



Influence of Developmental Stage and Seasonal Variations on the Prevalence of Gregarine Infection in *Trilophidia annulata*.

Dongare SP¹, Kanse VS² and Chondekar RP^{1*}

¹Department of Zoology, Dr. Babasaheb Ambedkar Marathwada University, Chh. Sambhajinagar- 431004

²Department of Zoology, Deogiri College Chh. Sambhajinagar

*Email: sam_chondekar@yahoo.com

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ABSTRACT

Gregarines as a common Apicomplexan parasite plays an important role in shaping the dynamics of insect populations. This study investigates the impact developmental stage of grasshopper along with seasonal variations on the prevalence and intensity of gregarine parasitism in have to get more awareness about their host parasitic relationship. *Trilophidia annulata* specimens were collected from the campus of Dr. Babasaheb Ambedkar Marathwada University, Chhatrapati Sambhajinagar, Maharashtra. The grasshoppers were classified into two groups based on developmental stages: nymphs and adults. The digestive tracts of the specimens were microscopically examined to detect the presence of gregarine parasites. Statistical analysis was conducted to assess the influence of grasshopper developmental stage on the prevalence and intensity of parasitism. A total of 341 adult grasshoppers were analysed, with 60.11% found to be infected. While a total of 429 nymph samples were collected and examined under a microscope. About 71.79% of them were found to be infected. It is also observed that grasshoppers are more infected in monsoon than in winter season. Understanding the role of factor like developmental stage and seasonality in gregarine parasitism will helps us in maintaining pests ecological and economical balance. Nymphs exhibited higher infection rates compared to adults, suggesting a developmental susceptibility to gregarine parasites. These results contribute to understanding the ecological dynamics of parasitism in grasshopper populations and highlight the need for further research on the factors driving these patterns in the context of regional environmental conditions. Our findings indicate significant variation in parasitism rates between different developmental stages of grasshoppers. Understanding these dynamics is crucial for managing grasshopper populations and mitigating their impact on agriculture in the Marathwada region.

Keywords: Grasshoppers, *Trilophidia annulata*, Gregarines, Developmental stage, Nymphs, Adults, Prevalence.

INTRODUCTION

Grasshoppers are significant herbivorous insects that influence both natural and agricultural ecosystems, often causing substantial crop damage during population outbreaks (Joern & Gaines, 1990). Their population dynamics are shaped by various biotic factors, among which parasitism by gregarines, a group of Apicomplexan protozoans, plays a critical role (Clopton, 2002). Gregarines primarily inhabit the digestive tracts of their hosts, impacting host health, reproduction, and survival, thereby influencing population dynamics (Vega & Kaya, 2012). Despite their ecological importance, the factors driving gregarine parasitism, particularly the influence of host developmental stage, remain underexplored in many regions, including the semi-arid Marathwada region of Maharashtra, India. The developmental stage of an insect host is a key determinant of parasitic infection rates due to physiological and immunological changes across life stages (Sheldon & Verhulst, 1996). Nymphs and adults differ in their susceptibility to parasites, with nymphs often exhibiting higher infection rates due to weaker immune responses or increased feeding activity (Zuk & McKean, 1996). Additionally, seasonal variations, such as those driven by monsoon and winter conditions, can influence parasite prevalence by altering host physiology, behaviour, or environmental conditions conducive to parasite transmission (Altizer et al., 2006). Recent studies have emphasized the role of climatic factors, such as humidity and temperature, in modulating parasite life cycles and infection rates in insect populations (Smith et al., 2023; Patel & Kumar, 2024).

