Morphological and reproductive dimorphism in *Zabrotes subfasciatus* (Boh.)

Deepinderjit Kaur, H.R. Pajni and P.K. Tewari[1]

Abstract

Two different morphs occur in the laboratory culture of Indian strain of *Zabrotes subfasciatus* (Boh,). The normal morph remains available throughout the year The abnormal morph appears only during the summer months and is represented by females, which look different from the normal females The abnormal morph has its pygidium uniformly covered with the pale-white setae as compared to the black pygidium with a median white streak in the normal female. The sterile females start appearing in the laboratory culture in the middle of April when atmosphere starts warming. The population of this morph gradually increases to attain a peak during the third week of June With the onset of monsoon, the percentage of abnormal morph starts declining and it disappears from the culture in the month of October The results are interesting in the light of previous reports in this laboratory according to which the abnormal females do not turn completely sterile but show decreased fecundity. The pattern of dimorphism of *Z. Subfasciatus* is also different from another bruchid species *C. Maculatus* (F.), in which both the abnormal sexes appearing of sterile individuals is presumably meant for controlling the population during the adverse seasonal conditions. However, the phenomenon also appears to show a deeper ecological significance.

Introduction

After the reported dimorphism in *C. Maculatus* (Utida, 1954; Arora & Pajni, 1959), and its confirmation by several workers in different regions of the world (Caswell, 1960; Asrora et al., 1967; Bawa et al., 1974a, 1974b; Taylor & Aludo, 1974) attention was focused on another species of bruchids for knowing the status of dimorphism/ polymorphism in them Preliminary reports have indicated different extent of polymorphism in *Callosobruchus chinensis* (L) (Nakamura, 1966, 1969; Applebaum et al., 1968; Pajni et al., 1987), *C. Analis* (F.) (Pajni, 1986a, 1986b), *Acanthoscelides obtectus* (Say.) (Huignard & Biemont, 1981), *Zabrotes subfasciatus* (Boh.) (Meik & Dobie, 1986, Pajni, 1986a, 1986b; Credland & Dendy, 1992a, 1992b) We have made detailed observations on the dimorphism of *Z Subfasciatus* by highlighting the differences in the morphology and reproductive organs of the normal and abnormal forms. The same is reported in the present communication.

[1] Department of Zoology, Panjab University, Chandigarh-160 014, India

Observations and Results

A critical examination of the laboratory-maintained cultures revealed that there occur two types of females. They differ in the colour of the setae covering their pygida (Ph. 1 – 2). On this basis, one can easily make out females with 'Black Pygidium' and 'Pale Pygidium'. These two morphs can be separated easily but there also occur certain intermediate forms and some criterion has to be adopted to allocate such forms to one category or the other The typical colour pattern and the range of variation in the two forms is given below:

Normal Morph or Black Pygidium Morph

The normal female has black and bare pygidium with one prominent median streak of white setae. The lateral sides of the streak are totally black in most of the cases and the margin of the pygidium touching the elytra is covered with pale setae (Ph. 3) However, a few normal females with laterally black pygidium have a marginal row of whitish setae along with the pale setae (Ph. 4). Sometimes, there are a few pale setae distributed along the meargins of the white streak and over the lateral black areas accompanied by a row of white setae along the anterior margin of the pygidium (Ph. 5) In all these case, the pygidium appears to be dominantly black and the median whitish streak is conspicuous

Abnormal Morph or Pale Pygidium morph (Ph.6 – 8)

The abnormal female has almost entire pygidium covered by the pale setae with a few small bare black patches, making the median white streak less conspicuous. The number of black spots on either side of white streak varies from 2 to 3 in different examples (Ph. 6 – 7). Some abnormal females have their pygidium covered both with pale and white setae (Ph. 8).

