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BIOLOGY AND IMMATURE STAGES OF *OXYRHACHIS RUFESCENS* (MEMBRACIDAE: OXYRHACHINAE) WITH REFERENCE TO IMMATURES OF TWO OTHER SYMPATRIC SPECIES AND THEIR RELATIONSHIPS[1]

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Abstract. Biology and immature systematic studies of *Oxyrhachis rufescens* on *Prosopis juliflora* were carried out in the field and in the laboratory in order to determine the period required for complete life cycle and to help separate the eggs and immature stages of this species from 2 other closely related sympatric species found on the same host plant. The period for the complete life cycle was 35–43 d under 11–41 °C and 14–90% RH. A key to separate immature stages of *D. rufescens* from 2 sympatric species, *O. taranda* and *O. serratus*, is given.

Capener (1962) has emphasized the value of immature characters in differentiating the tribes of Oxyrhachinae and in a later paper (1968) has stressed the need for employing the right techniques for the preservation of immatures. Recently Yasmeen & Ahmad (1975, 1979) and Ahmad & Abrar (1976) have shown that closely related sympatric species of the genera *Gargara* Amyot et Serville, *Tricentrus* Stål and *Oxyrhachis* Germar (Membracidae) can be separated more readily in their immature stages. It is difficult to separate *Oxyrhachis rufescens* Walker, of which neither the eggs nor the immature characters are known in the literature to date, from collections also containing sympatric species *O. taranda* (Fabricius) and *O. serratus* Ahmad & Abrar which are found on the same host plants (Ahmad & Abrar 1974). The present studies were undertaken to describe and illustrate the eggs and immatures of *O. rufescens*, with special reference to measurements and proportions of various components of the body and to enable us to construct a key to separate the immature stages of *O. rufescens*, *O. taranda*, and *O. serratus*.

MATERIALS AND METHODS

Several pairs collected from the host plant *Prosopis juliflora* were kept in the laboratory, 1 pair per glass chimney, with fresh twigs of the host plant in a beaker of water. Eggs laid by females in the laboratory did not hatch but first and later instars brought from the field molted in the normal way. Hatching was observed only in the field by releasing several pairs each in muslin bags tied on branches of the host plant. Also, the time and frequency of copulation were observed only in the field. The