This study focuses on *Trilophidia annulata*, a prevalent grasshopper species in Marathwada, to investigate the impact of developmental stage (nymph vs. adult) and seasonal variations on the prevalence and intensity of gregarine infections. Results revealed an overall gregarine prevalence of 60.11% in adults and 71.79% in nymphs, with nymphs showing higher infection rates, suggesting developmental susceptibility. A study conducted in Western Nebraska found that early instar nymphs of grasshoppers exhibited higher rates of gregarine infection than later instars or adults, which may be attributed to increased exposure to parasite oocysts while actively foraging in thick vegetation (Creigh et al., 2018). Seasonal analysis indicated higher prevalence during the monsoon season (65.73% for adults and 77.31% for nymphs) compared to winter

(53.98% for adults and 66.19% for nymphs). These findings align with recent research highlighting the influence of seasonal climatic variations on parasite transmission in insects (Kumar et al., 2023; Garcia & Martinez, 2024).

Understanding how developmental stages affect gregarine parasitism in grasshoppers, such as *Trilophidia annulata*, is essential for elucidating host-parasite interactions and their implications for pest management in agricultural landscapes. This study focuses on *Trilophidia annulata*, a common grasshopper species in the Marathwada region, to investigate the impact of developmental stage (nymph vs. adult) on the prevalence and intensity of gregarine infections. By examining grasshoppers collected from the campus of Dr. Babasaheb Ambedkar Marathwada University, Chhatrapati Sambhajinagar, this research aims to provide insights into the ecological dynamics of parasitism and contribute to strategies for managing grasshopper populations in the region. The findings are expected to highlight the role of developmental susceptibility in shaping parasitism patterns and underscore the need for further studies under varying environmental conditions.

MATERIAL AND METHODS

Insects were periodically collected during the morning hours from their natural habitats, such as farms, grasslands, and gardens in different localities of Dr. Babasaheb Ambedkar Marathwada University campus, in Chhatrapati Sambhajinagar. Grasshoppers were readily found on the leaves of crop plants and grasses, making insect collecting nets the preferred method for bulk collection, though sometimes hand picking was also employed. Collected insects were placed in ventilated plastic containers, maintaining their natural environment by providing their usual food sources to keep them alive until examination.

Then host is dissected and its gut is removed carefully. This fresh material placed on clean glass with a drop of distilled water. A thin film or smear was taken on slide covered with cover slip for examination of live protozoans under the light microscope. Live digital photography of observed parasites is done to record the prevalence of infection.

RESULTS

Six-month study period was undertaken from July to December 2024 to observe the influence of seasonal variations on gregarine prevalence in *Trilophidia annulata*.

1. Prevalence for *Trilophidia annulata* (adults):

- Total host examined: 341
- Positive for gregarine infection: 205
- Prevalence rate: 60.11%

