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IMPACT OF PARASITISM ON HEXAMERMIS VISWAKARMA DHIMAN ON ITS HOST LEPTOCORIS AUGUR FABR.

Pooja Arora
Dept. of Zoology,
K.L. D.A.V. (PG) College, Roorkee (India)

ABSTRACT

H. viswakarma is a parasitoid of ***L. augur***. This parasitoid cause morphological, anatomical, physiological and biochemical changes in the host. Preparasitic juvenile enters healthy host body through weak body points and grows enormously at the expense of host body tissues and brings these changes. At last parasitic nema merges out from host body and the bug dies within 15 to 30 minutes.

Keywords: Impact, ***H. viswakarma***, ***L. augur***, morphological effect, anatomical effect.

INTRODUCTION

Leptocoris augur Fabr. (Heteroptera Coreidae Rhopalidae) is a pest of Kusum plant, ***Scheichera oleosa*** Lour (Sapindaceae) on which lac of best quality is cultivated. The bug population causes serious damage to the various parts of host plant particularly seeds which loss viability. The bug has its various natural enemies which naturally control the bug population upto some extent (Jain, 201). Among these natural enemies, ***H. viswakarma***, Dhiman (Nematoda-Mermithidae) has good potential to be used as biocontrol agent various studies have been carried out time to time on this parasitoid by Dhiman (1984) Dhiman and Singh (1991) and Tomar and Dhiman (2017). In present paper an effort is made to describe the impact of parasitism of this nematode on its host bug, ***L. augur***.

MATERIALS AND METHOD

1. **Collection of bugs:-** Nymphal as well as adults population of host bug, ***L. augur***, were collected along with fresh leaves and seeds of Kusum plant from the field area (Horticultural Experiment and Training Centre, Saharanpur) by hand picking method, during rainy season and were brought alive to the laboratory in polyethylene bags. Collection was made randomly.
2. **Rearing of bugs in hurricane glass lamp chimneys:-** In the laboratory the collected bugs were reared at room temp. [20°-25°C) on fresh tender leaves and moist crushed seeds of Kusum plant placed in a petridish in hurricane glass lantern chimney covered at top by fine muslin cloth to allow circulation of air. The stale food was replaced daily with fresh one and the rearing petridishes were examined daily. A moist cotton swab was also kept in the chimney to maintain necessary relative humidity.

3. **Separation of emerged post-parasitic nematodes:-** The nematodes emerged out from the host body were collected with the aid of fine camel hair brush and were kept into the petridishes filled with moist sterilized coarse sand. Now each such petridish was placed into another big petridish filled with water to avoid escape of post-parasitic nematode as well as to prevent the entry of ants etc. (double tray system). Now, these petridishes were arranged into a big tray measuring 45 cm x 30cm with 3.75 cm boarder base filled with water. The whole tray was kept covered by fine cloth so as to avoid the occurrence of micro-organism through air. The petridishes were observed daily.

   During the active period (24-48 hours), the nematodes underwent into sterile coarse sand, some of them remained on the sand in coiling conditions. In each coil about 1-7 nematodes were found. After searching their desired position, they transferred into inactive state. This stage is termed quiescent stage which moulted twice within two to four weeks period.

4. **Rearing technique for obtaining adults, eggs, and pre-parasitic nematodes:-** When adults moulted finally, from inactive post parasitic nematodes (quiescent stage], they copulated. The process of copulation, oviposition and hatching (development till pre-parasitic stage of the nematode] was keenly observed under the stereoscopic binocular microscope. The eggs were also kept and the content of the petridishes were examined daily under binocular microscope and light compound microscope to note the process of incubation and hatching. Necessary moisture was maintained by adding boiled cold water whenever needed. Content of petridish was also observed for the yield of pre-parasitic stage.
5. **Rearing of pre-parasitic nematodes:-** For the rearing of pre-parasitic nematodes moist crushed seeds of Kusum plants along with some dried leaves were taken in a petridish and the pre-parasitic juveniles were added to it from the culture. These survived for a week in 100% R.H. and 24°-30°C.
6. **Parasitization of healthy bugs:-** Spray of newly emerged pre-parasitic nemas was carried out on healthy bugs (II to V instar nymphs and adults) reared in wooden wire gauge cages @50:1. The effect of parasitism on the bug and its behaviour was closely observed. Several dissections of the parasitized bugs were also made time to time under binocular microscope to ascertain the effect of parasitism on the anatomy of the host bug. Physiological and biochemical changes were also observed in the host bug by analysing haemolymplh.

   Regarding the rearing of ***Hexamermis*** sp; although no successful technique is available, however, the aforesaid method was found successful by repeating the experiment several times.

