

HISTOLOGY AND HISTOCHEMISTRY OF DEVELOPMENT AND RESORPTION IN THE TERMINAL OOCYTES OF *LEPTOCORIS COIMBATORENSIS*

AMARJIT KAUR, B. KISHEN RAO,
S. S. THAKUR and S. SABITA RAJA
*Department of Zoology, Osmania University,
Hyderabad 500 007, India.*

CARBOHYDRATES, proteins and the lipids are the three main metabolites which play an important role in egg production. A combined diet of sugars and protein is thus an essential constituent for egg production¹. Lipids are also essential for reproduction². According to Monroe³, 20% of eggs will not hatch if *Musca domestica* is fed on a diet deficient in cholesterol. Deprivation of food will also lead to changes in the gonads and their glands. Dietary insufficiency of nutrition, on the reproduction is not a direct one; it affects reproduction through the corpus allatum (CA) which secretes the gonadotropic hormone. Hence an attempt was made to study the distribution of the main metabolites in the ovaries after deprivation of food and after treatment of the adults of *Leptocoris coimbatorensis* with hydroprene (JHA).

The adults of *Leptocoris coimbatorensis* were fed on soaked soapnuts and were maintained at $30 \pm 1^\circ\text{C}$ and RH of $65 \pm 5\%$. In an experiment the freshly ecdysed adult females were completely deprived of food but provided with water and in another experiment the freshly eclosed females were applied topically with $1 \mu\text{l}$ of $0.01 \mu\text{g}/\mu\text{l}$ /insect of hydroprene. These experiments were repeated thrice. The ovaries of the experimental insects were dissected and processed for histological and histochemical studies. Paraffin sections of aqueous bouins fixed ovaries were stained with periodic acid Schiff's (PAS) stain for carbohydrates⁴, for glycogen the sections were stained with Best's Carmine⁵ whereas acid mucopoly-saccharides were stained by thionin stain. For better results both glycogen and acid mucopolysaccharides were demonstrated by a combination of PAS and alcian blue stain⁴. For the localization of lipids, paraffin sections of Bakers calcium formal fixed ovaries were stained in Sudan black B stain⁶ whereas for the study of proteins sections of Carnoy fixed ovaries were stained in mercuric bromophenol blue⁷. Nucleic acids were demonstrated by methyl green pyronin method⁸ and gallocyanin⁹. The volume of CA was calculated according to Tobe and Pratt's procedure¹⁰.

Histological and histochemical studies

(i) *Control adults*: The ovaries of the control adults showed that normally vitellogenesis started on the second day after emergence. At this time the follicular epithelium underwent the morphological changes and changed from columnar to a layer of thin elliptical cells at the completion of yolk deposition. Histochemical analysis of the different metabolites, carbohydrates, lipids, proteins and nucleic acids in the trophocytes, follicular epithelium and the oocytes showed a gradual daywise increase in these metabolites reaching a maximum on day 4 (the day of completion of vitellogenesis). The distribution of these metabolites is shown by the intensity of the stain taken by the tissue. The volume of CA increased daywise and reached its peak level on the 4th day after adult emergence (table 1, figure 1).

(ii) *Starved adults*: The ovaries of most of the starved adults did not show yolk deposition and if it did, there was rapid resorption of the oocytes. The follicular epithelium did not show the morphological changes as in the controls and this may be the reason for the resorption of the oocytes. Histochemical studies showed that carbohydrates, lipids, proteins and nucleic acids decreased rapidly. Negligible amounts of these metabolites were observed on day 4. The volume of CA also showed a rapid fall in its activity. (table 1, figure 1).

(iii) *Hydroprene treated adults*: The treated adults showed the initiation of yolk deposition, but before it could come to completion, there were signs of resorption of the oocytes. The follicular epithelium underwent the normal morphological changes in most of the oocytes but soon it took over the function of resorbing the yolk in the oocytes, thus performing a dual function. Studies on the distribution of the different metabolites showed an increase initially but gradually the amount decreased as the oocytes were undergoing resorption. However the intensity of the stains showed that the decrease in the metabolites was not so rapid as in the starved adults. The volume of CA also showed that in most of the cases the level was found to fall compared to the controls. (table 1, figure 1).