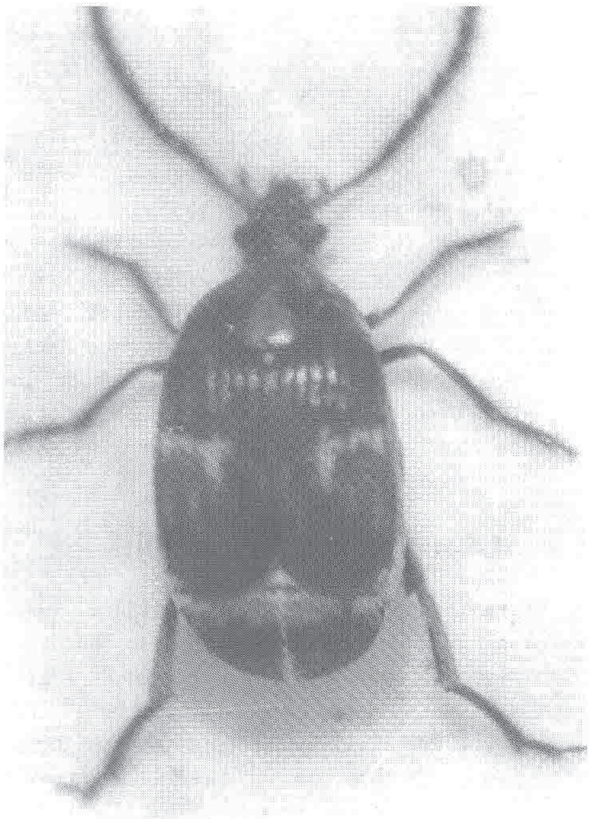


Photo 1. Normal female or black pygidium morph of *Z. Subfasciatus*

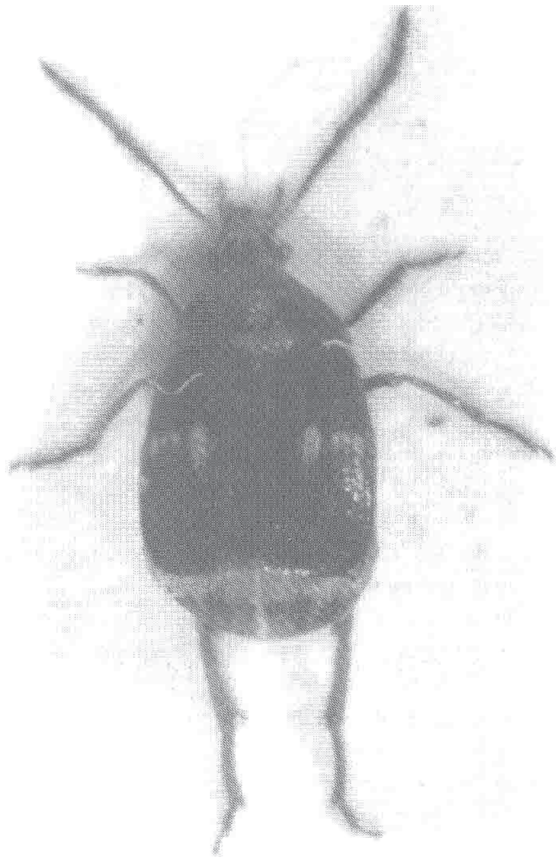


Photo 2. Abnormal female or pale pygidium form of *Z. subfasciatus*

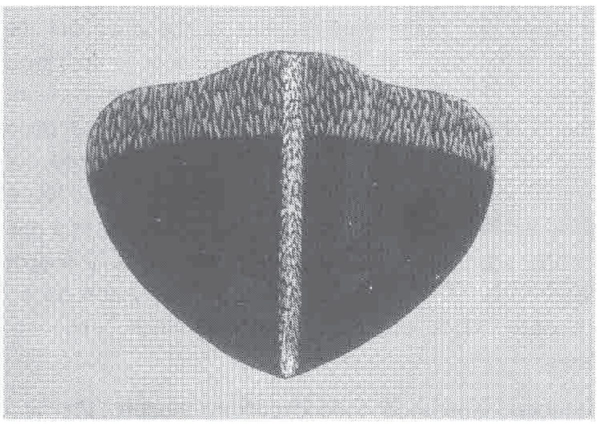


Photo 3. Pygidium of normal female bearing a prominent median longgitudinal white streak with black leteral areas

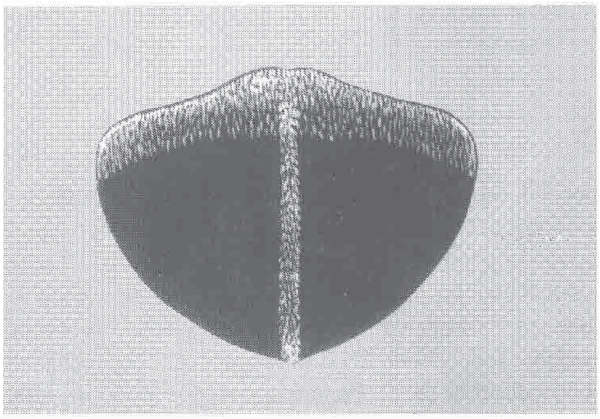


Photo 4. Pygidium of an intermediate black pygidium morph showing the presence of a marginal row of white setae along with the pale setae

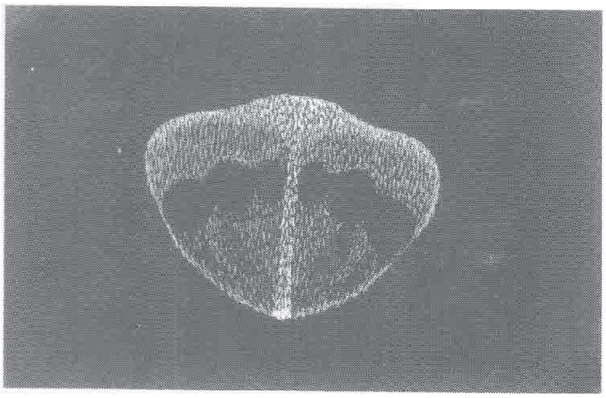


Photo 5. Pygidium of an intermediate black pygidium morph bearing some pale setae on the lateral black areas and arow of white setae along anterior margin of the pygidium

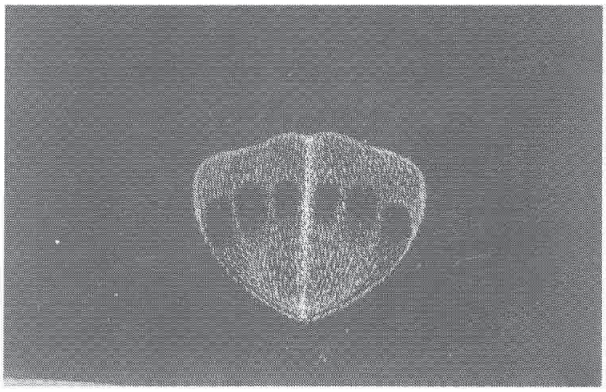


Photo 6. Pygidium of abnormal female covered almost entirely with pale setae leaving tree small bare black patches on either side of the less narrow conspicuous median white streak

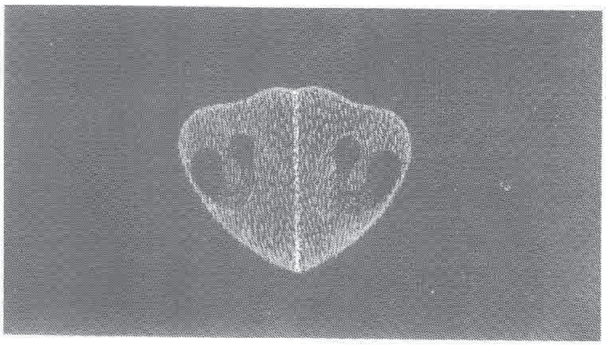


Photo 7. Pygidium of intermediate pale pygidium morph with too small bare black patches on the either side of the white streak

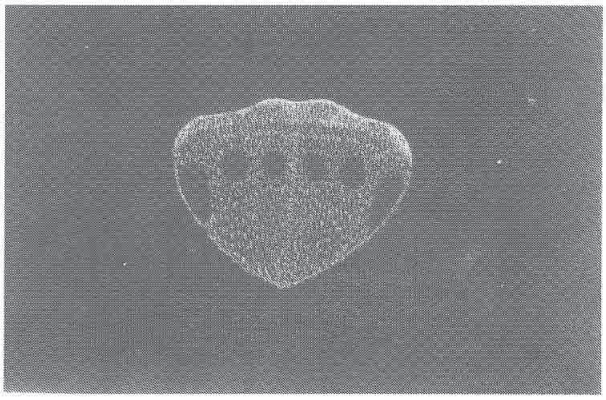


Photo 8. Pygidium of intermediate pale pygidium morph showing the presence of white setae along with the pale setae

In addition to the recognition marks provided by the colour pattern of the pygidium, the two types of females also show differences in the size of different part. As expected, the internal reproductive organs show a very poor development in the abnormal female with different parts like ovarioles, oviducts and bursa copulatrix showing a great reduction in size. Moreover, there is no indication of ova formation in the ovarioles. The spermatheca along with its accessory gland also shows a stunted growth in this morph. Moreover, the abnormal females, being more active and better flier, has relatively longer hind wings and elytra. The dimensions of the pygidium in the normal females are larger than those in the abnormal females, presumably due to normal development of reproductive parts. The data inclided in Table IV show that the differences in the length of some of body parts are significant irrespective of the fact that overall body length is insignificant different in the two types of females. The differences are more significant in those parts, which are associated with the sterility and active flight of the abnormal form.