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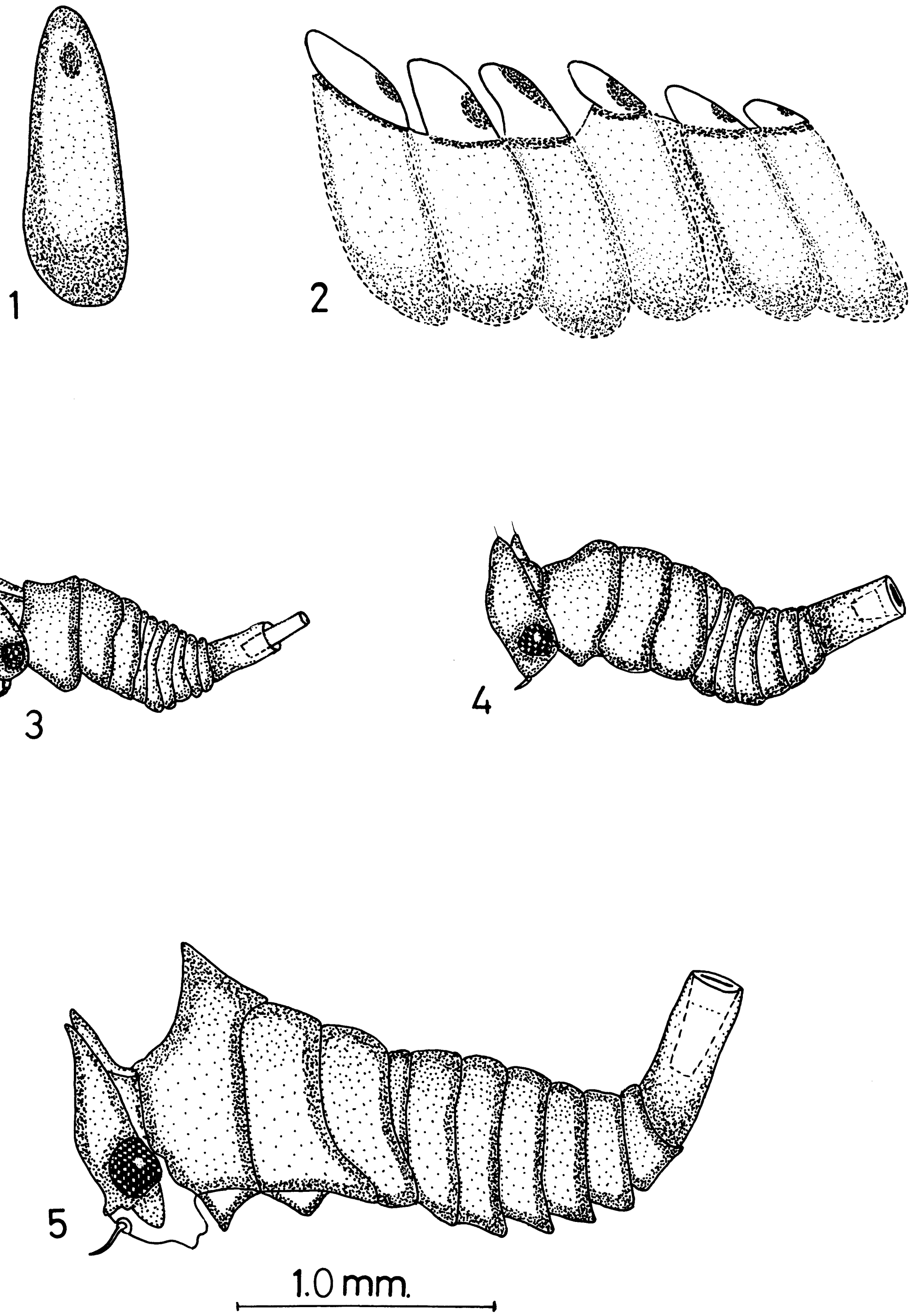

FIG. 1–5. *Oxyrhachis rufescens,* eggs and lateral view of immature stages: **1,** egg; **2,** cluster of eggs; **3,** 1st instar; **4,** 2nd instar; **5,** 3rd instar.

molting of each stage from 1st to 5th instar was observed in both field and laboratory. In all, 11 observations were made of hatching and molting of each stage. Samples of eggs and immature stages were also preserved in alcohol and glycerine. Measurements of eggs and of structures of immatures were taken in glycerine, using a micromillimetre slide. Drawings were made by setting the representatives of each stage in glycerine on cotton. Standard deviations were calculated to help find the range of all observations at 99% confidence limit, using standard statistical procedures (Table 1, 2). The correlation coefficient (r) of length and width of eggs was also calculated to help compare with these values of eggs of 2 other sympatric species, *O. taranda* and *O. serratus*, given by Ahmad & Abrar (1976).

BIOLOGY AND IMMATURE STAGES OF *O. RUFESCENS*

Copulation

Copulation takes place in an end to end position. The pair remains coupled for 3–5 h if left undisturbed. Each pair has been observed to copulate more than once.

Egg laying

Oviposition takes place 3–6 d after copulation. Eggs are laid on branches and stems and fixed with a covering of whitish jelly-like substance, so that only a small anterior portion of them remains visible. The eggs are laid in parallel rows or haphazardly. The number of rows and number of eggs per row are variable (usually between 1–8). The oviposition period is 1–2 d. After ovipositing, the female remains over the eggs until they hatch. If the female dies or is removed from the eggs, the eggs fail to hatch (Ahmad & Afzal, unpublished).

Immature stages

Egg (Fig. 1–2). Range length and width 1.23 ± 0.04 and 0.30 ± 0.01 at 99% confidence limit, respectively; elongate; when fresh, appears transparent, whitish, and gradually becomes darker. After 4–6 d, turns brownish and gradually blackish brown and then a pair of red eye spots appear; after 12–15 d the 1st stage immature emerges, head in front, from the egg.