It was found in the present study that all the metabolites gradually decreased in the ovary as it was undergoing resorption. The glycogen and acid mucopolysaccharides were utilised rapidly for the supply of energy to the insect. The other metabolites were also greatly reduced. It may be assumed that this fall in the amount of these metabolites may probably be due to

Table 1 Histochemistry of the ovariole of control, starved and Hydroprene treated adults (4 days old)

Metabolites	Germarium			Vitellarium			
	TC	D	FE	DC OP	FE	TC OP	
Glycogen	+	+	+	++	+	++++	C
	-	-	-	-	-	++	S
Carbohydrates	+	+	+	+	+	++	T
Acid	+	+	++	+	+	+	C
mucopolysaccharides	-	-	-	-	+	-	S
	-	+	+	-	+	+	T
Lipids	++	+++	+++	++	+++	++++	C
	+	-	+	+	++	+++	S
Proteins	++	+++	++	+	++	+++	T
	+++	+++	+++	++	++	++++	C
	++	+	+	+	+	++	S
	++	++	++	++	+	+++	T
RNA	+++	+++	+++	++	+++	++++	C
	+	-	+	+	+	++	S
Nucleic acids	++	++	+	+	+	++	T
DNA	++	++	++	+	++	+	C
	+		+	-	+	-	S
	+	+	+	-	+	-	T

++++ Very strong reaction; +++ strong reaction; ++ moderate reaction; + weak reaction; - negative reaction.
 TC—trophocytes; D—developing oocytes; DO—distal oocytes; FE—follicular epithelium; OP—ooplasm; TO—terminal oocytes; C—control adults; S—starved adults; T—Hydroprene treated adults.

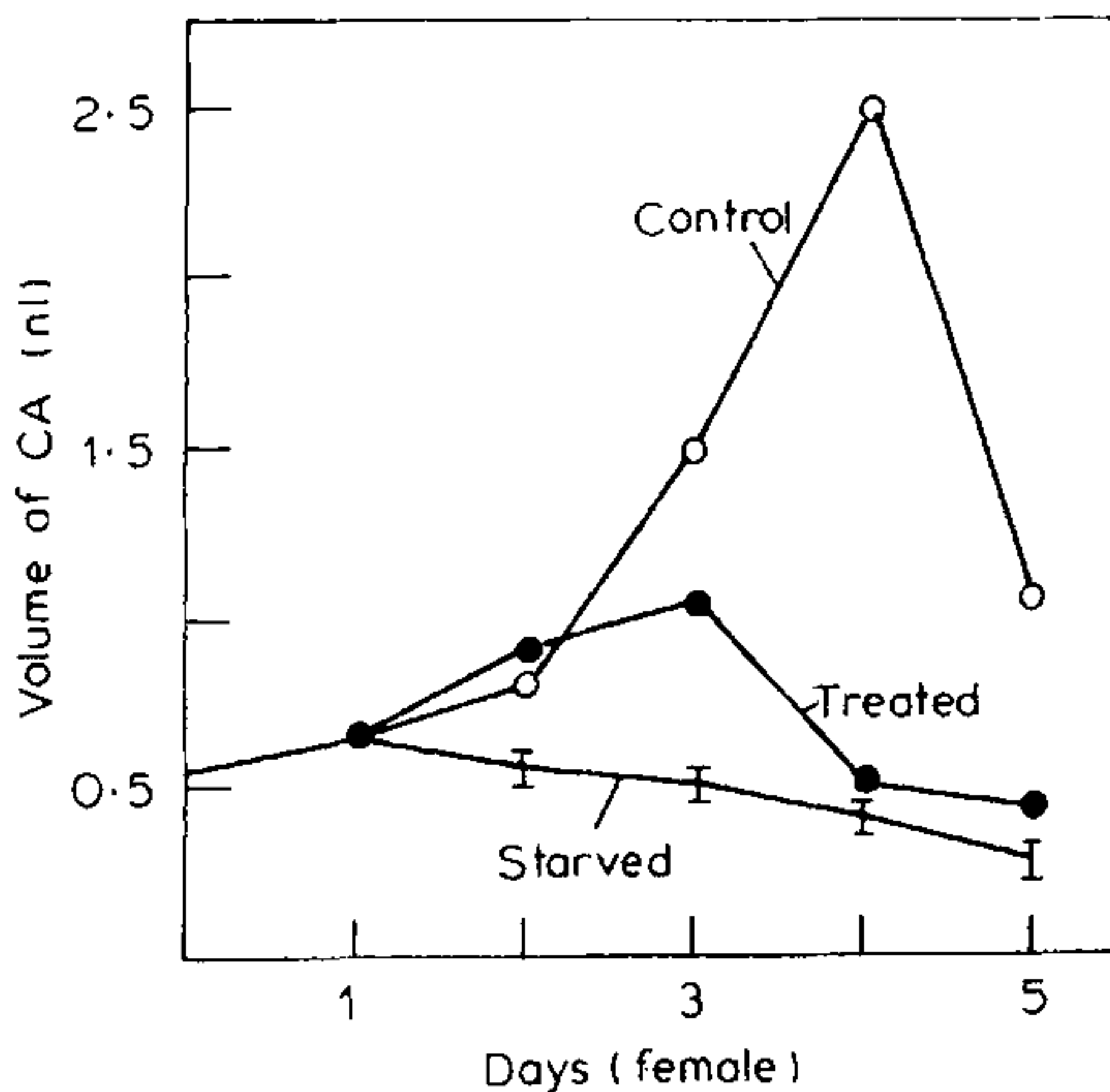


Figure 1. Volume of corpus allatum in the control, starved and hydroprene-treated adults.

