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HATCHING BEHAVIOUR OF AN AMBLYCERAN AND AN ISCHNOCERAN LOUSE (INSECTA, PHTHIRAPTERA)

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With 3 figures

A. Introduction

Very few workers have studied the hatching behaviour of the lice. The eclosion of nymph after the completion of incubation period involves a series of steps and a structure called hatching organ. The process of hatching of some lice is noted by WIGGLESWORTH (1932), MARTIN (1934), ANSARI (1943), ARORA & CHOPRA (1959), EICHLER (1963) and AGARWAL (1959, 1967). KUMAR & SOMADDER (1974) have given an account of hatching organ of three anopluran species. The hatching organ is a special part of prelarval skin which is shed at the time of hatching of phthirapteran egg (WIGGLESWORTH, 1932). It is also considered as a modified part of embryonic cuticle (KUMAR & SOMADDER, 1974) and is given different names viz. egg burster, egg tooth or egg ruptorovi. This organ helps in hatching of embryo from the egg by cutting vitelline membrane of the egg with the help of hatching spines present on the disc. AGARWAL & SAXENA (1982) have studied the hatching organ of *Lipeurus lawrensis tropicalis* but the hatching behaviour of this louse escaped attention. AGARWAL (1967) has given an account of hatching behaviour of *Falcolipeurus frater* infesting White Scavanger Vulture (*Neophron percnopterus*). There is hardly any published information relating to the structure of hatching organ of any amblyceran louse. However, AGARWAL (1959, unpublished) has noted the features of the hatching organ of *Laemobothrion percnopteri* (Amblycera). In the present studies an attempt has made to study the hatching behaviour of the ischnoceran louse *Lipeurus lawrensis tropicalis* PETERS and the amblyceran species *Menopon gallinae* (L.).

B. Material

The incubation period of the eggs of *Lipeurus lawrensis tropicalis* at 35 ± 1 °C and 82 – 83 % R. H. was found to be 5 days. Freshly laid eggs, obtained from the stock were incubated for 4 days at the above said temperature and R.H. On the fifth day the eggs were taken out and examined under the microscope. The eggs likely to hatch were kept under microscope for direct observation. Similarly, the freshly laid eggs of *Menopon gallinae* were incubated till 3rd day and then kept under constant observation under microscope (low power). The events observed were recorded simultaneously.

The structure of the hatching organ of *Menopon gallinae* was studied by taking out the hatched eggs. Such eggs were dehydrated and then pressed from the posterior to the anterior end before mounting with the help of fine needles. In many of the eggs the hatching organ remains inside the eggs even after the eclosion of the nymph. When the pressure is applied at the rear end of the eggs, the hatching organ comes out through the mouth of the egg. The same is then mounted and studied.

C. Observations

Lipeurus lawrensis tropicalis

The eggs (chorion) are milky-white in appearance when freshly laid but become transluscent on second day. On third day, the yellow coloured nymph becomes visible through the egg-shell, lying somewhat obliquely and making pulsating movements of the body. Five days after the incubation a fully developed embryo begins to show rhythmic movements about 12 hours before the emergence.

Nearly 2.15 minutes before the hatching, embryo starts gulping air which enters possibly through micropylar pores. The air then traverses the alimentary canal, escapes out of the anus and accumulates behind the embryo (in the space between embryonic cuticle and egg membrane). Initially the frequency of inhalation is slow but it increases gradually and a rapid stream of air bubbles entering the gut is clearly visible under the microscope (fig. 1). The air bubbles coalasce with one another and a large air bubble is produced in about 7–8 seconds. Ultimately it bursts and air escapes through the anus of the embryo. The process is continued for nearly 1 hour 30 minutes. The embryo originally occupying the entire