First instar (Fig. 3). Brownish gold; head about as wide as long, cranial processes short, rounded but prominent and terminating in a hairlike bristle, head deflexed, extending ventrally to 1st pair of legs, eyes large, rounded, reddish; antennae setaceous, 3-segmented; labium 2-segmented, reaching to 6th abdominal segment, 1st segment longer than 2nd; pronotum about 3× as wide as long, longer and broader than head; mesonotum shorter and broader than pronotum; metanotum shortest, posterior margin concave.

Second instar (Fig. 4). Slightly larger than 1st instar; lateral margin of subocular expansions with ridges; labium now reaching to 4th abdominal segment; lateral lobes of prothorax and mesothorax appearing; median pronotal process appearing as a projection.

Third instar (Fig. 5). Cranial processes fully developed, darker; labium reaching to 3rd abdominal segment; apical ½ of 3rd antennal segments turning pale; lateral lobes of thorax more developed than that of mesothorax; mesothoracic wing pads concealing lateral portions of metanotum; metathoracic wing pads reaching to ¼ of 2nd abdominal segment; small pleural

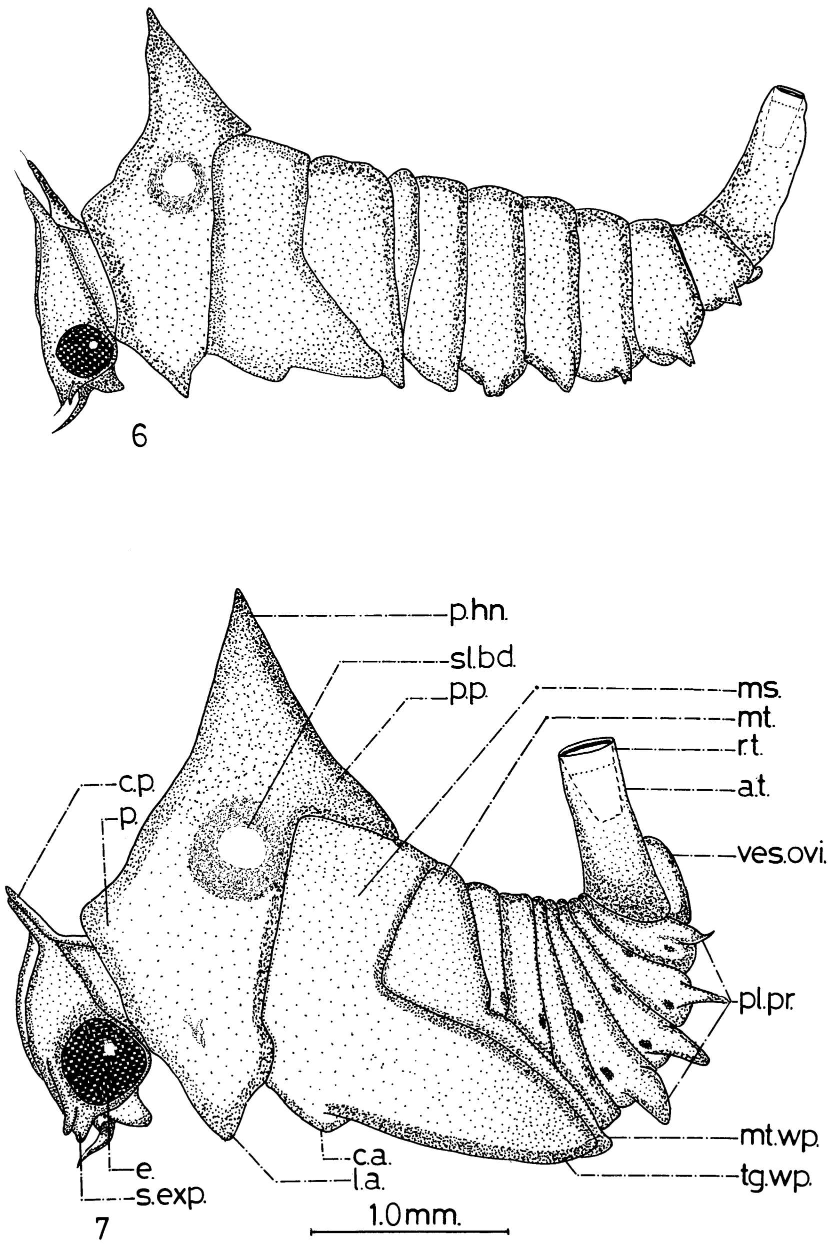

FIG. 6–7. *Oxyrhachis rufescens,* lateral view of immature stages: **6,** 4th instar; **7,** 5th instar. a.t., anal tube; c.a., costal angle; c.p., cranial process; e., eye; l.a., lateral angle; ms., mesonotum; mt., metanotum; mt.wp., metanotal wingpad; p., pronotum; p.hn., pronotal horn; pl. pr., pleural process; p.p., posterior process; r.t., retractile tube; s.exp., subocular expansion; sl.bd., suprahumeral bud; tg.wp., terminal wingpad; ves.ovi., vestigial ovipositor.

TABLE 1. Duration of life cycle of *Oxyrhachis rufescens* on the basis of 11 observations.

	DAYS TO HATCH		RANGE AT 99% CONFIDENCE LIMIT
	DAYS	MEAN	
Egg	12–15	13.27	13.27 ± 0.86
1st instar	4–5	4.45	4.45 ± 0.41
2nd instar	4–5	4.54	4.54 ± 0.41
3rd instar	4–5	4.54	4.54 ± 0.41