the starvation; hydroprene treatment generally decreases the metabolic activity by interfering with the synthetic activity of the corpus allatum (CA). During starvation or after hydroprene application, the normal hormonal balance is disturbed, hindering the metabolic activity, as such the insect does not have sufficient reserve food material for its survival. Hence it tries to utilize whatever metabolites, especially the carbohydrates present in the ovary, thus accounting for the depletion of these metabolites in the gonad. Similar reports were presented by Jutsum *et al*¹¹ and Minks¹². The failure of the follicle cells to differentiate properly may be the cause of resorption of the oocytes which is a JH dependent event or the paucity of JH leads to oocyte resorption¹³. Hence we see that there is a specific interaction of hormonal and dietary factors directly influencing the ovary. Thus malnutrition or starvation or interrupted feeding due to Hydroprene treatment results in a partial or complete developmental arrest.

The authors (AK and SR) thank UGC, New Delhi for financial assistance.

25 November 1985; Revised 15 February 1986

1. Rasso, S. C. and Fraenkel, K., *Ann. Ent. Soc. Am.*, 1954, **47**, 639.
2. Vanderzant, E. S., *J. Insect Physiol.*, 1963, **9**, 683.
3. Monroe, R. E., *Nature (London)*, 1959, **184**, 1513.
4. Pearse, A. G. E., *Histochemistry, theoretical and applied.*, 1968, III edition, Vol. 1.
5. McManus, J. F. A. and Mowry, R. W., *Staining methods: histologic and histochemical*, New York, Paul B Hoeber and Co. 1960.
6. McManus, J. F. A., *Nature (London)*, 1946, **158**, 202.
7. Mazia, Daniel, Brewes, Philips, A. and Alfred Max., *Biol. Bull.*, 1953, **104**, 57.
8. Kurnick, N. B., *Stain Technol.*, 1955, **30**, 213.
9. Einarson, L., *Acta Pathol. Microbiol. Scand.*, 1951, **28**, 82.
10. Tobe, S. S. and Pratt, G. E., *J. Exp. Biol.*, 1975, **62**, 611.
11. Jutsum, A. R., Agarwal, H. C. and Goldsworthy, G. J., *Acrida*, 1965, **4**, 47.
12. Minks, A. K., *Archs Neerl Zool.*, 1967, **17**, 175.
13. Bell, W. J. and Bohm, M. K., *Biol. Rev.*, 1975, **50**, 373.

ADDITIONAL EARLY CAMBRIAN (BOTOMIAN) BRACHIOPOD FOSSIL LOCALITIES IN TAL FORMATION, LESSER HIMALAYA, INDIA, AND THEIR SIGNIFICANCE

C. TRIPATHI, G. KUMAR, S. MEHRA,
D. K. BHATT, V. K. MATHUR,
ASHUTOSH JOSHI and B. S. JANGPANGI
Geological Survey of India, Lucknow 226 006, India.

THE recent find¹ of an assemblage consisting of ten taxa of fossil brachiopoda of Early Cambrian (Botomian Stage) from the Tal Formation of Lesser Himalaya, Uttar Pradesh, is of great biostratigraphic significance. This discovery was made from a shaly horizon in the basal part of the 'Upper Tal', viz Quartzite Member² or Phulchatti Quartzite Member³, in the southern limb of the Mussoorie

Syncline from a locality southeast of Loarkha. In view of its great importance a thorough search for new localities was made during May 1985, which resulted in finding of two more fossiliferous localities in the northern limb of the Syncline. The localities belong to the same stratigraphic level and contain the same fauna as recorded earlier¹. A brief account of the new localities and the significance of this brachiopod level are discussed in this paper.

