



FUNCTIONAL RESPONSE AND PARASITISM BY *COTESIA SESAMAE* (CAMERON) ON LARVAE OF *TARUCUS ROSACEA* (AUSTAUT) FROM IRAQ

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ABSTRACT

This study evaluated the biocontrol potential of the parasitoid *Cotesia sesamae* (Cameron) on jujube blue butterfly, *Tarucus rosacea* (Austaut), under field and laboratory conditions in Iraq, from September 2023 to May 2024. Field surveys recorded natural parasitism rate of 85.7% (± 5.2). A strong density-dependent relationship was observed ($R^2 = 0.9276$). Laboratory studies revealed a clear host stage preference, with the highest parasitism occurring in the fourth larval instar. Functional response analysis indicated that a Holling Type III model best described the parasitoid's behaviour, with an adaptive attack rate of 0.139 and a handling time of 0.043, demonstrating efficient host searching and adaptive foraging. These results strongly support the incorporation of *C. sesamae* in a biocontrol based IPM program.

Key words: Biological control, Braconidae, functional response, *Cotesia sesamae*, koinobiont, *Tarucus rosacea*, parasitism, host stage preference, *Ziziphus spina-christi*

The jujube tree, *Ziziphus spina-christi* (L), is known from arid and semi-arid ecosystems across the Middle East and Africa. Its productivity and health are frequently threatened by herbivorous insects, among which the jujube blue butterfly, *Tarucus rosacea* (Austaut) (Lepidoptera: Lycaenidae) is a prominent pest. Its larvae caused considerable damage by feeding on young leaves and floral buds, leading to reduced photosynthetic capacity and significant yield losses, thereby compromising the tree's overall vitality (Alle et al, 2024). Using insecticides causes many ecological hazards and more sustainable and ecologically sound IPM strategies are necessary, of which the biological control is a major one (Krishnamoorthi et al. 2024). Parasitoid wasps of the family Braconidae, Subfamily Microgasterinae, particularly those in the genus *Cotesia*, are among the most effective and widely studied agents in biological control (Pennacchio and Strand, 2006). Recent field surveys have identified *Cotesia sesamae* (Cameron) as a key natural enemy of *T. rosacea* (Al-Jorany, 2018). To properly assess its potential, it is crucial to quantify its parasitic capabilities. Key parameters for such an evaluation include host stage preference and the functional response, which describes how a parasitoid's attack rate changes with host density (Nonaka and Kaitala, 2020). This study provides the first comprehensive assessment of *C. sesamae* as a biocontrol agent against *T. rosacea*. Objectives include: confirming species identity; seasonal dynamics and field

parasitism rates; determine host instar preference under laboratory conditions; and characterize its functional response.

MATERIALS AND METHODS

Initial populations of *T. rosacea* and its parasitoid, *C. sesamae*, were sourced from larvae collected from jujube trees *Ziziphus spina-christi* of Basra, Iraq, Al-Jabbasi (30.65°N, 47.65°E), and Abu Al-Khasib (30.45°N, 47.98°E). Parasitoid identity was confirmed using the keys of Wilkinson (1932) and the Wasp Web database (Mifsud et al. 2019). Both insect colonies were maintained at 26 \pm 2°C, 65 \pm 5% RH, and a 14:10 h (L:D) photoperiod. The *T. rosacea* colony was reared on fresh jujube leaves, while the *C. sesamae* culture was sustained by weekly exposure to fourth-instar host larvae. All experiments used 2- to 3-day-old, mated female parasitoids, pre-conditioned by 24 hr of host deprivation. Regular field surveys assessed the dynamics of natural populations and the rate of parasitism from September 2023 to May 2024, with weekly visits to each site. During each visit, randomly selected trees were examined, and the number of larvae present on 3 branches from each tree was recorded. Larval samples were collected and reared in the laboratory to determine the natural parasitism rate. To identify the preferred host instar (2nd, 3rd, 4th, or 5th), a single female wasp was exposed to 20 larvae of a single instar in a Petri dish for 24h. Larvae were then reared individually to

calculate the parasitism % (Ehteshami et al., 2023). The experiment was replicated ten times for each instar in a completely randomized design (CRD). Parasitism data were arcsine-transformed and analyzed using ANOVA followed by LSD test in SPSS v26. Using the preferred instar from the first experiment (host stage preference), a single female wasp was introduced into an arena with one of five host densities (2, 4, 8, 16 and 32) for 24 hr (n=5 replicates). The number of parasitized hosts was determined by dissecting all larvae after exposure, in addition to the physiological changes that occur in the larvae of the parasite, with corresponding controls. The model Holling's Type II and Type III which apply to most parasitoids were used (Papanikolaou et al., 2021).