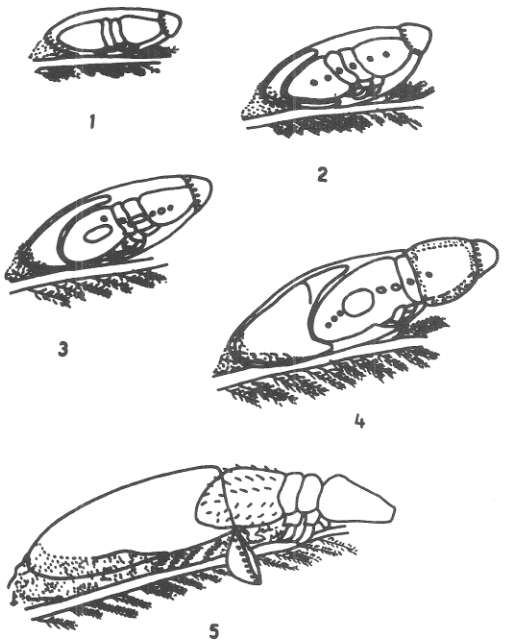

Fig. 1. Diagrammatic representation of events during hatching of the nymph of *Lipeurus lawrencis tropicalis*.

egg is driven towards the opercular end as more and more air accumulates at the rear. Due to constant pressure the operculum gets loosened along the circumference. The process is probably assisted by the rotation of spines present on the hatching organ. Finally, the operculum detaches from the egg along the circumference. As the operculum detaches, a girdle like swelling pushes forth from the mouth of the egg. The swelling goes on increasing until embryonic cuticle splits open suddenly.

First of all the head, thorax and the first pair of legs emerge out and keep moving for sometime. The nymph then grasps barbs and barbules of feather with the help of the mandibles and legs. Now the abdomen undergoes a series of muscular contractions which enable it to release itself from the embryonic cuticle inside the egg. The meso- and meta-thoracic legs are seen moving inside the egg. Due to the movements of the abdomen and legs, these become free and gradually emerge out of the egg shell. The hatching organ and the operculum may remain attached on to the side of the mouth of the egg shell or may fell down. After the eclosion of the nymph the empty egg shell remains attached to the feather with the help of cementing material glued at the posterior end. Freshly emerged first instar nymph does not show any sign of movement till 20 minutes and then starts feeding.

The duration of different stages of hatching of five eggs are shown in Table 1. The entire process starting

Tab. 1. Showing duration of different stages during hatching of 5 nymphs of *Lipeurus lawrensis tropicalis.*

Sl. No.	Time required for detaching the operculum	freeing from the egg	complete process
1.	2 h, 3 min	7 min	2 h, 10 min
2.	2 h, 3 min	9 min	2 h, 12 min
3.	2 h, 6 min	8 min	2 h, 14 min
4.	2 h, 5 min	11 min	2 h, 16 min
5.	2 h, 3 min	10 min	2 h, 13 min
∅:	2 h, 4 min	9 min	2 h, 13 min

from the sucking of air upto the complete eclosion takes an average about two hours 13 minutes. The embryo requires only 9 minutes to come out of the egg after the emergence of the head, thorax and first pair of legs. The rest of the time i.e. two hours 4 minutes is needed for sucking in of the air and building necessary pressure at the rear end of the egg which helps in detachment of operculum (Table 2).

AGARWAL & SAXENA (1982) have described the hatching organ of *Lipeurus lawrensis tropicalis.* The present studies do not indicate any departure from the account given by them. It consists of two components, the anterior one is in the form of small, oval, saucer shaped thickened disc (0.025 to 0.03 mm in diameter) which bears 24–29 hatching spines. There are 6–7 spear shaped spines (out of which one located in the centre is the largest and 18–22 small bluntly pointed spines). From the either end of the disc arise thickened, chitinous, rod-like structures which after running for short distance descend down, make a loop and finally run inwards to join a large cup like structure (the posterior component).