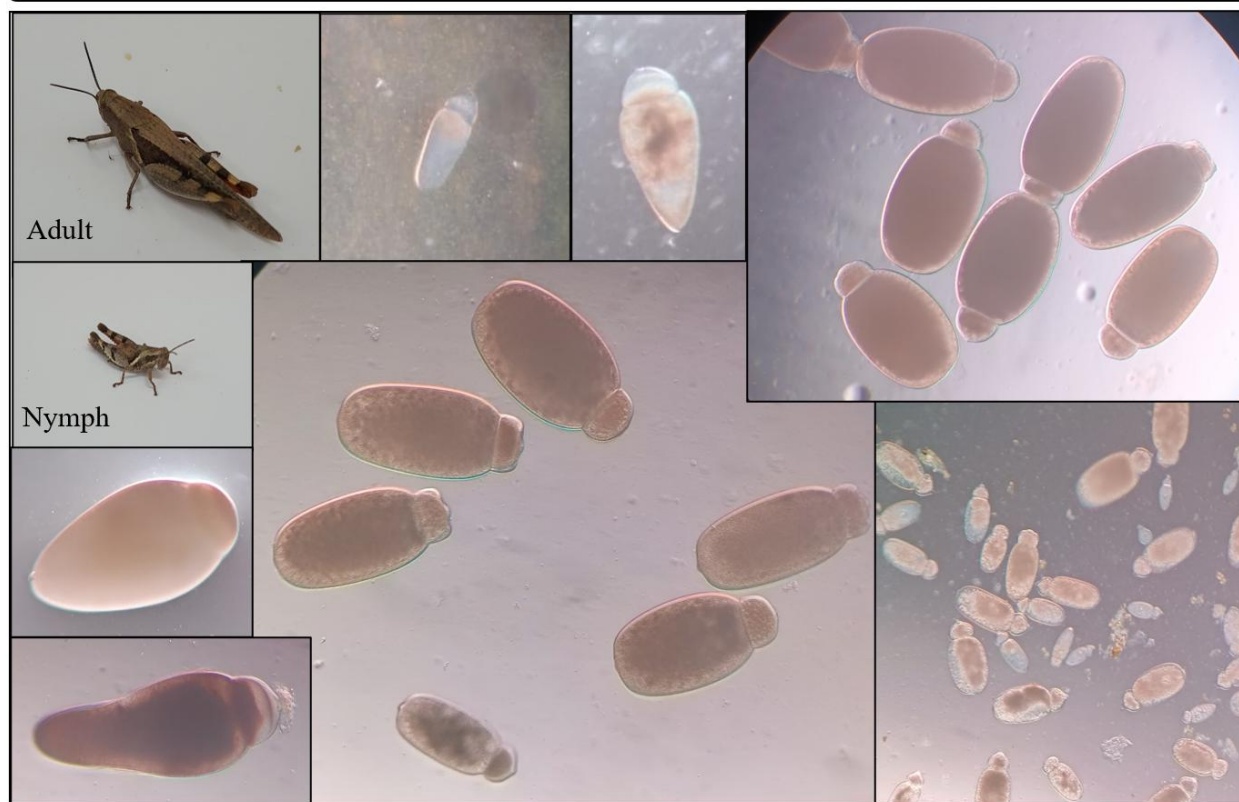
During the study period month wise analysis of gregarine parasitism in adults, showed the maximum percentage of prevalence during September 2024 i.e. 71.23% and the minimum percentage of prevalence is in December 2024 i.e. 49.05%.

Seasonal variations in occurrence of Gregarines in *Trilophidia annulata* (adults)-

- Monsoon (July-September 2024):
 - Total number of hosts examined: 178
 - Number of hosts infected with gregarines: 117
 - Prevalence: 65.73%
- Winter (October-December 2024):
 - Total number of hosts examined: 163
 - Number of hosts infected with gregarines: 88
 - Prevalence: 53.98%

The prevalence of gregarines parasitism in adults was higher in Monsoon (65.73%) and lower in Winter (53.98%). It indicates the effect of seasonal variations on gregarines parasitism in the adults, with the more prevalence occurring during the months of Monsoon as compare to winter.

Different life cycle stages of various gregarine parasite species found in *Trilophidia annulata*

