



Short Communication

A comprehensive study on relationship between *Euploea core* and *Nerium indicum* present in Fergusson College, Pune, India

Sameer Terdalkar¹, Snehangshu Das², Priyanka Patil³ and Minakshi Mahajan^{3*}

¹Department of Zoology, Fergusson College, Pune-411004, Maharashtra, India

²Department of Botany, Shivaji University, Kolhapur-416004, Maharashtra, India

³Department of Botany, Fergusson College, Pune-411004, Maharashtra, India
minakshi7mahajan@gmail.com

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Abstract

The life cycle of Danaidae butterfly *Euploea core core* Cramer (Lepidoptera: Rhopalocera: Danaidae) (Common Indian Crow) and its larval association with one of its known host plant, *Nerium indicum* (Gentianales: Apocynaceae), was investigated at Fergusson College campus by an effortful scientific observational approach by keeping them intact in their "chosen barrier free" natural habitat. Three particular sites were chosen where matured plants of *N. indicum* were already present. The morphological and detectable behavioural aspects of fourth and fifth instar larvae, pupae and the emergence of adult out of its pupal stage were observed in the selected study site at temperature ranging from 26°C-28.7°C. This study presents a pilot approach towards ovipositional behaviour of adult *E. core* in choosing a suitable site for laying eggs in the natural environment by studying the height, coordinates, daily weather conditions and neighbouring environmental factors. In addition to the lifecycle, the current study also adds a note on the larval food relationship with *N. indicum* by focusing on the sequestrational power of larvae of *E. core* to the harmful cardiac glycosides present as a defense mechanism in the foliage of Indian Oleander.

Keywords: *Euploea core*, oviposition, life cycle, sequestration, cardiac glycosides, *Nerium indicum*, larval relationship.

Introduction

The phytophilous insects such as butterflies also provide a pecuniary and ecological aids to the mankind. The adults and larvae of butterflies are dependent on greenery as they share a convoluted feeding liaisons with plants. A 'botanical instinct' is clearly shown by the larvae as they are customarily related with a definitive host and mainly feed on the on the foliar components of the greens¹.

With recent emerging demands for the conservation of insects booming with each season, the butterflies are now being contemplated to be an imperative bellwether for insect protection^{2,3}. This school of thought has driven many objective oriented human minds to record and describe the faunal diversity of their respective educational campus. The faunal and floral^{4,5} diversity of Fergusson college campus have been studied with much scientific zeal and precision and the 109 acre campus holds an array of fauna which includes 90 spider (28 families), 93 butterfly (6 families), 5 amphibian (3 families), 26 reptile (9 families), 137 bird (52 families) and 19 mammal (13 families) species⁶⁻⁸.

Till date there is only one well-documented list of butterfly diversity in campus which dates back to 1984 which showed the presence of 90 species belonging to 8 families⁹. Even in

discursive manner butterfly species were recorded from the campus area^{10,11}. An exclusive faunal review of the campus has so far reported 93 species, 53 genera of butterflies belonging to 6 families⁸.

According to most of the conservationists and butterfly ecologists, current insights of the literal requirements of different butterflies is severely sketchy in the country¹. Accordingly, the biological, ecological and conservational efforts are being contrived at Fergusson College to investigate the butterfly fauna present at the campus and its surroundings. The present study focuses on an innate alliance between the common crow butterfly-*Euploea core core* (Cramer) (Lepidoptera: Rhopalocera: Danaidae) with its larval host plant *Nerium indicum* (Mill.) with a description of its metamorphological pattern, larval food resources and how the larvae of *E. core* develops a definitive relationship with the toxic foliage of *N. indicum*.

Materials and methods

The study was conducted from the month of December 2017 to August 2018. The average temperature of the environment surrounding the *N. indicum* was recorded to be 27.25°C during the investigation period. The average humidity and wind velocity was measured to be 47.8% and 1.78 km/h during the

period of observation. Three observational sites were selected in the campus were the likelihood of the *E. core* selection of the plants for their oviposition would be greater. These sites were relatively marked alphabetically according to their occurrence in the campus as A, B and C respectively. All the three sites were kept under daily and hourly observation during daylight to note the behavioral pattern of *E. core* to choose only one favorable *N. indicum* plant for its oviposition. The coordinates and height of all the three plants have been noted down (Table-1). The coordinates were measured with the help of Garmin GPS Etrex-20X Receiver. The photographical observations of the larval stages were made only after the larvae reached its fourth larval instar stage as the investigation team was more focused on the study of “choosing pattern” of host plants by *E. core* in its natural habitat. It was ensured that no external harm occurred to the selected plants and individuals of butterflies during the tenure of observational period. The investigation team deliberately didn’t transfer the eggs to the laboratory for the ex-situ growth of the larvae of *E. core* by externally providing phytonutrition to the growing stages because a prominent and concrete observation of their natural behavior could only be made in the “barrier-free” nature.