Tab. 2. Showing the description of the behaviour of the embryo and the time taken during different stages prior to hatching of the eggs of *Lipeurus lawrensis tropicalis.*

Duration of different stages of hatching	Description of the behaviour of the embryo prior to the hatching
1.10 p.m.	Air sucking starts (5–10 air bubbles in 10 seconds).
1.22 p.m.	Larger number of air bubbles.
1.30 p.m.	Air bubbles in plenty and form long air bubble, the long air bubble bursts and air accumulates behind the embryo.
2.55 p.m.	Air sucking at climax (20–23 bubbles in 10 seconds).
3.00 p.m.	Egg hatches, operculum detaches.
3.05 p.m.	Embryonic cuticle splits.
3.08 p.m.	Operculum and hatching organ comes to hang at the mouth of the egg shell.
3.10 p.m.	First pair of the legs gets free.
3.12 p.m.	Second pair of the legs gets free.
3.13 p.m.	Third pair of the legs gets free.
3.15 p.m.	Completely out of the egg shell.

Menopon gallinae

The egg of *M. gallinae* remains transluscent white till 2nd day (as in fresh condition). It turns pale yellow on 3rd day. One red spot (embryo's eye spot) becomes visible on either side of the egg, below the opercular disc. As the yolk filled inside the egg is quite thick and gelatinous and also the embryo is more or less transparent (very feebly sclerotised), any embryonic structure remains unclear. Further, it has been noticed that the tentacle (or to say the tencacle-like membranous processes) occurring around the mouth of the egg remain tilted or collapsed in a fresh egg start erecting as the development proceeds. Before the emergence they give a lotus like appearance to the opercular end. The opercular tentacle also straightens one day before eclosion. Due to the pressure developing inside the egg during the process, the egg often gets tilted and turned from its original position. Furthermore, it has also been noted that the embryo inside the egg remains nearly motionless and does not show any pulsating movements. The viscous liquid filled inside the egg shows some sort of vibrations but the eye spots (visible as red dots) remain stationary.

The embryo starts sucking the air nearly for 35 minutes to 1 hour before the hatching. The sequence of events that follow the entry of air bubbles have been listed in the Table 4 and also shown in Fig. 2. The time taken for the detachment of the operculum and freeing of the nymphs from five eggs have been indicated in Table 3. The air sucking, visible in the form of entry of air bubbles, leads to the formation of a large air bubble at the postero-lateral side of the egg. As the sucking continues the size of the air bubble also increases. It may result in building a pres-

Tab. 3. Showing duration of different stages during hatching of 5 nymphs of *Menopon gallinae.*

Sl. No.	Time required for detaching the operculum	freeing from the egg	complete process
1.	54 min	3 min	57 min
2.	45 min	2 min	47 min
3.	38 min	2 min, 40 sec	40 min, 40 sec
4.	42 min	2 min, 20 sec	44 min, 20 sec
5.	33 min	2 min, 24 sec	35 min
∅:	42 min, 24 sec	2 min, 24 sec	44 min, 36 sec

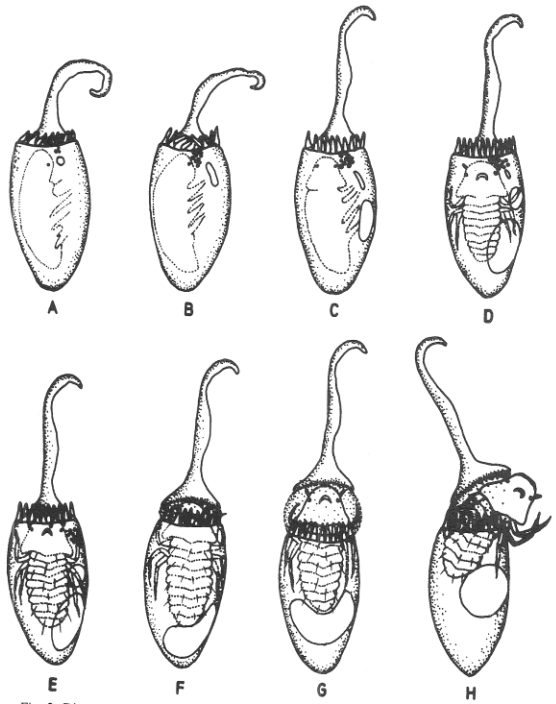

Fig. 2. Diagrammatic representation of events during hatching process in *Menopon gallinae.*

sure inside the egg which ultimately causes detachment of the operculum. It requires on an average 42 minutes (Table 3). It may be noted that the embryo remains in its position inside the egg (on the lateral side) and is not pushed anteriorly during the process.