   Emerged post-parasitic juveniles were also successfully reared in 1% saline water- filled petridishes upto the pre-parasitic stages to avoid fungal infection which often occurred during moist sterile coarse rearing technique.

OBSERVATIONS

EVALUATION OF THE IMPACT OF PARASITIZATION ON THE HOST:- Effect of parasitization on the host has been studied in following different headings:-

a) **Morphological effect:** Influence of parasitism on this nematode on physical changes of ***L. augur*** was studied by watching the behaviour of parasitized bugs during laboratory rearing and in field also.

(i) First entrance of pre-parasitic juveniles in the host body makes the bug slightly irritated and it moves frequently but as the development of parasitic stage proceeds inside the host, there appears letharginess in the behaviour of the bug and at last prior to the emergence, the bug becomes quite sluggish.

(ii) The abdomen of the parasitized bug swells up greatly and attains biconvex shape.

(iii) Movements of antennae, legs and wings are greatly affected and the parasitized bug developed inability in flight.

(iv) A bluish spot (preferable towards left side) develops on the postero-ventral part of the abdomen of the bug.

(v) Body cuticle of the hosts abdomen becomes thin and transparent so that the coils of the parasitic ***Hexamermis*** can be seen externally under binocular microscope or even by naked eye having good eye sight.

(vi) The parasitized bug usually live alone and avoid company.

(vii) Finally, after the emergence of parasitic stage of Vishwakarma the host appears much tired and died soon.

b) Anatomical effect: Parasitic juvenile causes great anatomical changes in the host body which are-

(i) As the parasitic juvenile of Vishwakarma. resides in haemocoelomic cavity of the host ***L. augur***, hence it first takes nutrients from haemocoelomic fluid, causing quantitative loss in it.

(ii) Secondly, the fat bodies of the host which lie around the gut, reproductive organs and in haemo coelomic fluid are first dissolved and then consumed by diffusion through body wall.

(iii) Thoracic muscles, including flight muscles (dorsal longitudinal, dorso-ventral and oblique muscles as well as wing auxiliary muscles) and abdominal muscles are first dissolved and absorbed. In late stage, when the parasitic juvenile extends in head also, antennal intrinsic muscles and some mouth parts muscles are also devoured.

(iv) Highly parasitized bugs have negligible amount of haemocoelomic fluid, since, most of their body is filled up by the coilings of parasitic juveniles.

(v) However, nervous system and tracheal systems are not seen much affected.

c) Physiological and biochemical effect:

(i) Mermithids stimulate catabolism and inhibits anabolism of host fat body proteins, which thus provides the necessary dietary amino nitrogen via the host haemolymph. Cessation of vitellogenesis in the host bug can be attributed to an inability of the oocytes to sequester available proteins from the haemolymph. Subsequent oocyte resorption probably results from a depletion of vitellogenic proteins in the haemolymph due to the nutrient requirements of the developing mermithids. This results in sterilization of the female host.

(ii) Vishwakarma infections diminish the efficiency of the excertory system. A heavy infection of second and third instar nymphs increased the uric acid level of the haemolymph five times more than that of the controls. Furthermore, the concentration of faecal uric acid was reduced to one quarter that of uninfected controls.

(iii) Amino acids are taken up in large quantity by developing mermithid larvae and incorporated into protein at varying rates during mermithid development. Dipeptides and polypeptides are not taken up by the developing larvae. Under experimental conditions protein was synthesize most rapidly from leucine by 17 days old larvae.

This timing of maximum synthesis concides with the rapid increase in total dry weight and in protein level that occurs mostly between 17 and 21 days after infection [Gordon and Webster, 1972] as the developing nematode accumulates stored proteins and lipids in the trophosome (Chitwood and Jacobs, 1938; Gordon and Webster, 1972) prior to emergence from the host. These observations are seen true of viswakarma. In which rounded spheres of proteins and fats are stored in large number in trophosomes.

(iv) Parasitic larvae take up glucose from the haemolymph of the host rather than trehalose, the hosts main blood sugar. They do this faster in the mid phase (i.e., about 10 days old parasitic stage) than in the closing phase (i.e., after 18 days of their parasitic development). Glucose absorption through the cuticle is probably mediated by an active transport system in the trophosome which sets up a diffusion gradient across the pseudocoel, hypodermis and cuticle.

(v) The fatty acid composition of the host haemolymph is not significantly changed by mermithid parasitism, but the levels of cholesterol and cholestanol appears to be increased. The rapid and persistant uptake of glucose modifies the fat body haemolymph for the parasite. The depletion of the glycogen phosphorylase in the fat body prevents further glycogenolysis there and helps to maintain a constant low level of glycogen in the host fat body.