Table-1: The height and coordinates of the plants.

Spot	Height (m.)	Coordinates (Location)
A	1.6	18°32'18.7"N; 73°63'43.3"E
B	2.5	18°33'46.7"N; 73°57'28.9"E
C	4.21	18°33'22.8"N; 73°60'39.8"E

Results and discussion

The selection of plant for ovi position from spot A was seen to be the highest and both spots B and C didn’t have a single egg or larvae or pupal stages present during the study period. The factors primarily responsible for selection of spot A could be due to the presence of plants under the shade with the last low intense rays of sun reaching the spot only during the twilight phase of the day and could also be due to the low height of the plants as compared to the plants from spot B and C. Other factors responsible could possibly depend on trophic interactions that includes the hypothesis of preference-performance, insect-insect probable competition and in addition to that it can also depend on non-trophic interactions which includes features like inter and intraspecific resource competition and “intraguild” predation¹².

The metamorphosis of *E. core* has been studied well with precise measurements of the different larval forms and the incubation time for their growth^{1,13-16}. The fourth instar and fifth instar stages (Figure-1) of 12 larval forms were recorded from the studied larval host plant^{1,17}. The freshly emerged fourth instar stage was present for 1-2 days. The larvae was dark brown in colour with white striated bars. A bright yellow patch

was seen. There was a sequential growth of tentacles which were fleshy and occurred in two doublets of which longer length was seen in the first duo. A blackish tinge, curved and upright tentacles were observed at its anterior end. And from the fourth instar larval stage, the growth rate seemed to be increased. The caterpillar started to adjust its point of recession, time and again on the ventral side of foliage of the host plant. Colour of the fifth instar larvae was grey and the duo of white striations of the body grew broader. The longitudinal patch of yellow colour decreased and formed spots that were found along the black spots. The rate of growth of larvae was faster than the previous stage. The fifth instar larval stage was present for 3-4 days.



Figure-1: X: fourth larval stage; Y: fifth the larval stage.

The matured caterpillar now starts looking for a suitable pupation site, it takes rest near on mature leaf’s lower surface, just near to its midrib. After a brief pause of 2-3 hours, the caterpillar starts secreting adhesive substances from its labial pore on the surface of the leaf. The larvae starts fixing its anal pro legs by hanging itself upside down with the secreted substances. The caterpillar which was matured its colours bleached which initiated the process of casting of molt. The whole process was completed in less than an hour. Initially, chrysalis’s colour was cream yellow. Within four days of pupation, it got converted into a dazzling golden colour. On the eight day of pupae, the chrysalis turned charcoal black and the matured wings of the butterfly being observed through the wall of chrysalis. This chrysalis stage lasted for 8 days. The measurements of the studied stages have been recorded Table-2.

Table-2: Measurements of the 12 studied larval forms and their chrysalis stages.

Stages of growth	Measurements (mm.)	
	Length	Breadth
Fourth Larval Instar stage	20.1 – 25.3 (22.85 ± 1.07)	2.6 – 2.9 (2.72 ± 0.62)
Fifth Larval Instar stage	37.3 – 41.1 (39.43 ± 1.31)	4.9 – 5.2 (5.11 ± 0.37)
Chrysalis	17.7 – 19.1 (18.42 ± 0.87)	6.9 – 9.4 (7.56 ± 0.66)

On the very early morning of ninth day of the matured pupae, the butterfly tries to break open the chrysalis by the force of wings and the juvenile wings are wet and small, still not ready for the flight motion. As usual butterflies pump some unknown fluids from its abdominal region through the veins present in its wings, which causes the expansion of the wings to their maximum size. Steadily, the wings gets dried up and butterfly exercises its flight muscles before its first fly in the environment¹⁸. The whole process of breaking open of the chrysalis to its first flight has been represented in a sequential manner (Figure-2). The whole life cycle was concluded in a span of 22 days. *Nerium indicum* (Mill.) (Hindi: "Kaner", commonly known as Indian Oleander) is a member of the Apocynaceae family which is perpetually green throughout the

year and shows a limited, obtusely radiating shrub habit. A whorled structured type of leaves are present which is leather like, slightly ovoid to entirely horizontal. The colours of the flowers ranges from pink to red and white and contains a beautiful aroma¹⁹. The *N. indicum* contains non digitalis cardiac glycosides in each and every component of itself. It comprises of neriodorin ($C_{22}H_{32}O_7$, m.p. 86-87°C), neriodorein ($C_{23}H_{34}O_{11}$, m.p. 106-107°C) and karabin ($C_{21}H_{49}O_6$) but the principle cardiac glycosides present in the leaves is oleanderin ($C_{32}H_{48}O_9$, m.p. 250°C) which is also an active principle of the leaves of *Nerium oleander*²⁰. Four hours after the ingestion, oleander poisoning can be visible through symptoms which are cardiac and gastrointestinal in nature²¹.