Antenna

The antenna of *Z. Subfasciatus* is 11-segmented. The scape or the basal segment, attached to the head, is cylindrical. The second segment or pedicel is about half the length of the scape. The remaining nine segments constitute the flagellar segments. Antennae of both the morphs are almost of equal size and appear alike when observed under the microscope (Figs. 1 – 2). However, the sensilla covering thin surface show some differences, which are revealed, through scanning under electron microscope.

Three major types of sensilla have been recognised which are named as sensilla trichodea, sensilla basiconica and sensilla auricillicum (Fig. 3). This terminology has been taken from Callahan (1975) and Wang et al. (1975).

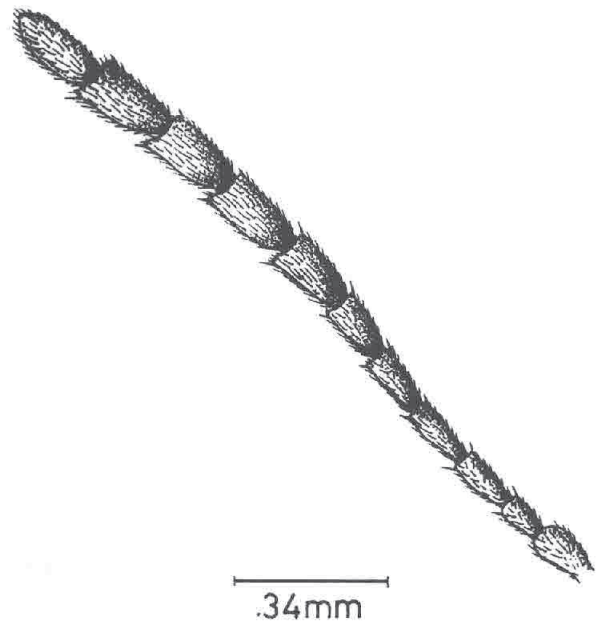


Fig. 1. Antenna of normal female of *Z. subfasciatus*.

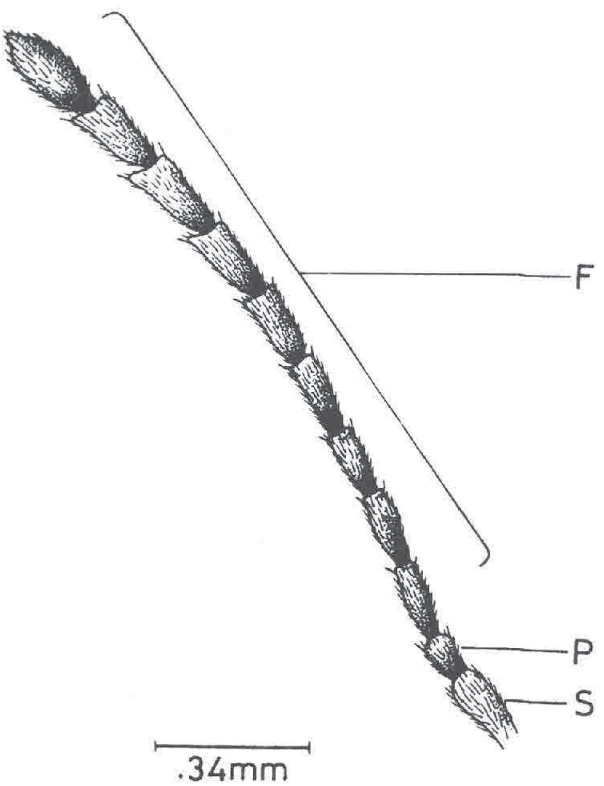


Fig. 2. Antenna of abnormal female of *Z. Subfasciatus*.
F – Flagelum; P – pedicel; S – Scape

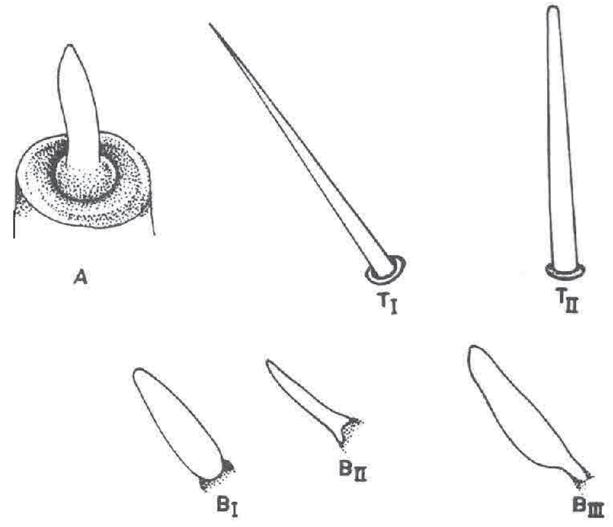


Fig. 3. Types of sensilla.
TI – Sharp-tipped sensilla trichodea;
TII – Blunt-tipped sensilla trichodea;
BI – Blunt-straight sensilla basiconica;
BII – Pointed-straight sensilla basiconica;
BIII – Curved sensilla basiconica;
A – sensilla auricillicum

Sensilla Trichodea

The sensilla trichodea are the true setae. Each sensillum arises from a pit, is broad at base and gradually tapers distally. These are of two types, sharp-tipped and blunt-tipped, the former being comparatively longer. However, both the types show good variation in length.

Sensilla Basiconica

These sensilla are also called peg-like sensilla. Each ensillum is without a prominent socket. Two major types present are the blunt-straight type and the pointed-straight type. However, a few curved sensilla are also sometimes present.