The hatching organ presumably helps in detachment of the operculum. The exact location and function of the hatching organ during the process remained obscured even under the microscope. Perhaps, the cup-like component of the hatching organ is pushed anteriorly towards the operculum causing the detachment of the operculum.

After the detachment, a girdle like swelling (embryonic cuticle filled with air) containing the head of the nymph comes out of the egg. As soon as the cuticle splits the head as well as the first pair of legs become free. It is followed by jerky movements of the abdomen (inside the egg). Soon, the thorax and abdomen also comes out along with 2nd pair of legs totally out while 3rd pair of legs get released after some more efforts by the embryo (Table 4). Thus, the entire process of freeing from the egg takes hardly 2–3 minutes only (average 2 minutes 24 seconds). Sometimes, a few abdominal setae remain attached

Tab. 4. Showing the description of the behaviour of the embryo and the time taken during different stages prior to the hatching of eggs of *Menopon gallinae.*

Duration of different stages of hatching	Description of the behaviour of the embryo prior to the hatching.
4.02 p.m.	First air bubble appeared below the opercular disc.
4.04 p.m.	Air sucking continued at the rate of 15 bubbles per minute.
4.09 p.m.	Brisk air sucking at the rate of 34 bubbles per minute. Very small bubbles appearing below the opercular disc and uniting to form slightly bigger bubbles which pass to the postero-lateral side and accumulate to form a larger one.
4.14 p.m.	Bubbling continued at the rate of 17 bubbles per minute. The size of the large bubble (located at the postero-lateral end) increases and it covers ⅓ of the egg (posterior part).
4.35 p.m.	Air sucking continued at the rate of 19 bubbles per minute. The minute bubbles moving in zig-zag manner (after entry) and colliding with each other frequently, then finally passing to the posterior end.
4.42 p.m.	Brisk vibrations in the liquid below the opercular disc but air sucking at the rate of 15 bubbles per minute.
4.47 p.m.	Frequency of air sucking increases (31 bubbles per minute).
4.51 p.m.	The egg gets turned automatically. The size of large bubble exceeds to half of the egg. Air sucking slows but vibrations in the liquid below the operculum increase.
4.52 p.m.	Air sucking stops.
4.54 p.m.	Few small bubbles vibrate briskly below the operculum (probably due to vibrations in the liquid).
4.56 p.m.	Operculum lifting and a girdle-like swelling coming out of the egg mouth.
4.57 p.m.	The size of the girdle-like swelling increases. The head of the embryo becomes visible in the swelling.
4.58 p.m.	Head as well as the first pair of legs come out of the egg (by rupturing of the embryonic cuticle). The abdomen moving sideways inside the egg.
4.59 p.m.	Abdomen and the second pair of legs also come out. Only the third pair of legs still inside the egg. The embryo struggles hard to pull it out.
5.00 p.m.	The nymph completely out of the egg.

to the egg shell and the nymph shows wriggling movements in order to get rid of the egg shell. After being completely free, the nymph remains quiescent for 3–4 minutes and then starts walking over the black paper sheet comfortably. It does not require any assistance of feather barbs or barbules, at least in vitro conditions.

After the eclosion, the hatching organ remains attached to the anterior end of the empty egg shell and can be taken out after proper processing. On the other hand, the operculum normally detaches and falls down from the egg. The empty egg consists of a very thin, chitinous egg shell bearing tentacles at the anterior end. They are very light and get easily displaced even by the exhaled air during the normal breathing of the observer. This may be one reason of the non-availability of the hatched eggs of *M. gallinae* during the examination of feathers.