Figure-2: The series of photographs from (A-G) depicting the stages of matured butterfly break open its pupae.

Our observations suggest that growth of the caterpillar of *E. core* did not differ in feedback to cardiac glycosides present in the leaves which shows a considerable discernment in the uptake of cardiac glycosides into their tissues. According to the angle of plant defense mechanisms, cardiac glycosides which enter the caterpillar haemolymph may be the most germane because they are motile and can reach the neurological Na^+/K^+ -ATPase by the simple mechanism of diffusion²². The feeding mechanism of larvae of *E. core* suggests that its midgut epithelium is impervious to cardiac glycosides as its haemolymph analysis showed no microtraces of cardiac glycosides²³. The process of breakdown of cardiac glycosides in their body is still a mystery. Probably, the machinery of disintegration of harmful cardiac glycosides have led them successfully to have many larval host plants, showcasing an amazing evolutionary pattern for their survival in the present time.

Conclusion

The study of taxonomy of Lepidoptera always includes the list of ovi position host and forage plants of the species. But unfortunately, the studied data are generally not specified as a “taxonomic index”²⁴. The trend of adjustment to endure phyto-toxins have always been stated as an “evolutionary origin” and these traits have indeed been explained as a coevolutionary transitions to phyto-defence mechanisms²⁵⁻³⁰. The evolutionary mechanism of phyto-toxin in butterflies are yet to be studied deeply with a comprehensive analysis of internal biochemical machinery that provides sequester rational power to the Lepidopterans. Rather than a laboratory approach to study and incubate the butterflies with an artificial or natural food, researches should be carried out in their natural habitat by setting up laborious and scientific field observation methods to carefully observe their behavior in their own home. The need of the hour is to not only to merely study the lifecycles of various butterflies but also to provide an extensive analysis of the development of the relationship between its ovi positional or/and food plant and Lepidopterans by the means of biochemical ecological studies.

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References

1. Chandar G.S., Rao K.E. and Atluri J.B. (2016). Metamorphosis of *Euploea core* (Common Crow) on *Nerium indicum* (Mill.) at Andhra University Campus, Visakhapatnam. *World Journal of Pharmacy and Pharmaceutical Sciences*, 5(9), 1435-1450.
2. New T.R., Pyle R.M., Thomas J.A., Thomas C.D. and Hammond P.C. (1995). Butterfly conservation management. *Annual review of entomology*, 40(1), 57-83. <https://doi.org/10.1146/annurev.en.40.010195.000421>
3. Smetacek P. (1996). Restoring past glory. *Santuary Asia*, 16(6), 26-29.
4. Vartak V.D. (1958). The flora of the Fergusson College campus, Poona dist. *Fergusson College Magazine*, 50(2), 7-11.
5. Nerlekar A.N., Lapalikar S.A., Onkar A.A., Laware S.L. and Mahajan M.C. (2016). Flora of Fergusson College campus, Pune, India: monitoring changes over half a century. *Journal of Threatened Taxa*, 8(2), 8452-8487. <http://dx.doi.org/10.11609/jott.1950.8.2>.
6. Nerlekar A.N., Gowande G.G. and Joshi P.S. (2014). Diet of the Spotted Owlet *Athene brama* in an urban landscape. *Indian BIRDS*, 9(2), 45-48.
7. Nerlekar A.N., Gowande G.G. and Joshi P.S. (2014). Behavioural ethogram of Spotted Owlet *Athene brama* (Temminck, 1821). *Journal of the Bombay Natural History Society*, 111(3), 172-179. <http://dx.doi.org/10.17087/jbnhs/2014/v111i3/82356>
8. Nerlekar A.N., Warudkar A.M., Gowande G.G., Salve S. S., Raut A., Patankar S.R. and Nalavade S.B. (2016). A review of the faunal diversity of the Fergusson College campus, Pune, India. *ZOO's PRINT*, 31(10), 4-25.
9. Kumar N. (1984). A Checklist of the butterflies of Fergusson College. *Fergusson College Magazine*, 75, 14-17.
10. Chhaya K., Mujumdar N., Mhaske P. and Patwardhan A. (2012). A new larval host record for the Pea Blue butterfly *Lampides boeticus* (Linnaeus) (Insecta: Lepidoptera: Lycaenidae) from Pune, Maharashtra, India. *Bugs R ALL Newsletter of the Invertebrate Conservation & Information Network of South Asia*, 19, 6-9.
11. Warudkar A. and Patankar S. (2013). Butterflies in Fergusson College. *Fergusson College Magazine*, 104, 3-4.
12. Shiojiri K., Sabelis M. and Takabayashi J. (2015). Oviposition preference of cabbage white butterflies in the framework of costs and benefits of interspecific herbivore associations. *Royal Society open science*, 2(12), 150524.
13. Saikia K., Kalita J. and Saikia P.K. (2010). Biology and life cycle generations of common crow-*Euploea core core* Cramer (Lepidoptera: Danainae) on *Hemidesmus indica* host plant. *Int J NeBio*, 1(3), 28-37.
14. Wynter-Blyth M.A. (1957). Butterflies of the Indian region. Today and Tomorrow's Printers and Publications, New Delhi, 523.
15. Varshney R.K. (1993). Index Rhopalocera Indica Part III. Genera of Butterflies from India and neighbouring