Sensilla Auricillicum

The sensilla auricillicum are short and thick, each of which is fitted in a well developed dome-shaped socket. They ahow variation in shape and size and the type met within the present species is the rabbit-eared shoe-horn type.

Distribution of Sensilla on the Antennal Surface of the Normal Female (Emg. 1 – 11)

The flagellum, the pedical and the scape of the normal female are densely covered with sensilla (Emg. 1). The number of sensilla per segment generally increases from proximal to the distal flagellar segments. All the three types of sensilla described above are present in the normal morph.

Sensilla Trichodea

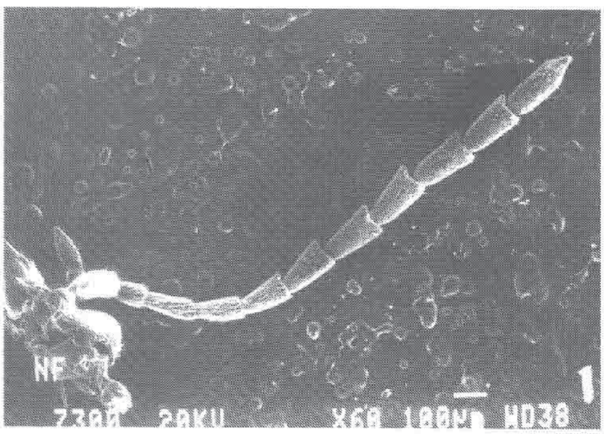
Both the types of sensilla trichodea are present on all the antennal segments, although the blunt-tripped sensilla are restricted only to the tip of the last segment (Emg. 6) and are missing from the surface (Emg. 5). The sharp-tipped sensilla are always more in number on all the segmens (Emg. 11). Inner distal margins of the flagellar segments also show the presence of both the types of sensilla trichodea (Emg. 9 – 10).

Sensilla Basiconica

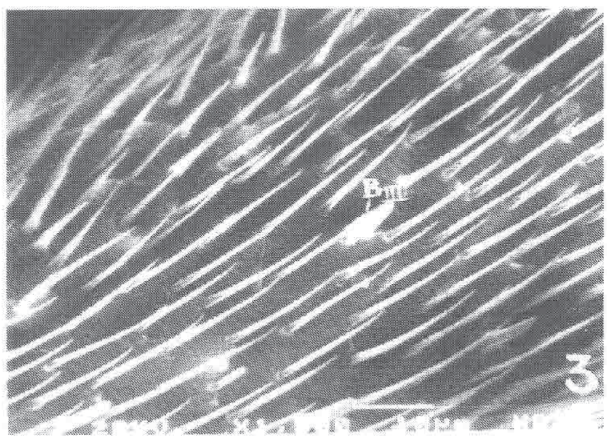
The sensilla basiconica are distributed on the pedicel and all flagellar segments but are missing from the scape (Emg. 2). Blunt-straight basiconica are more in number on all the segments as compared to pointed-straight basiconica. The number of sensilla basiconica is always less in comparison to sensilla trichodea but the number increases from the proximal to distal flagellar segments. The inner distal margins of the flagellar segnments (Emg. 9 – 10) also show the presense of blunt-straight and pointed-straight sensilla basiconica. Curved sensilla basiconica are very few in number and are present only on the pericel (Emg. 3 – 4).

Sensilla Auricillicum

The sensilla auricillicum are present mainly on the last flagellar segment (Emg. 7 – 8) and on the inner distal sensilla are very few in numbers. They are missing from the surface of the flagellar segments, the scape and the pedicel.

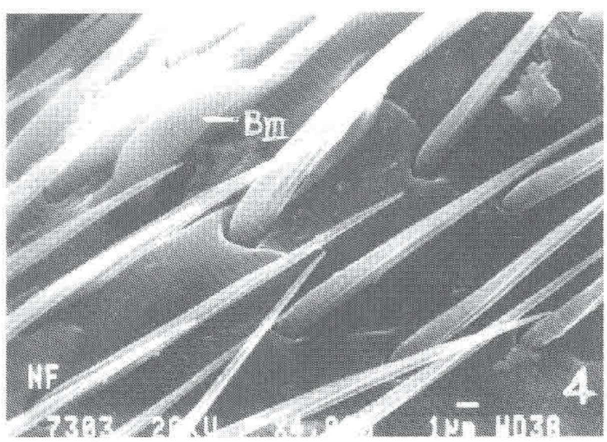


Emg. 1. Antenna of normal female of *Z. subfasciatus* (×60)

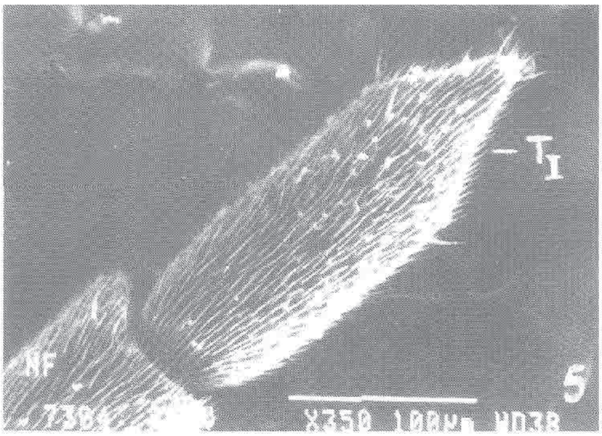


Emg. 3. Distribution of the sensilla on the pedicel of normal female (×1,500)

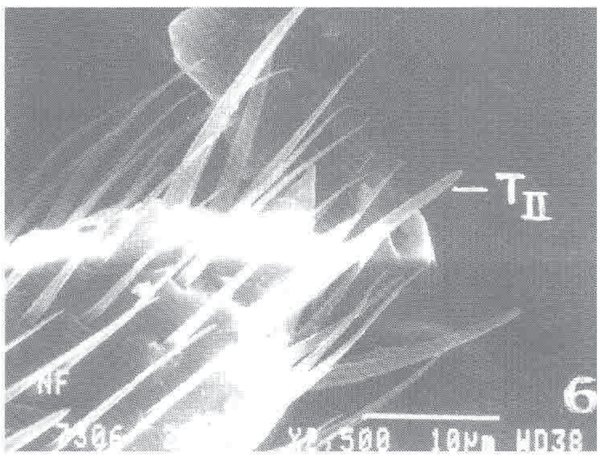
Emg. 2. Scape of the normal female covered with sharp-tipped sensilla trichodea – T_1 (×500)



