotches, while the ventral side is light brown and plain. The pupal body, which is humped in the middle, is attached to the substrate by a loose body band and weighs approximately 85.5 mg.

Duration of Life Cycle: The entire life cycle of the pea blue butterfly ranges from 17 to 22 days, with the egg stage lasting 3 days, the larval stage 7-9 days, and the pupal stage 7-10 days.

Food Consumption, Growth, and Utilization: Newly hatched larvae bore into the buds of the host plant, primarily consuming the androecium and gynoecium. They move to new flowers as the inner contents of the previous ones are depleted. Molting is marked by the presence of shed skin. Interestingly, larvae do not enter new flowers until after they have molted.

Table 1: Food consumption, growth and food utilization efficiencies of *Lampides boeticus* larva fed with *Crotalaria laburnifolia buds*.

Instar number	Wt. of food Ingested (mg)	Wt. of Faeces (mg)	Wt. gained by larva (mg)	GR (mg/day/mg)	CI (mg/day/mg)	AD (%)	ECD (%)	ECI (%)
I	--	--	--	--	--	--	--	--
II	17.0 ± 01.60	0.5 ± 00.20	0.80 ± 0.15	0.57	12.14	97.05	04.84	04.70
III	20.0 ± 02.25	0.8 ± 00.85	1.00 ± 0.56	0.40	08.00	96.00	05.20	05.00
IV	100.0 ± 03.30	4.0 ± 01.10	10.80 ± 1.35	0.64	05.95	96.00	11.25	10.80
V	550.0 ± 18.72	159.6 ± 14.20	86.30 ± 5.65	0.37	02.38	70.98	22.10	15.69

Fig 1: (a) Adult pairing, (b) Eggs, (c) Instar I, (d) Instar II, (e) Instar III, (f) Instar IV, (g) Instar V, (h) Pupa

Table 1 provides data on food consumption and corresponding weight gain by larvae. Due to technical challenges, no data was collected for the first instar. However, as the larvae grew, their food consumption increased significantly. The proportion of food consumed by instars II to V accounted for 2.47%, 2.91%, 14.55%, and 80.05% of the total, respectively, with the final instar alone consuming 80% of the total food intake. This increase in food consumption was mirrored by a corresponding growth in larval weight, with the final instar accounting for over 87% of the total growth. Fig 2 illustrates the direct relationship between food consumption and weight gain by each instar.

While the values of the Consumption Index (CI) decreased progressively from instar II to V, an irregular pattern was observed in the values of the Growth Rate (GR) across the instars. CI values ranged from 2.38 to 12.14 mg/day/mg, with the highest value associated with the second instar and the lowest with the fifth instar. GR values varied between 0.37 and 0.64 mg/day/mg, with the highest value seen in the fourth instar and the lowest in the fifth instar. Additionally, Table 1 presents data on the Approximate Digestibility (AD), Efficiency of Conversion of Digested food (ECD), and Efficiency of Conversion of Ingested food (ECI). The AD values decreased from a high of 97.05% in the second instar to a low of 70.98% in the final instar, while ECD and ECI values increased progressively from the second to the final instar. The ECD values ranged from 4.84% to 22.10%, and ECI values from 4.70% to 15.69%, indicating an inverse relationship between AD and both ECD and ECI across the instars.

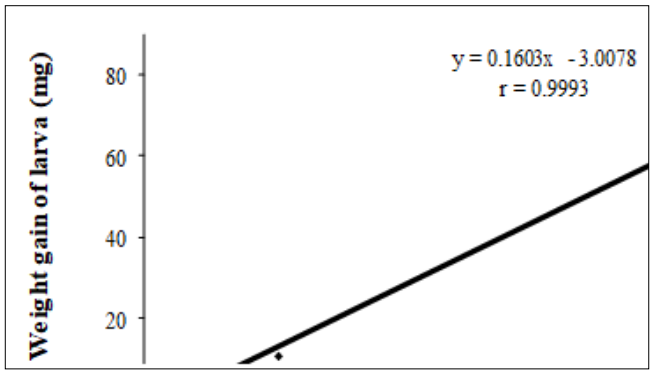

Fig 2: Relationship between food consumption and growth in *Lampides boeticus* on *Crotalaria laburnifolia*.

Discussion

The developmental cycle of the pea blue butterfly (*Lampides boeticus*), from egg to adult emergence, was observed to take approximately 20.20 ± 0.84 days under controlled laboratory conditions, where the temperature was maintained at around 28 ± 2°C. This duration is consistent with the relatively brief life cycles typical of tropical butterflies, as noted by Owen (1971) [21]. Since temperature plays a crucial role in the timing of each developmental stage (Pathak & Pizvi, 2003 [25]; Braby, 2003) [6], variations in ambient temperatures could lead to changes in the lifecycle length.