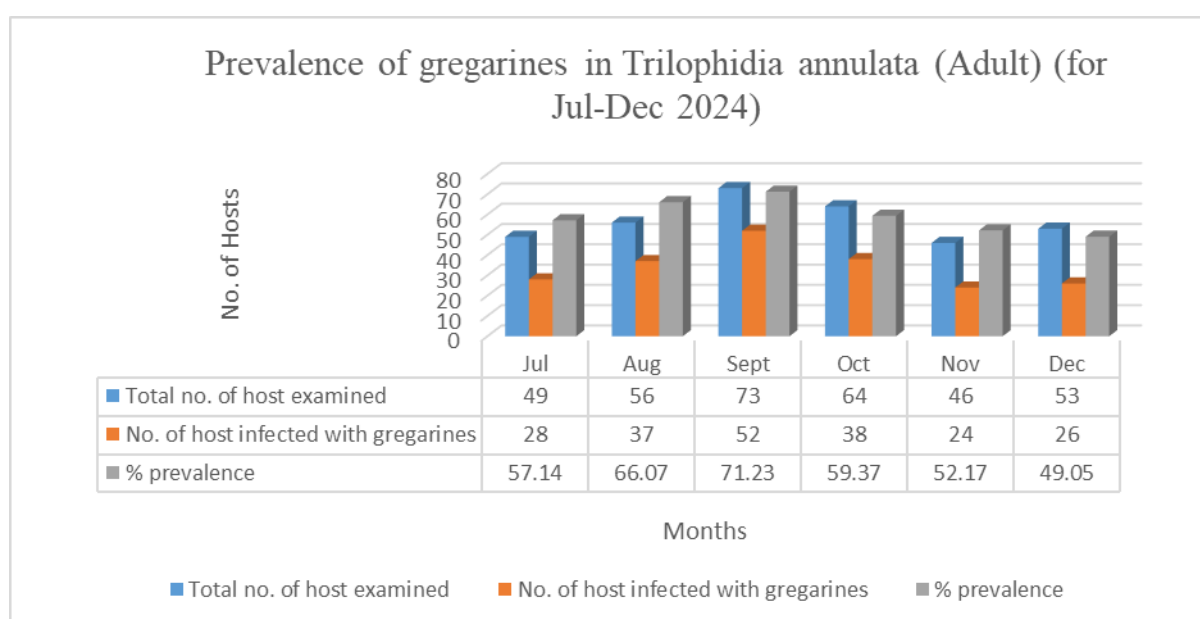


Parasites are observed under 10X x 10X magnification

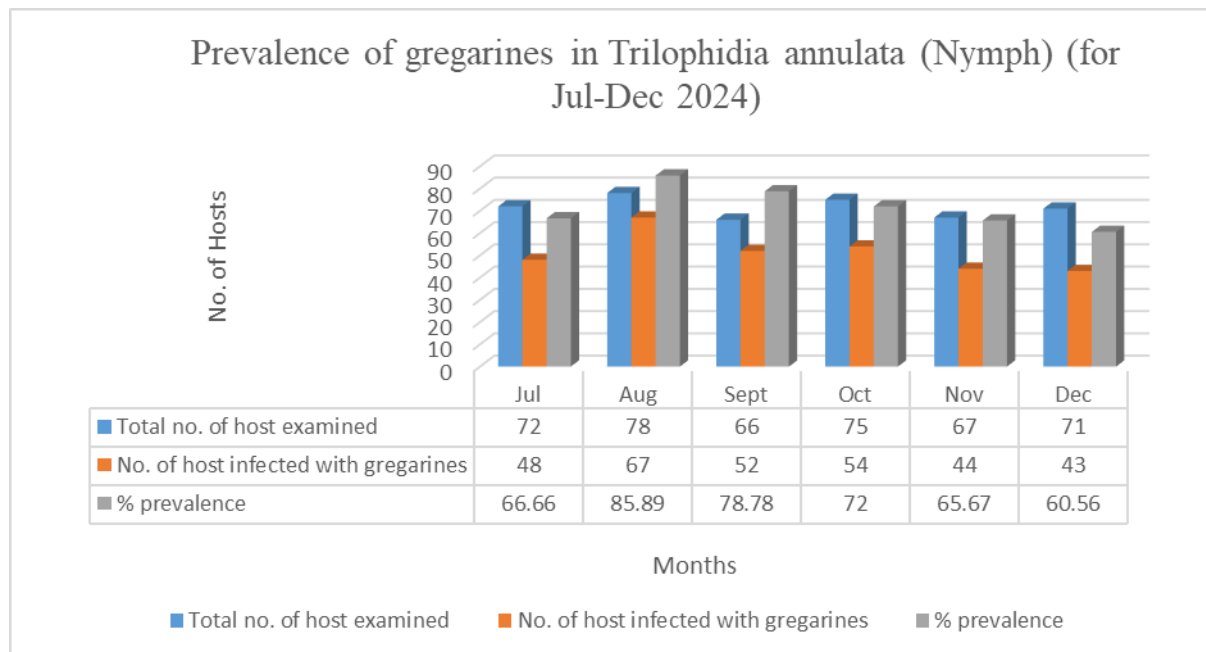
Photo plate 1. Different life cycle stages of gregarines parasitising *Trilophidia annulata* (Adult & Nymph)