4th instar	5–6	5.54	5.54 ± 0.41
5th instar	6–7	6.64	6.64 ± 0.39
Total time	35–43	39	39 ± 1.70

lobes appearing as lateral projections of 4th, 5th and 6th abdominal segments and bearing small bristles; abdominal segments appearing dark brown in middle.

Fourth instar (Fig. 6). Labium reaching to 2nd abdominal venter; ocelli visible; posterior pronotal process concealing mesothorax; supraocular callosities and suprahumeral buds present on sides of pronotum; mesothoracic wing pads concealing ¾ of metathoracic wing pads, latter reaching to 2nd abdominal segment; pleural lobes present on 7th segment.

Fifth instar (Fig. 7). Labium reaching hind coxae; wing pads extending to 4th abdominal segment; metanotal wing pads more or less completely overlapped by mesonotal wing pads.

KEY TO THE IMMATURE STAGES OF *Oxyrhachis taranda*, *O. serratus* AND *O. rufescens*

1. Ocelli present, labium shorter, never passing beyond 2nd abdominal venter 10
   Ocelli absent, labium longer, always distinctly passing beyond 2nd abdominal venter 2
2. Mesothoracic wing pads present (3rd stage) 3
   Mesothoracic wing pads absent 5
3. Labium shorter, never reaching beyond 3rd abdominal venter ..... **rufescens**
   Labium longer, reaching to 4th abdominal venter 4

TABLE 2. Range of measurements of different stages of *O. rufescens.*

	INSTAR				
	1ST	2ND	3RD	4TH	5TH
Total length	1.26 ± 0.01	1.79 ± 0.04	2.52 ± 0.06	3.38 ± 0.04	4.78 ± 0.11
Width of head	0.58 ± 0.01	0.79 ± 0.02	1.10 ± 0.08	1.33 ± 0.02	2.00 ± 0.10
Length of head	0.45 ± 0.01	0.56 ± 0.01	0.74 ± 0.01	0.89 ± 0.01	1.28 ± 0.10
Length of cranial processes	not observed*	0.85 ± 0.01	2.80 ± 0.03	0.32 ± 0.01	0.33 ± 0.02
Width of pronotum	0.61 ± 0.01	0.68 ± 0.02	1.33 ± 0.01	1.90 ± 0.06	2.59 ± 0.07
Length of pronotum	0.18 ± 0.01	0.28 ± 0.01	0.48 ± 0.01	0.81 ± 0.03	0.99 ± 0.01
Length of pronotal processes	not observed*	not observed*	0.28 ± 0.01	0.57 ± 0.02	1.06 ± 0.04

* Too short to measure.