RESULTS AND DISCUSSION

Morphological examination confirmed its identification as *Cameron sesamiae* (Cameron, 1906) (= *Apanteles sesamiae* Cameron, 1906) as documented by Wilkinson (1932) and the Wasp Web database (Mifsud et al. 2019) (Fig. 1). This genus is characterized by the presence of a specific pattern on the first segment of the abdomen (propodeum), called the areola, which is an important diagnostic feature. Many (but not all) Cotesia species have bicolored legs, where the base (coxa and femur) is dark (black or dark brown), while the ends (tibia and tarsus) are light (yellow or light brown). General body colour tends to be entirely black or with brown markings. Cocoon: The parasitic larva often (but not always) spins a silken cocoon that is distinctively white, light yellow, or golden. Observations from field surveys as given in Fig. 2 show a clear pattern of population dynamics showing that the infestations began in late September and continued until the end of May, with peak activity occurring from October to April. The results revealed a strong relationship between

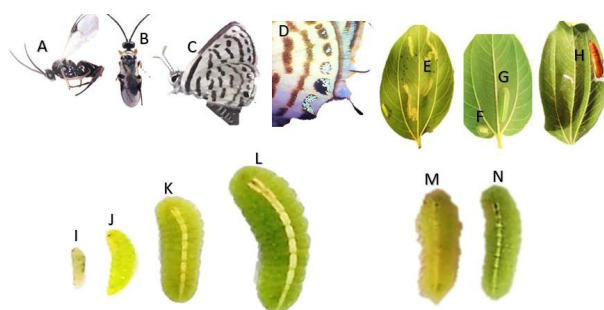


Fig. 1A= Lateral view of the parasitoid, B= Dorsal view of the parasitoid, C= Lateral view of the adult host, D= Wing spot patterns, E= Symptoms of infestation, F= Parasitoid pupa, G= Host larva, H Host pupa, I- L= Larval instars (1-4), M= Parasitized larva, N= Healthy larva.

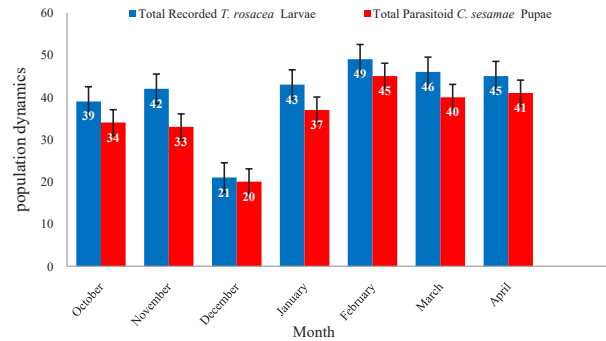


Fig. 2. population dynamics between *T. rosacea* and *C. sesamiae*

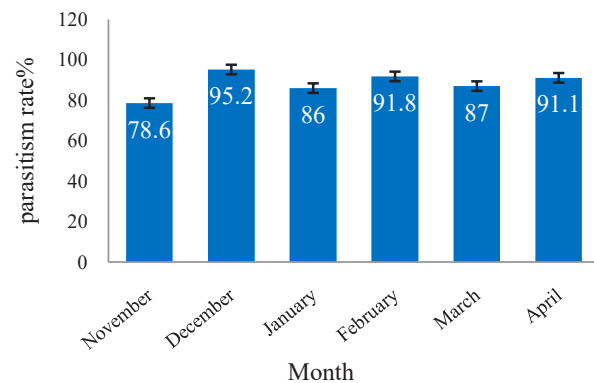


Fig. 3. Monthly parasitism rate% of *C. sesamiae* on *T. rosacea*

host larval density and parasitoid activity. The number of infested larvae on leaves ranged from 28-86 larvae/ three examined branches, while the number of observed parasitoid pupae ranged from 5-22/ site. Thus, revealing high natural parasitism rate (80-90%), and showing parasitoid's effective role (Fig. 3). Parasitism rate was remarkably high and stable throughout the season $-85.7 \pm 5.2\%$; far exceeding the threshold required for successful biological control; and relationship between the parasitoid and the host was highly significant ($R^2 = 0.9276$, Fig. 4). Parasitism rates varied significantly

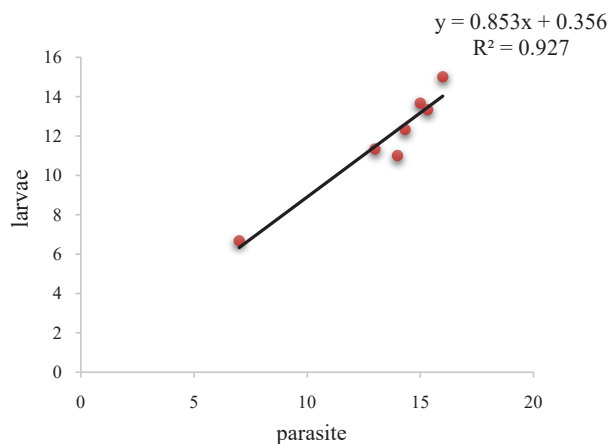


Fig. 4. Correlation- *T. rosacea* vs *C. sesamiae*

among host instars ($p < 0.05$); fourth instar was the most preferred (89.3%, followed by the third (76.2%); second and fifth instars were less preferred (Fig. 5).