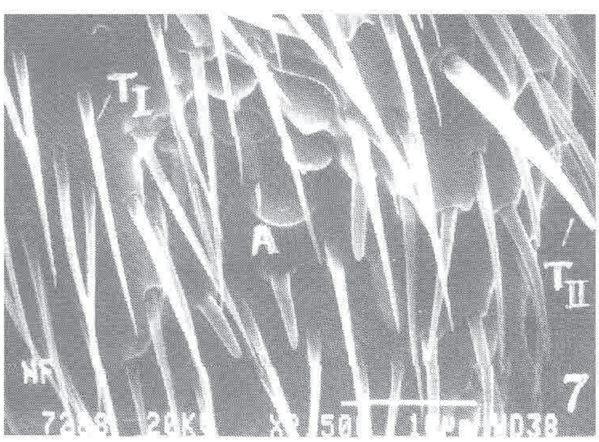
Emg. 4. Magnified view of Emg. 3 showing the presence of curved sensilla basiconica – B_{III} (×4,000)



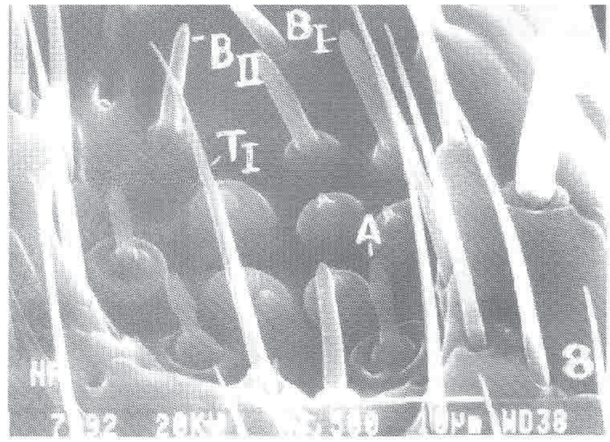
Emg.5. Distal flagellar segment of the normal female(× 350)



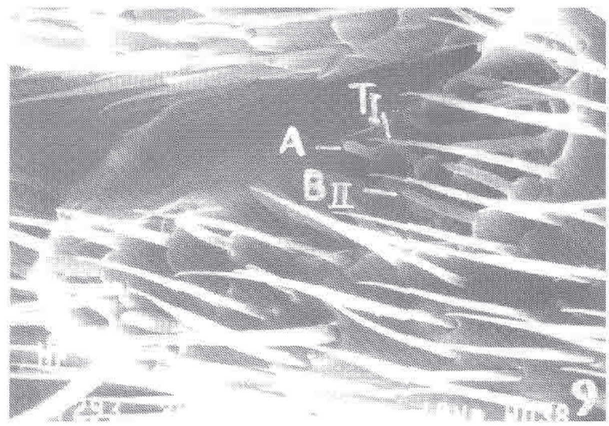
Emg.6. Magnified view of the distal flagellar segment of the normal female showing blunt-tipped sensilla trichidea – T_{II}(× 2,500)



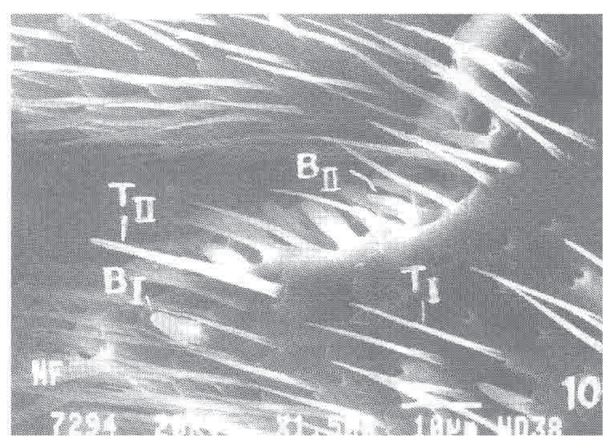
Emg.7. Magnified view of the distal flagellar segment of the normal female (× 2,500)



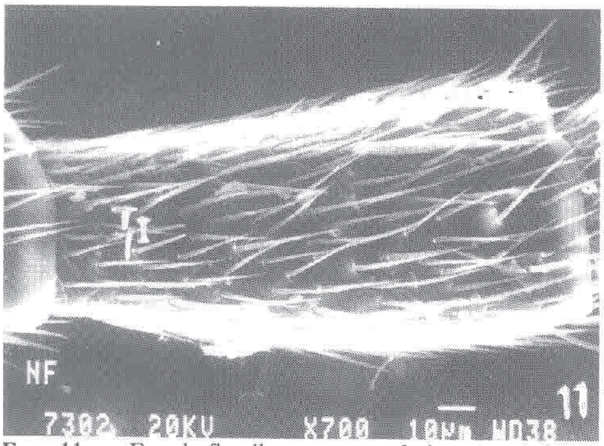
Emg.8. Magnified view of the Emg. 7 to highlight sensilla auricillicum-A, blunt-straight sensilla baciconoca-B_I and pointed-straight sensilla basiconica – B_{II}(× 3,500)



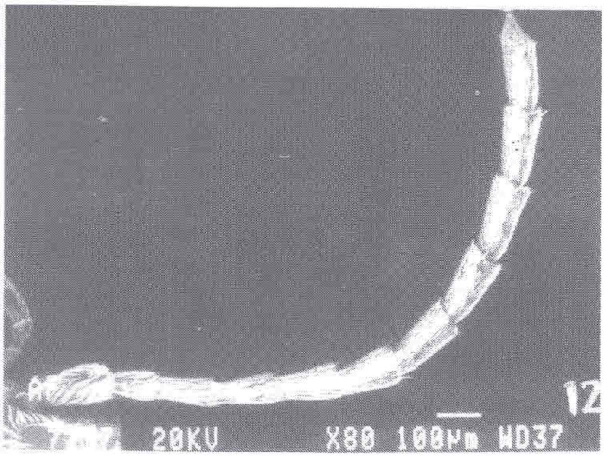
Emg.9. Inner distal margin of the fourth flagellar segment of normal female (× 950)