The hatching organ of *Menopon gallinae* apparently consists of a thick chitinous cup like structure measuring 35 to 50 μ in length and 18 to 22μ in width (Fig. 3). The posterior end of the cup tapers and is supported by thin chitinous rods (on either side) which after running for short distance sideways, turns and make a loop to run towards the central area which appears like bunch of feebly chitinized fibres. From the central area arises a long, thick, spine like structure bearing 4–6 tooth like projections (giving it a somewhat saw like appearance). The spine extends to the entire length of the cup and is often supported by membranous structures. The presence of a hatching disc could not be recorded in this louse. Except the anterior ¼ part, the entire cup remains sorrounded by a very thin, transparent, chitinous structure (presumably the embryonic cuticle).

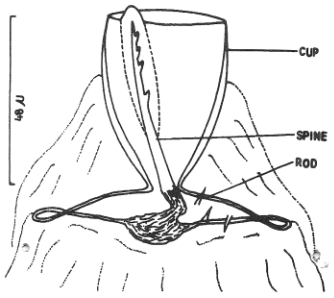

Fig. 3. Hatching organ of *Menopon gallinae.*

D. Discussion

The hatching behaviour of *Lipeurus lawrensis tropicalis* does not present any departure from other species studied from this point of view by WIGGLESWORTH (1932), MARTIN (1934), ANSARI (1943), ARORA & CHOPRA (1959) and AGARWAL (1967). AGARWAL (1967) has made studies on the hatching behaviour of an ischnoceran louse *Falcolipeurus frater* and also recorded the time required during different stages of hatching. In *F. frater* the egg requires 1 hour 2 minutes to 1 hour 40 minutes for detaching the operculum, 13–15 minutes for freeing from the egg and thus the complete process lasts 1 hour 15 minutes to 1 hour 55 minutes. In *Lipeurus lawrensis tropicalis,* the duration is slightly longer as the process lasts 2 hour 10 minutes to 2 hour 16 minutes (average 2 hour 4 minutes for detachment of operculum and 9 minutes for freeing from the egg). The hatching organ of *L. lawrensis tropicalis* has already been studied by AGARWAL & SAXENA (1982). It probably helps in detachment of operculum (with the help of spines present on the hatching disc) apart from protecting the head of the embryo from injury during the process.

The structure of the egg, nature of hatching organ and the hatching behaviour of *Menopon gallinae* has been described here for the first time. Moreover, there is no published information regarding the hatching organ or hatching mechanism of any amblyceran Phthiraptera. The only description of the hatching organ of any amblyceran species (*Laemobothrion percnopteri*) is available from the work of AGARWAL (1959, unpublished). In *L. percnopteri,* it consists of a median rod (proximally bifid) bearing spines. The distal end of the median rod on *L. percnopteri* supports a semicircular disc bearing spines. The hatching organ of *Menopon gallinae* differs completely with that of *Laemobothrion percnopteri* but shows some resemblance with *Lipeurus lawrensis tropicalis.* In the later, the hatching organ consists of two components, anterior, the hatching disc and posterior, the cup like portion. The anterior component (the hatching disc) seems to be lacking in *Menopon gallinae* and is represented by only the posterior component which contains an additional structure, a big sized toothed spine. Due to minute size, its function remains obscure during the process but most probably it is also used for tearing the embryonic cuticle before eruption.

The account of hatching mechanism of any amblyceran species is being given here for the first time. The hatching process of *Menopon gallinae* seems to be relatively less time consuming than *Lipeurus lawrensis tropicalis* and requires only 33–54 minutes. However, in case of *M. gallinae,* the air does not accumulate behind the embryo (as in *L. lawrensis tropicalis*) but at the posterolateral side in the form of a large air bubble. Furthermore, after the detachment of the operculum, the time required by the nymph for coming out of the egg is quite less (2–3 minutes) as compared to *L. lawrensis tropicalis* (7–11 minutes). Thus, being an active menoponid, *M. gallinae* exerts little as compared to the ischnoceran *L. lawrensis tropicalis* in which the embryo holds the feather barbs by its pro-and mesothoracic legs and struggles hard to draw itself out of the egg.