- countries Lepidoptera: (A) Papilionidae, Pieridae, and Danaidae). *Oriental Insects.*, 27, 347-372.
16. Sidhu A.K., Kaur M. and Rose H. (2007). Life History of The Common Indian Crow *Euploea Core* (Cramer) (Nymphalidae: Lepidoptera: Insecta). *Rec. zool. Surv. India*, 107(Part-1), 119-123.
 17. Kunte K. (2000). India, a Lifescape: Butterflies of Peninsular India. Universities Press Pvt. Ltd., Hyderabad, 149. ISBN 8173713545
 18. Metamorphosis (2018). Butterfly school. <http://www.butterfly-school.org/new/meta.html>. Accessed on 03/09/2018.
 19. Frohne D. and Pfänder H.J. (1984). A colour atlas of poisonous plants. Wolfe Publishing Ltd. London, 190.
 20. Deshaprabhu S.B. (1966). The Wealth of India – Raw Materials VII (N-Pe). CSIR, New Delhi, 15-17. ISBN: 81-85038-00-7
 21. Behcet Al., Yarbil P., Dogan M., Kabul S. and Yildirim C. (2010). A case of non-fatal oleander poisoning. *BMJ Case Reports*, 2010. bcr02.2009.1573. <http://doi.org/10.1136/bcr.02.2009.1573>
 22. Petschenka G., Offe J.K. and Dobler S. (2012). Physiological screening for target site insensitivity and localization of Na⁺/K⁺-ATPase in cardenolide-adapted Lepidoptera. *Journal of insect physiology*, 58(5), 607-612.
 23. Petschenka G. and Agrawal A.A. (2015). Milkweed butterfly resistance to plant toxins is linked to sequestration, not coping with a toxic diet. *Proceedings of the Royal Society B: Biological Sciences*, 282(1818), 20151865. <http://doi.org/10.1098/rspb.2015.1865>
 24. Downey J.C. (1962). Host-plant relations as data for butterfly classification. *Systematic Zoology*, 11(4), 150-159. <https://www.jstor.org/stable/2411462>
 25. Krieger R.I., Feeny P.P. and Wilkinson C.F. (1971). Detoxication enzymes in the guts of caterpillars: an evolutionary answer to plant defenses?. *Science*, 172(3983), 579-581.
 26. Dussourd D. and Denno R. (1994). Host Range of Generalist Caterpillars: Trenching Permits Feeding on Plants with Secretory Canals. *Ecology*, 75(1), 69-78.
 27. Jongsma M.A. and Bolter C. (1997). The adaptation of insects to plant protease inhibitors. *Journal of Insect Physiology*, 43(10), 885-895.
 28. Engler H.S., Spencer K.C. and Gilbert L.E. (2000). Insect metabolism: Preventing cyanide release from leaves. *Nature*, 406(6792), 144. <https://doi.org/10.1038/35018159>
 29. Wittstock U., Agerbirk N., Stauber E.J., Olsen C.E., Hippler M., Mitchell-Olds T., Gershenzon J. and Vogel H. (2004). Successful herbivore attack due to metabolic diversion of a plant chemical defense. *Proceedings of the National Academy of Sciences of the United States of America*, 101(14), 4859-4864. <http://doi.org/10.1073/pnas.0308007101>
 30. Singer M.S. and Stireman III J.O. (2005). The tri-trophic niche concept and adaptive radiation of phytophagous insects. *Ecology Letters*, 8(12), 1247-1255. <https://doi.org/10.1111/j.1461-0248.2005.00835.x>