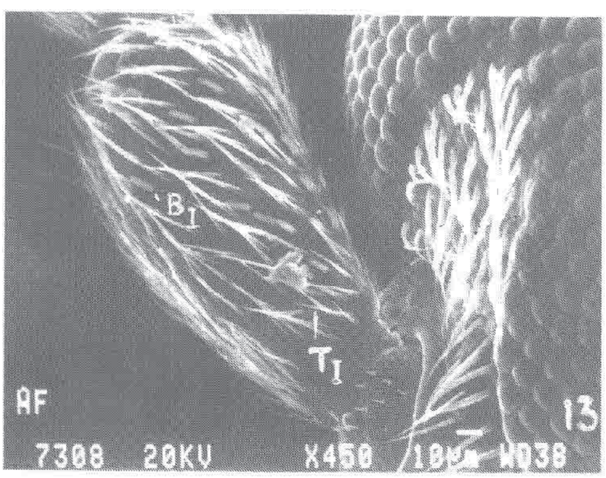
Emg.10. Magnified view of Emg. 9 (× 1,500)



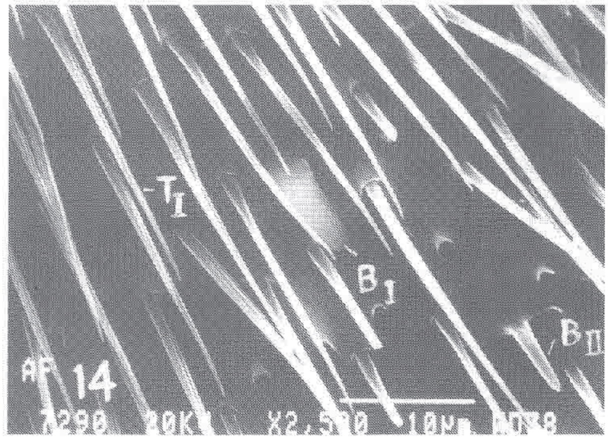
Emg. 11. Fourth flagellar segment of the normal female covered with sharp-tipped sensilla trichodea – T_I ($\times$700)



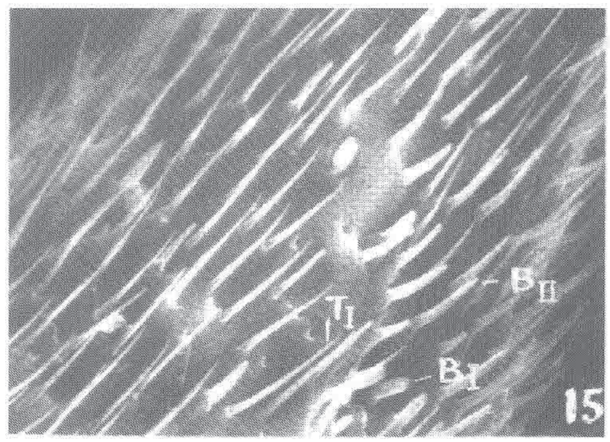
Emg. 12. Antenna of abnormal female of *Z. Subfasciatus* ($\times$80)



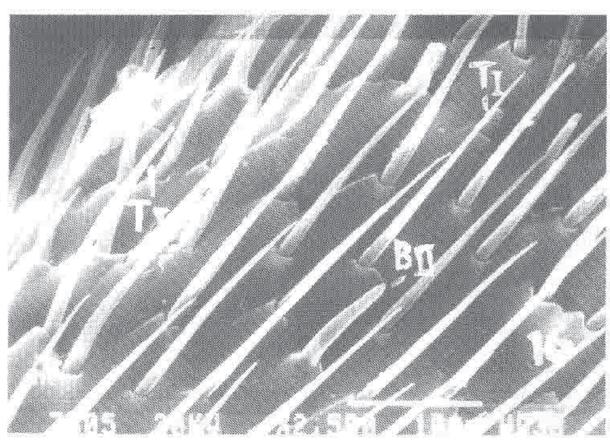
Emg. 13. Scape of abnormal female covered with blunt-straight sensilla basiconica-B_I and sharp-tipped sensilla trichodea-T_I ($\times$450)



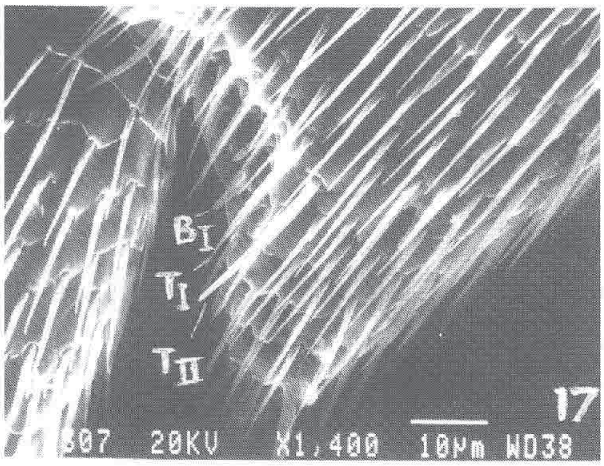
Emg. 14. Distribution of sensilla on pedicel of abnormal female ($\times$2,500)



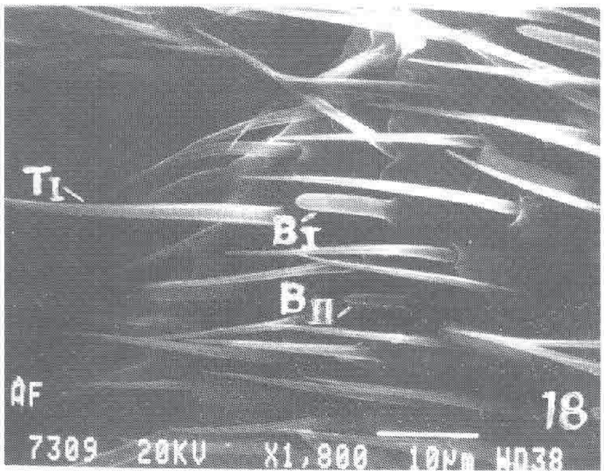
Emg. 15. Distribution of sensilla on the surface of the fourth flagellar segment of abnormal female ($\times$950)



Emg. 16. Magnified view of Emg. 15 (2,500)



Emg. 17. Inner distal margin of the fourth flagellar segment of abnormal female (×1,400)



Emg. 18. Magnified view of Emg. 17 (×1,800)

Differences Observed in the Abnormal Form of Female

The pattern of sensilla on the antennae of abnormal female deffers from that of the normal in several ways. First of all, the sensilla auricillicum are completely absent (Emg. 17 – 18). Secondly, the number of sensilla basiconica is much less as compared to the normal antenna (Emg. 15). Morover, the blunt-tipped trichodea also have reduced number on different segments of the flagellum (Emg. 16). Further, the surface of scape in the abnormal form has only blunt-straight sensilla basiconica (Emg. 13) while they are absent from the scape of the normal form. Again, the curved sensilla basiconica are not present on the pedicel (Emg. 14) unlike then pedicel of normal antenna.