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Summary

The duration of different stages and the behaviour of embryo during hatching process has been determined in case of an ischnoceran *Lipeurus lawrensis tropicalis* and an amblyceran louse, *Menopon gallinae* by direct observation. The hatching organ of *M. gallinae* has also been studied.

Zusammenfassung

Auskriechen aus dem Ei bei einer ischnozeren und bei einer amblyzeren Tierlaus-Art (Insecta, Phthiraptera). Unter dem Mikroskop wurde mehrfach das Auskriechen aus dem Ei bei den auf indischen Haushühnern (*Gallus gallus* f. *dom.*) lebenden Federlingsarten *Lipeurus lawrensis tropicalis* Peters, 1931[1] [Ischnocera, Lipeuridae] und *Menopon gallinae* (L., 1758) [Amblycera, Menoponidae] beobachtet. Bei *M. gallinae* wird erstmals für eine Amblyzere Schlüpfen aus dem Ei und Eizahnorgan beschrieben.

Lipeurus lawrensis tropicalis: Nach dem 5. Tag der Eiablage ist der Embryo voll entwickelt und beginnt 12 Stunden vor dem Auskriechen mit rhythmischen Bewegungen. Etwa zwei Stunden und 15 Minuten vor dem Auskriechen saugt er Luft an, die, über den Anus wieder abgegeben, hinter und zwischen ihm und dem Chorion eine größer werdende Blase bildet. Infolge des dadurch wachsenden Innendrucks löst sich nach ca. zwei Stunden und 3 Minuten der Eideckel an der vorgebildeten Bruchlinie. Nachdem die Embryonalkutikula gerissen ist, erscheinen zuerst Kopf und Thorax mit erstem Beinpaar. Mit Mandibeln und erstem Beinpaar hält sich die Larve an der Feder fest. Durch abdominale Peristaltik löst sie sich schließlich vollständig von der in der Eihülse zurückbleibenden Embryonalkutikula. Bis zu 20 Minuten nach dem Auskriechen verharrt sie reg-

[1] Anmerkung der Red. – Diese Art lebt ursprünglich auf Perlhühnern (*Numida* spp.) und hat sich sekundär auf Haushühnern in Asien und Nordamerika angesiedelt. Sie gehört sicher in die Gattung *Numidilipeurus* Tendeiro, 1955. – E. M.

los, dann beginnt sie zu fressen. Vom ersten Luftschlucken bis zum Verlassen des Eies benötigt die Larve durchschnittlich 2 Stunden und 13 Minuten, davon 7 bis 11 Minuten zum Schlüpfen. – Das Eizahnorgan (Oviruptoren) wird kurz beschrieben.

Menopon gallinae: Nach dem 3. Tag der Eiablage ist der Embryo transparent und noch bewegungslos. Etwa 35 Minuten bis zu einer Stunde vor dem Schlüpfen beginnt er Luft anzusaugen, die posterior-lateral eine große Blase bildet. Durchschnittlich 42 Minuten benötigt der Embryo, den Eideckel zu sprengen. Dabei waren Lage und Funktionsweise des Eizahnorgans nicht zu bestimmen. Nach dem Aufreißen der Embryonalkutikula erscheint die Larve zuerst mit Kopf und erstem Beinpaar aus dem Ei. Mit ruckartigen Bewegungen befreit sie sich von diesem. Das Auskriechen dauert nur durchschnittlich zwei bis drei Minuten. Danach verharrt die Larve drei bis vier Minuten, um schließlich geschwind davonzulaufen. Zum Auskriechen benötigt sie keine Feder. – Das Eizahnorgan wird ausführlich beschrieben und mit denen von *Lipeurus lawrensis tropicalis* und *Laemobothrion percnopteri* (Gervais) verglichen.

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