4. Cranial processes rounded or subacute at apices . . . . **serratus**
   Cranial processes pointed at apices . . . . **taranda**
5. Labium at least reaching to 6th abdominal venter (1st stage) 6
   Labium never reaching beyond 5th abdominal venter (2nd stage) 8
6. Labium reaching to 6th abdominal venter . . . . **rufescens**
   Labium reaching to 7th abdominal venter 7
7. Cranial processes blunt . . . . **serratus**
   Cranial processes pointed . . . . **taranda**
8. Labium reaching to 4th abdominal venter . . . . **rufescens**
   Labium reaching to 5th abdominal venter 9
9. Cranial processes rounded at apices . . . . **serratus**
   Cranial processes pointed at apices . . . . **taranda**
10. Labium shorter, at the most reaching to middle of 2nd abdominal venter or to hind coxae, wing pads at least reaching to 4th or 5th abdominal segments (5th stage) 11
    Labium comparatively longer, reaching to posterior margin of 2nd abdominal venter, wing pads relatively less elongated, hardly passing beyond 3rd abdominal segment (4th stage) 13
11. Labium reaching to hind coxae . . . . **rufescens**
    Labium reaching to middle of 2nd abdominal venter 12
12. Cranial processes only slightly more than ⅓ of head length and rounded at apices . . . . **serratus**
    Cranial processes distinctly more than ⅓ of head length and pointed at apices . . . . **taranda**
13. Labium reaching to 2nd abdominal venter . . . . **rufescens**
    Labium reaching to 3rd abdominal venter 14
14. Cranial processes shorter about ⅓ of head length and rounded at apices, median pronotal process round . . . . **serratus**
    Cranial processes longer, distinctly more than ⅓ of head length and pointed at apices, median pronotal process pointed . . . . **taranda**

DISCUSSION

According to Ahmad & Abrar (1976), the length of an egg increases with an increase in its width in *O. serratus* ($r = 0.7$) in contrast to that in *O. taranda,* in which the length of an egg decreases with an increase in its width ($r = -0.79$), but in *O. rufescens* we now find that there is no correlation between the length and width of an egg ($r = -0.07$).

Membracid egglaying, site, and mechanics are discussed by Funkhouser (1917). The eggs are most commonly deposited on or under the bases of twigs by species of the genera *Oxyrhachis, Tricentrus, Gargara,* and *Otinotus.* However, a number of membracid species deposit their eggs in buds, on leaves, or on axils of leaves. Yasmeen (1977) reported that eggs of all the above genera are protected only by bark or bud scales, but we have observed eggs of *Otinotus* spp. and *Oxyrhachis* spp. in the field as well as the latter under laboratory conditions and have found that the eggs of both are protected by a gelatinous sheath and are firmly bonded.

There appears to be no egg burster, micropyle, or cap in the eggs of the species of *Oxyrhachis* we studied, but Funkhouser (1917) has reported a large cap which swells and wrinkles before hatching in the eggs of most of the membracid species he studied.

Capener (1962) regarded the immature characters of oxyrhachines useful at tribal level but underrated their importance for separating closely related genera or species. He considered them varying from one instar to another and intergrading between species so that the differences could be of no more than subsidiary use in separating species. But we have found the immature characters to be an excellent tool for the separation of not only genera but also for the separation of closely related species-groups and species (Yasmeen & Ahmad 1975, 1979; Ahmad & Abrar 1976).

Our findings agree with those of Capener (1962): in species of the subfamily Oxyrhachinae the last instar possesses a pair of large and rudimentary cranial tubercles, a pronotum with a prominence in front that sometimes is developed into a distinct horn, and a rudimentary posterior process extending to the mesonotum.

Immature stages of oxyrhachine species showed marked differences, especially in the later stages (i.e., from 3rd to 5th), that proved helpful in separating closely related species on the basis of the following characters (described in description and key): size; color and pigmentation; shape of cranial processes; length of labium; and length of wing pads.

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