During its larval stage, an average of over 0.69 grams of leaf material was consumed, with the greatest intake occurring during the final two instars. This pattern of increased consumption during the later instars is characteristic of many lepidopteran larvae (Waldbauer, 1968 [34]; Mathavan & Pandian, 1975 [17]; Scriber & Slansky, 1981 [29]; Palanichamy *et al.*, 1982 [22]; Selvasundaram, 1992; Ghosh and Gonchaudhuri, 1996), compensating for the energy demands during the pupal stage when feeding does not occur (Pandian, 1973) [23]. The rate at which food is consumed is related to how efficiently the ingested material is converted into biomass (ECI), with higher consumption often correlating with lower conversion efficiency, and vice versa (Slansky & Scriber, 1985). This was evident in the second instar, where a consumption index (CI) of 12.14 mg/day/mg corresponded with low ECI values. Generally, growth rates tend to be higher in penultimate instars compared to the final instars, a trend also observed in the growth rates (GR) of *Lampides boeticus* (Scriber & Feeny, 1979) [28].

The Approximate Digestibility (AD) values obtained in this study align with those reported for other lepidopteran larvae, such as the range of 19-81% cited by Pandian and Marian (1986) [24] and 28.7-84.6% for *Pericallia ricini* (Ghosh and Gonchaudhuri, 1996). The high AD percentage of over 90.01% in this study supports the idea that foliage chewers often exhibit high digestibility, especially when their diet is rich in nitrogen and water (Pandian & Marian, 1986) [24]. Similar findings have been documented in studies involving *Pieris brassicae* (Yadava *et al.*, 1979) [35], *Ariadne merione merione* (Atluri *et al.*, 2010) [1], *Byblia ilithyia* (Bhupathi Rayalu *et al.*, 2011) [5], *Helicoverpa armigera* (Hemati *et al.*, 2012) [4], *Rathinda amor* (Bhupathi Rayalu *et al.*, 2012) [4], *Junonia iphita* (Bhupathi Rayalu *et al.*, 2013) [13], and *Spodoptera frugiperda* (Chaithra *et al.*, 2020) [7].

The observed increase in Efficiency of Conversion of Digested food (ECD) from early to late instars, as described

by Slansky & Scriber (1985) [31], was also evident in *Lampides boeticus*, with the lowest ECD recorded in the second instar and the highest in the fifth instar. Although ECD values were generally lower than AD values, this is not uncommon (Waldbauer, 1968) [34] and may indicate reduced efficiency in converting digested food into body tissues, potentially due to nutrient deficiencies or elevated metabolic energy demands (Bailey & Mukerji, 1976 [2]; Muthukrishnan, 1990) [19]. The pattern of Efficiency of Conversion of Ingested food (ECI) closely mirrored that of ECD, with values ranging from 4.70% to 15.69%, which is within the expected range for foliage chewers (1-78%) (Slansky & Scriber, 1985 [31]; Chaithra *et al.*, 2020 [7]; Hemati *et al.*, 2012) [14]. The similarity between the patterns of ECI and AD, as suggested by Waldbauer (1968) [34], and the relatively high ECD and ECI values in the final two instars, highlight the substantial tissue growth and ecological growth efficiency that enable *Lampides boeticus* to thrive in urban environments.

Conclusions

Research has consistently shown that poor nutrition during the larval stages can have detrimental effects on flight capabilities and wing development in butterflies, fruit flies, and moths (Portman *et al.*, 2015 [26]; Reim *et al.*, 2019 [27]; Yama *et al.*, 2019 [36]; Zahran *et al.*, 2018) [37]. Additionally, the dietary choices of larvae are known to play a crucial role in shaping the performance of adult butterflies (Ebada *et al.*, 2023) [12]. This study contributes essential insights into the nectar sources for adults, the host plants for oviposition, and the larval performance of the pea blue butterfly, particularly in terms of food consumption, growth, utilization, and the overall duration of its lifecycle. These findings are significant for the effective conservation and management of this endangered species in various settings, including parks, zoos, butterfly houses, and natural habitats. Butterfly houses, especially in zoos, are recognized as vital tools for both education and conservation (Mathew, 2001 [18]; Veltman, 2009) [33]. The study also suggests that maintaining larvae at a controlled temperature of around 28 ± 2°C in captivity can help ensure an adequate supply of adults, facilitating the restocking of areas where pea blue butterfly populations are low.

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