(i) Betagad (Jabarkhet) section

The brachiopod yielding horizon consists of light grey micaceous shale within massive quartzite of Phulchatti Member about 260 m above its base, and is well exposed on the Dehradun-Mussoorie-Chamba road cuttings, about 425 m before Jabarkhet (Betagad) Toll barrier (figure 1). Only the upper 7 m thick sequence of shale is exposed while the lower part is covered (figure 2). The brachiopods occur as black dots mostly sporadic on bedding surfaces measuring 1-2 mm in diameter. One of the rare, relatively better preserved brachiopod taxon *Obolus* sp, is illustrated (Figure 3).

(ii) Marora section

This is located on the Maldeota-Kaddukhal-Dhanaulti road, about 100 m downstream of the bridge over Song river at Marora (figure 1). Here also the brachiopod fauna occurs in 26 m thick shale horizon within the thick quartzite beds of the Phulchatti Member 130 m above its base. The shale sequence reveals two fossiliferous bands interspersed

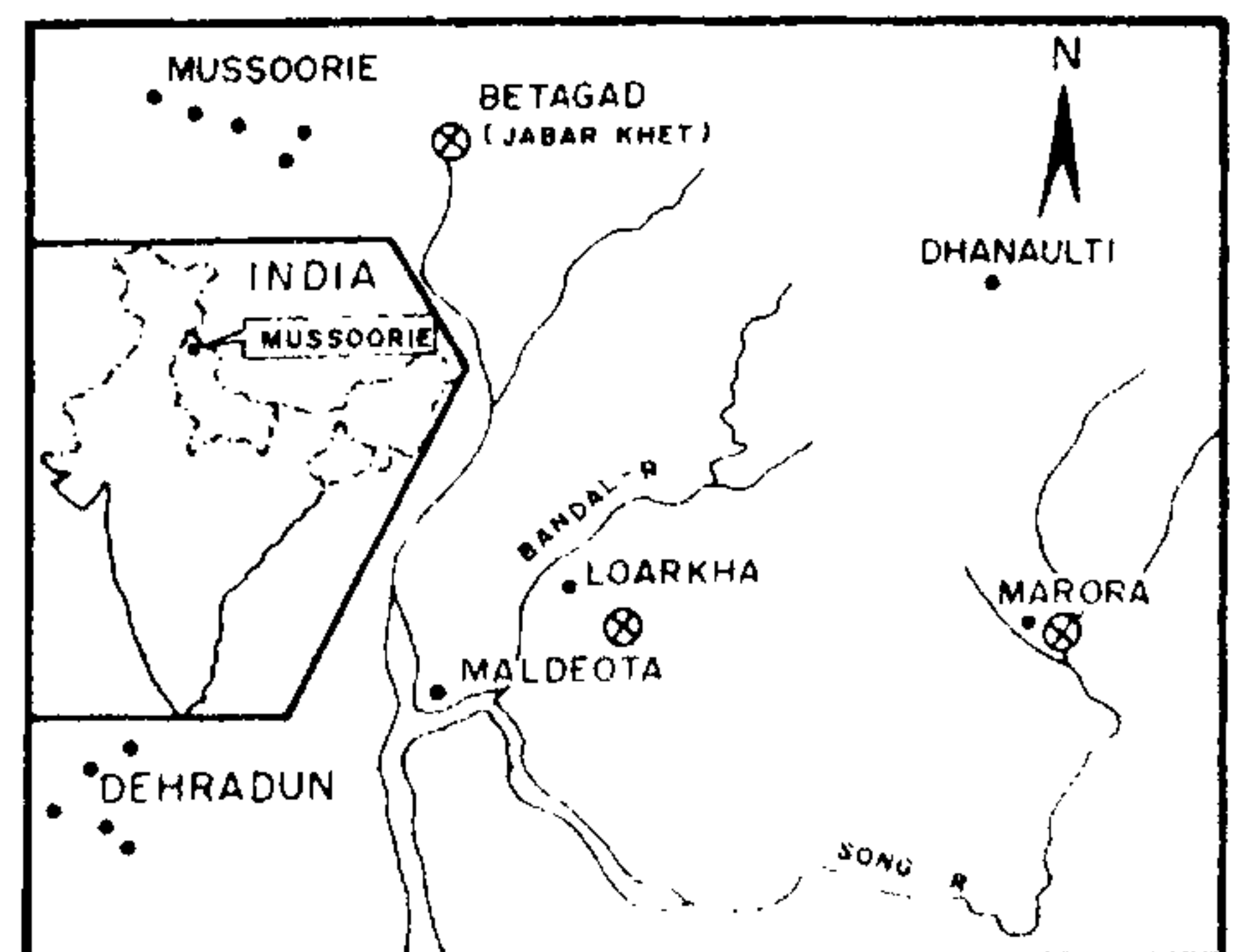


Figure 1. Map showing fossil localities.