(vi) The parasitic mermithids may digest the protein reserves of the host fat body by secreting hydrolytic enzymes, it is more likely that mermithids indirectly utilize host fat body proteins by inducing changes in the hosts metabolism. The level of haemolymph, proteins and amino-acids remains relatively constant in adult host parasitized by mermithids, but a significantly decrease in fat body proteins and amino acids occurs (Gordon and Webster, 1971). Viswakarma not only utilize fat body proteins but also the lipid content of the fat as a result of which fat bodies disappear from the haemolymph.

d) On feeding capabilities:- During the host searching, sensory organs of the pre-parasites (amphids and phasmids) play a vital role which detect the presence of host in nearby vicinity and on contact with a host it searches the weak points of the body for penetration such as wing auxilaries, coxal joints etc. After entrance in the host, the parasitoid derive its complete nourishment from the haemolymph, adipose tissue and body muscles of the host bug. It causes increase demand of food and thus the feeding requirement of host bug greatly increases and it devotes more time in feeding on ***S. oleosa*** seeds and sucking sap from the leaves of adjacent host plants [Dhiman and Gulati, 1986).

Super parasitism, which is a general tendency, further inhance the continuous apply of nutrition by which feeding time is increased many fold.

e) On copulation and reproductive organs:

(i) No marked effect has been observed on the copulation ability of early parasitized adults' bug as the development of parasitoid occurs in the body of host bug, mating ability of the bug is reduced and finally no mating occurs in mid and late stage of parasitized bugs.

(ii) Testes are greatly reduced in size. Vasa deferentia are dissolved, hence, there is no connection between male gonads and ejaculatory duct. Bulbous ejaculatorius and genital muscles have also been observed damaged. Thus, 100% sterility occurs in male bugs by parasitism.

(iii) In female, maturation of ovum in ovary is greatly affected as fat and protein contents of the host body are consumed by the parasitic stage, hence, ovaries in parasitized bugs have been recorded smaller due to retardation in vitellogenesis. Moreover, oviducts are destroyed at several points and muscles of ovipositor reduced. Thus, like male bug, female sex also gets sterilized.

f) On fecundity:- Early parasitization of female bug has less effect on position and female lay eggs as usual but number of eggs laid decreases to the development of parasitic nema proceeds further in haemocoel, it after marked effect on ovaries and oviducts and fecundity is adversely affected and no oviposition occurs in mid and late parasitized female. Thus, fecundity in female, too, is reduced to zero.

g) On ecdysis:- (i) Parasitization in nymphal instars inhibits the ecdysis and nymph prolonged its life time till the emergence of nematode causes its.

(ii) If the pre-parasitic stage enters just prior to moulting in host instar, then the nymph prolonged its life time till the emergence of nematode causes its death.

(iii) If the pre-parasitic stage enters just prior to moulting in host instar, then the nymph moults into next stage which die with the emergence of nema.

DISCUSSION

Influence of parasitism of ***Hexamermis viswakarma*** on the host ***L. augur*** has been observed externally as well as internally. First entrance of pre-parasitic juveniles of ***Hexamermis*** in the host body makes the bug slightly irritated but as development of nematodes proceeds inside the host, the bug becomes lethargic and the abdomen swells up greatly and attains biconvex shape. All the parasitized bugs are incapable of taking flights. A bluish mark develops on the abdominal sternite preferably towards left side of the median body axis which has also been observed by Gulati (1989]. Finally, after the emergence of parasitic juveniles of ***H. viswakarma*** the host appears much tired and died soon. Since, its parasitic juveniles, first takes nutrients from haemocoelomic fluid, hence, there is quantitative loss in this vital body fluid and then fat bodies are consumed. Flight muscles as well as other thoracic muscles are devoured by the parasitic nematode. Highly parasitized bugs have negligible amount of haemocoelomic fluid, since, most of their body is filled up by the coiling's of parasitic juveniles. Taylor (1933) observed 1 .0 mm shorter wings than normal males in both mermithogynes and mermithanders (parasitized males) of ***Acanthomyops flavus***. He further said that in mermithanders, distention of abdomen resulted in protrusion of their genitalia. Sugiyama (1956) and Webster (1972) studied that the deformation in grasshoppers and locusts, such as crumpled and shortened wings by mermithids are not correlated with parasitic load but with the timing of infection. Petersen **et.al** (1968) mentioned that in ***Anopheles*** sp. parasitic nematodes of ***Romanomermis*** sp. apparently robbed the host of nourishment and thus prevented the development of fat body, leg rudiments and other structures parasitism was accomplished in all four larval instars, but no infected larvae pupated. Passera (1976) seems to be alone in observing true mermithostratiotes and found little modification induced in studying the effect of parasitism by Mermis sp. in ***Pheidola pallidula***, one of the obvious features of mermithized queens, termed mermithogynes by Mrazek (1908) is the tendency to shortening' of wings but there is also a distinct narrowing of the head and thorax.