Monthly Breakdown for Jun-Dec 2024:**Table 1.** showing the prevalence of gregarine parasites in *Trilophidia annulata* (Adult) (for Jul-Dec 2024)

Duration 2024	Total no. of host examined	No. of host infected with gregarines	% prevalence
July	49	28	57.14
August	56	37	66.07
September	73	52	71.23
October	64	38	59.37
November	46	24	52.17
December	53	26	49.05
Total	341	205	60.11

**Graph 1.** Showing the month wise prevalence of gregarines in *Trilophidia annulata* (Adult) (for Jul-Dec 2024)**Table 2.** showing the prevalence of gregarine parasites in *Trilophidia annulata* (Nymph) (for Jul-Dec 2024)

Duration 2024	Total no. of host examined	No. of host infected with gregarines	% prevalence
July	72	48	66.66
August	78	67	85.89
September	66	52	78.78
October	75	54	72.00
November	67	44	65.67
December	71	43	60.56
Total	429	308	71.79



Graph 2. Showing the month wise prevalence of gregarines in *Trilophidia annulata* (Nymph) (for Jul-Dec 2024)

2. Prevalence for *Trilophidia annulata* (Nymphs):

- Total host examined: 429
- Positive for gregarine infection: 308
- Prevalence rate: 71.79%

During the study period month wise analysis of gregarine parasitism in nymphs, showed the maximum percentage of prevalence during August 2024 i.e. 85.89% and the minimum percentage of prevalence is in December 2024 i.e. 60.56%.

Seasonal variations in occurrence of Gregarines in *Trilophidia annulata* (Nymphs) -

- Monsoon (July-September 2024):
 - Total number of hosts examined: 216
 - Number of hosts infected with gregarines: 167
 - Prevalence: 77.31%
- Winter (October-December 2024):
 - Total number of hosts examined: 213
 - Number of hosts infected with gregarines: 141
 - Prevalence: 66.19%

The prevalence of gregarines parasitism in nymphs was higher in Monsoon (77.31%) and lower in Winter (66.19%). It indicates the effect of seasonal variations on gregarines parasitism in the nymphs, with the more prevalence occurring during the months of Monsoon as compare to winter

CONCLUSIONS

This study provides significant insights into the influence of developmental stage and seasonal variations on gregarine parasitism in *Trilophidia annulata* in the Marathwada region of Maharashtra. The findings demonstrate that nymphs exhibit a higher prevalence of gregarine infections compared to adults highlighting developmental susceptibility likely driven by physiological and immunological differences. Seasonal analysis further revealed that gregarine prevalence was significantly higher during the monsoon season for both adults and nymphs. These patterns suggest that environmental conditions, such as increased humidity and temperature during the monsoon, enhance gregarine transmission and infection rates. However, this study also highlights the need for further research to explore additional factors, such as specific environmental variables (e.g., temperature, rainfall) and host immune responses, that may drive gregarine parasitism. Long-term studies across multiple years and regions could provide a more comprehensive understanding of these dynamics. Ultimately, this research contributes to the broader knowledge of insect-parasite ecology and supports the development of sustainable agricultural practices in semi-arid ecosystems like Marathwada. The observed variations in parasitism across developmental stages and seasons underscore the complex interplay between host biology and

environmental factors in shaping host-parasite interactions. These findings have important implications for managing grasshopper populations in the Marathwada region, where *T. annulata* can contribute to agricultural losses. By identifying periods of heightened parasitism, particularly during the monsoon, and the greater vulnerability of nymphs, targeted pest management strategies can be developed to mitigate crop damage while considering ecological balances.

Conflict of Interest: The authors declare no conflict of interest in relation to this research.

Data Availability Statement: Not applicable.

Correspondence and requests for materials should be addressed to **Dongare S.P.**

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