It thus appears that sensilla auricillicum and curved sensilla basiconica which are absent in the antennae of abnormal female have to do something with the perception of certain pheromones produced by the males. In the ansence of these sensilla, the abnormal females do not show any reaction to the male pheromones. At the same time, the number of the sensilla basiconica and sensilla trichodea is also reduced in the abnormal form indicating the requirement of some threshold number of these sensilla for eliciting response in the females.

Reproductive System

The female reproductive system ia consistuted by a pair of ovaries, the paired lateral oviducts, the common oviduct, the genital pouch (vagina + bursa copulatrix), the spermatheca and the accessory gland.

Reproductive System in the Normal Female

The ovaries lie one on either side of the alimentary canal in the second to fifth abdominal segments. Each ovariole inclides the usual parts, i. e., the germarium and the vitellarium. The average length of an ovariole is 0.39mm. The ovarioles of either side open into the corresponding lateral oviduct, which is divided into an anterior broad calyx and the posterior tubular part. The average length of the oviduct is 0.37mm. The two lateral oviducts join with each other to form a relatively shorter duct, which is reffered to, as the common oviduct. The average length of the common oviduct is 0.09mm. It opens posteriorly into the genital pouch which is a larger structure lying on the common oviduct, with its rounded end projecting anteriorly. It consists of two parts, the anterior dilated bursa copuulatrix and the posterior tubular vagina. The chitinous plates which separate the vagina and the bursa in other bruchid species (Singh, 1973) are missing. The genital pouch is 0.26mm long. The vagina narrows posteriorly and leads to the short ovipositor that has a length of 0.13mm. The spermatheca communicates dorsally with the posterior end of the bursa through a short spermathecal duct. The spermatheca is U-shaped, light brown and receives an elongated tubuilar accessory gland at its anterior end. The gland is narrow in proximal region but greatly swollen in its distal part. The average length of spermathecal duct, spermatheca ans accessory gland is 0.20, 0.085 and 0.30mm respectively.

Differences Observed in the Reproductive System of the Abnormal Female

The reproductive system of abnormal female is quite rudimentary in comparison to the well developed system in the normal female. Most of the parts show reduction in length (Table 1) and have stunted growth. The size of the ovariole is reduced to more than half and each measure 0.17mm in length. There is no indication of ova formation. The two lateral oviducts also undergo reduction in their length (0.19mm), lie very close to each other and the common wall between them is not clearly demarcated. The common oviduct is still shorter, i. e., 0.03mm in length.

The average length of the genital pouch is reduced to 0.18mm that of the ovipositor to 0.08mm. The spermatheca is not properly chitinized and the accessory gland surrounds the spermatheca in the form of a watch-spring in contrast to the free accessory gland in the normal female.

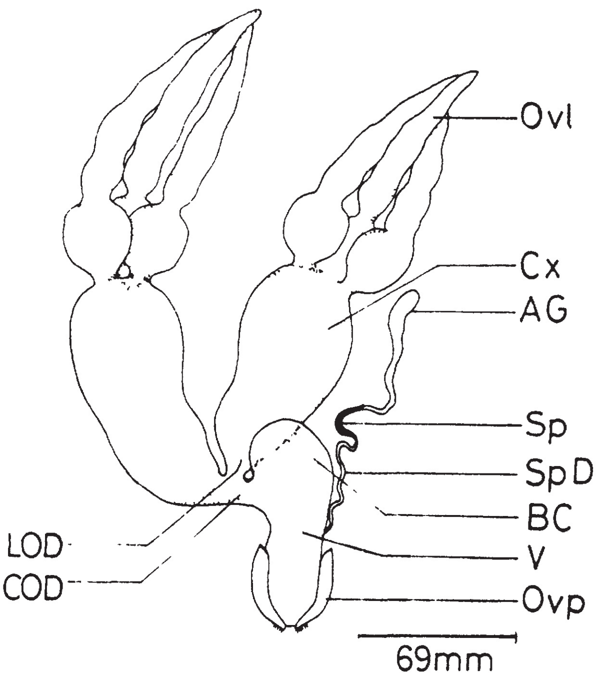


Fig. 4. Reproductive system of normal female of Z. Subfasciatus

AG – Accessory Gland; BC – Bursa Copulatrix; COD – Common Oviduct; Cx – Calyx; LOD – Latweral Oviduct; Ovl – Ovariole; Ovp – Ovipositor; Sp – Spermatheca; SpD – Spermathecal Duct; V – Vagina

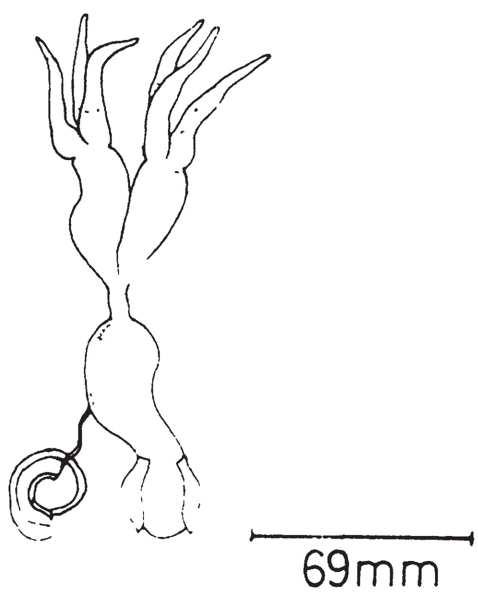


Fig. 5. Reproductive system of abnormal female of *Z subfasciatus*.

Table 1. Morphometric measurements of the normal and the abnormal female of *Z subfasciatus*.