Physiologically, mermithids stimulate catabolism and inhibits anabolism of host fat body proteins. Large quantities of amino-acids, lipids and carbohydrates are taken up by the parasitic larvae. Rubtsov (1967) and others have suggested that parasitic mermithids may digest the protein reserves of the host fat body by secreting hydrolytic enzymes, it is more likely that mermithids indirectly utilize host fat body proteins by inducing changes in the hosts metabolism. Gordon and Webster (1971) observed that the level of haemolymph proteins and amino acids remains relatively constant in adult ***S. gregaria*** parasitized by ***M. nigrescens***, but a significant decrease in fat body proteins and amino acids occur. He also studied that mermithid infections diminish the efficiency of the excretory system.

Amino acids are taken up in large quantities by developing ***M. nigrescens*** larvae and incorporated into protein at varying rates during mermithid development. Dipeptides and polypeptides are not taken up by the developing larvae. The developing nematode accumulates stored proteins and lipids in the trophosome (Chitwood and Jacobs, 1938; Gordon and Webster/1972) prior to emergence form the host which has also been studied in ***H. viswakarma*** Parasitic ***M. nigrescens*** larvae preferentially take up glucose from the haemolymph of ***S. gregaria*** rather than trehalose, the hosts main blood sugar. Rutherford and Webster (1974) observed that glucose absorption through the cuticle is probably mediated by an active transport system and lipids in the trophosome, which sets up a diffusion gradient across the pseudocoel, hypodermis, and cuticle. The fatty acid composition of the host haemolymph is not significantly changed by mermithid parasitism, but the levels of cholesterol and cholestanol appear to be increased (Rutherford and Webster, 1976). Condon and Gordon (1977) showed that a heavy (80 ova per host) ***M. nigrescens*** infection of ***L. migratoria*** second and third instar nymphs, increased the uric acid level of the haemolymph five times over that of the controls. Rutherford and Webster (1976) observed that the rapid and persistent uptake of glucose modifies the fat body haemolymph balance of carbohydrates. This causes a reduction in glycogenesis in the host fat body that, thus, ensures the availability of more glucose in the haemolymph for the parasites.

After penetration, the host bug feeding requirements greatly increases and it devotes more time in feeding on ***S. oleosa*** seeds and sucking sap from the leaves of adjacent host plants [Dhiman and Gulati, 1986]. Super parasitism which is a general tendency, further enhance the continuous supply of nutrition by which feeding time is increased many folds. Moreover, no copulation and oviposition has been observed in parasitized bugs. Testes are greatly reduced in size, vasa deferentia are dissolved and no

connection between male gonads and ejaculatory duct remains. Thus, male bugs are sterilized. In female, maturation of ovum in ovary is greatly affected and hence, ovaries have been recorded smaller. Moreover, oviducts are also damaged at several places. Muscles of external genitalia have also been observed destroyed. Parasitization in nymphal instars inhibits the ecdysis. Thus, parasitism is fatal for the host and brings sterility and 100% mortality of this bug.

Crawley and Baylis (1921) sectioned mermithogynes of ***Lasius*** sp. and found atrophied ovaries and nurse cells, an absence of fat body but hypertrophy of the trachea^ Christie (1936) studied that male grasshoppers infected with either ***A. decaudate*** or ***M. nigrescens*** retain the capability of producing sperm and the testes are "not materially diminished" in size. Kevan et. al. (1962) found that the number of eggs in the ovaries of naturally infected ***Metrioptera roeseli*** decreased with increasing parasite load. Denner (1976) observed that in heavy infections the gut and lateral oviducts sometimes become compressed and distorted due to pressure from the growing mermithid larvae in the haemocoel. The ovaries are diminished in size. Lesage and Harrison (1980) observed the effect of parasitism on Cricotopus by ***Neomesomermis*** sp. and ***Hydromermis*** sp. and said that parasitism results the production of intersex hosts. In the bug, under study, no such intersex was observed. Molloy (1981) studied the influence of parasitism on ***Simulium ornaturn*** by ***Mesomermis, Gastromermis*** and ***Isomermis*** and mentioned that mermithid parasitism modifies adult behaviour, parasitized males have been observed to attach to oviposition substrates and stimulate oviposition. Rhamhalinghan"! [1987) observed that ***Coccinellimermis*** when infects the ***Coccinella septempunctata*** retards the ovarian growth of the host. Movement of the endoparasite possibly causes the rupture of ovarioles, vitellarium wall and distortion of germaria. Thus, effect of parasitism on visceral organs of host is quite variable. However, in most cases it causes damage to reproductive organs and fertility of the host.

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