Functional response when analysed reveal that it increased with larval abundance; host density and parasitism by *C. sesamae* analyzed using Holling's functional response model revealed that both models (Type II and Type III) were suitable, but the statistical fit indicated that the Type III model was superior (Fig. 6); adaptive attack rate (Type III) shown was = 0.139 and handling time T_h was =0.043. Sigmoidal response curve shows that the parasite's efficiency. gradually increases with host density, reflecting an adaptive foraging behaviour. Eight-months field study confirmed a highly effective and stable host-parasitoid relationship in the Basra ecosystem (high parasitism rate of $85.7 \pm 5.2\%$ (Fig. 3), significantly exceeding the 50-60% threshold required for effective biological control (Van Driesche et al., 2010; Blazyte-Cereskiene et al.,2022). This is higher than rates reported for other *Cotesia* spp. (Wang et al., 2023). This exceptional performance indicates that *C. sesamae* acts as a dominant mortality factor for *T. rosacea* populations. The strong positive correlation ($R^2 = 0.9276$) between host and parasitoid densities

confirms a density-dependent relationship, which is crucial for natural pest regulation. This also suggests that *C. sesamae* possesses efficient host-searching capabilities (Holling, 1959). Mechanistic insights from the functional response analysis show that the superior fit of the Type III model (Fig. 6); with an adaptive attack rate of 0.139 and handling time of 0.043, indicates sigmoidal foraging behaviour (Fernández-Arhex and Corley, 2003; Aguirre et al., 2024). This adaptive response, where parasitism efficiency increases with host density up to a threshold, aligns with field observations of high parasitism rates and contrasts with the Type II model's decreasing efficiency at high densities (Li et al., 2024; Saini and Sharma, 2018; Clemente et al.,2024). These findings support the incorporation of this parasitoid into conservation biological control strategies (Gurr et al., 2017). *Cotesia sesamae* is a highly efficient and specific natural enemy of *T. rosacea*, and it is recommended for inclusion in IPM programs (Fortuna et al., 2024).

AUTHOR CONTRIBUTION STATEMENT

H A M and M A A conceived and designed the research. H AM conducted the experiments and performed data collection. M A A supervised the project and provided critical input. H A M and M A A analyzed the data. R M K wrote the initial draft of the manuscript. M A A reviewed and edited the manuscript. All authors read and approved the final manuscript.

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CONFLICT OF INTEREST

No conflict of interest.

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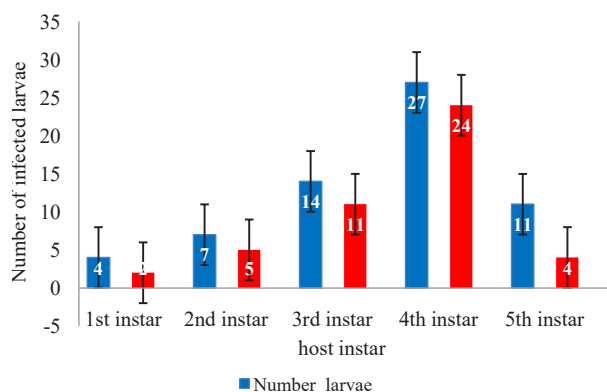


Fig. 5. Nutritional preference of *C. sesamae* vs. larval stages of *T. rosacea*

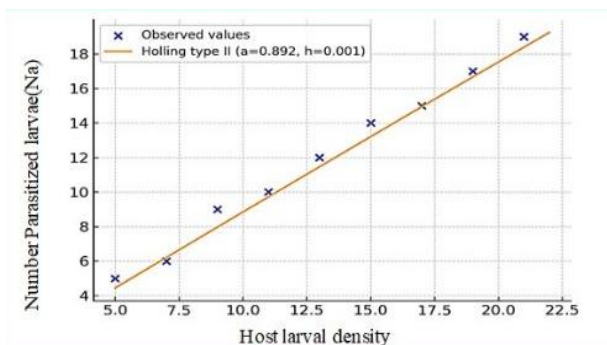


Fig. 6. Functional response (Holling Type II) of *C. sesamae* on *T. rosacea*

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