S No Body Parts	* Average length (mm) females		Significance (at 5%)
	Normal	Abnormal	
Antenna(l)	1.90	1.89	NS
Elytra (l)	1.69	1.86	S
Elytra (w)	0.83	0.84	S
Pygidium (l)	0.75	0.56	S
Pygidium (w)	0.74	0.53	S
Hind Wing (l)	2.75	3.50	S
Hind Wing (w)	0.87	1.12	S
Body length (without Antenna)	3.03	2.92	NS
Ovariole (l)	0.39	0 17	S
Lateral	0.37	0.19	S
Common	0.09	0.03	S
Spermathecal	0.20	0.18	NS
Spermathecal (l)	0.95	0.090	S
Accessory Gland (l)	0.30	0.21	S
Genital Pouch (l)	0.26	0.18	S
Ovipositor (l)	0.13	0.09	S

* based on 10 observations; l = length; w = width; S = significant; NS = Non-Significant

Discussion

The occurance of two types of females in *Z Sunfasciatus* with black and pale pygidium has also been previously reported by Pajni (1986a, 1986b). There is no other report on the dimorphism of this species in India or in other regions.

As far as other bruchid species are concerned, dimorphism in *C. Maculatus* is quite well known. The two forms, i e., the normal and the active, can be distinguished from the elytral and pygidial colour patterns. The normal form according to Utida (1954) is brownish whereas the flight form has tan body colour Southgate et al (1957) reported that the active form has completely black elytra which are dark brown in the normal form. They did not say anything about the colour pattern of thepygidium. According to Arora and Pajni (1959), the sterile female has the pygidium covered with white or pale setae while the normal female has black pygidium with a median band of white setae They further stated that the normal male has the elytra covered with brownish setae or at most a small part in the midddle is scarcely covered with setae and shows the black ground colour as against the more prominent black areas in the sterile male. Caswell (1960) also described the white-pygidium active female different from the black-pygidium normal female. He separated the two types of males on the basis of the colour pattern of the elytra. Arora et al. (1967) found the structure of male genetalia in the abnormal male entirely different from the normal male. The structure of the chitinized area at the base of bursa copulatrix has been found to show differences in the two types of females (Spirina, 1974) Kapoor (1980) found the sterile and normal morphs of Indian strain differing in the length of elytra, hind wings, abdomen and pygidium desides differences already described in the colour pattern of the pygidium and the elytra as well as the structure of the male genitalia Yasui (unipuplishe from Utida, 1972, 1981) reported the active and the normal forms differing in body weight, as well as the length of wings, pygidium and the body. Further studies on the Indian strain by George and Verma (1994) showed that there were five morphs found in *C Maculatus*, three of which corresponded to the normal/flightless form and the other two forms corresponded to the active/flight form of earlier authors. Different morphs differ in the body colouration and pubescence rotation of aedeagus, wing length, body weight, size of male accessory gland, size of bursa copulatrix and basal egg chambers of ovarioles, in addition to other discriminatory characters

In the absence of any other report on the structure of antenna in the two forms of *Z. Subfsciatus*, the results of the present study indicating the presence of three types of sensilla namely sensilla trichdea, sensilla basiconica and semsilla auricillicum is accepted for the time being for comparison with the antenna of the dimorphic/polymorphic forms of other bruchid species Earlier studies on *C. Maculatus* and *C. Chinensis* (Rip, 1988; Pajni and Gupta, 1989, 1990) have revealed the presenceof only two types of sensilla in both forms of *C. maculatus* and normal form of *C chinensis*. The presence of sensilla auricillicum has not been reported in any other bruchid species which has been detected in the normal female of *Z Sunfasciatus* but is missing in the sterile female. The results of the present study showing the reduction of sensilla trichodea and sensilla basiconica in the sterile female of *Z. Subfasciatus* gets support from the findings of Pajni and Gipta (1990) who have reported a similar eduction in the sterile males of *C maculatus*.

Earlier reports on the internal reproductive system of *C maculatus* (Arora and Pajni, 1959; Caswell, 1960; Utida, 1972, 1981) show that the ovaries and testes as well as the associated genital ducts present extremely stunted growth in the abnormal sexes The fat bodies have also been reported to be present in large quantities in the abnormal form (Utida, 1972, 1981). These findings are in agreement with the results of be present studies on the abnormal female of *Z Subfasciatus* which also show a poor development of reproductive system and excessive deposition of fat bodies.

Coming to the morphological differences in the discovered morphs of *C chinensis*, Applebaum et al (1968) were the first to record difference in the antennae and the arrangement of setae on the prothorax and the pygidium of two strains procured from Israel and Japan. Neither of these strains was, however, associated with any abnormality in behaviour or reduction in fecundity. Pajni (1986a, 1986b) and Pajni et al. (1987) recorded three different types of females in the laboratory culture of this species amd called these forms as 'Brown pygidium', 'White pygidium' and 'Black pygidium' females. Datta (1990) confirmed the occurrence of these three morphs and also reported a few forms intermediate between the major three categories. The forms of this species also show difference in the time of their appearance and in their fecundity, with the least fewcund white-pygidium form making its appearance during the summer months. The abnormolity in the females of *C chinensis* is accordingly on the lines of *C maculatus* and *Z. Subfasciatus*, the abnormal forms of which are totally infecund, have white pygidium and make their appearance during the summer part of the year.

Coming to *C analis*, Pajni (1986a, 1986b) reported two types od females and four types of males in this species differing in the colour pattern of their elytra and pygidia Bhartu (1990), on the contrary, reported three types of females and five types of males. The rcorded females of the three types also show difference in their fecundity and time of appearance in the laboratory Tiwary and Verma (1989a, 1989b) described three types of individuals or 'phases' namely the normal form, the active form and the intermediate form. The three phases exhibit characteristics morphological, developmental and behavioural features. Moreover, the normal male shows clockwise rotation of the aedeagus as agaonst the anticlockwise rotation of the aedeagus in the abnormal male.

Taylor (1981) examined the pygidium of *Callosobruchus subinnotatus* (Pic) and a few other bruchids and expressed the opinion that differences in the colour pattern of the pygidium appear to be common and hence the dimorphism in this family is supposed to be a wide spread phenomenon.

Observation of different workers on the occurrence of two or more morphs in different species of bruchids reveal that in most cases, the white pygidium females are generally less fecund than the females with black pygidium The less fecund or sterile females are also characterized by other behavioural and physiological features

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