from August to November, causing a maximum 32% parasitism. It was found to parasitize the earlier nymphal instars (up to 10 days old) of F. virgata. The parasitoid completed development in 25-30 days. Only one adult emerged from a parasitized mealybug. It was found to be uniparental. Adults lived for 18-24 days at  $25\pm2^{\circ}$ C and 60-70% RH under laboratory conditions. This is the first record of B. insularis on F. virgata in India. The previous records of B. insularis parasitizing F. virgata are from the Philippines<sup>6</sup> and Congo<sup>7</sup>.

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## SPERMATOGENESIS DURING ONTOGENY IN THE BLACK-HEADED CATERPILLAR OPISINA ARENOSELLA WALKER (LEPIDOPTERA: XYLORYTINAE)

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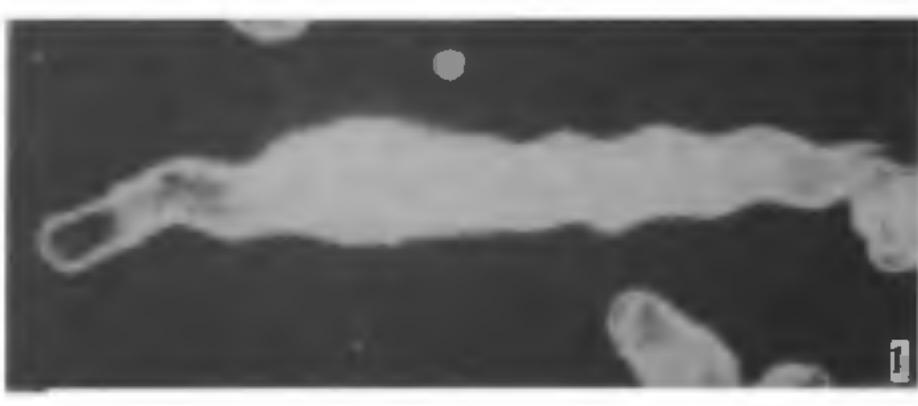
DETAILED information on spermatogenesis of insects is often essential for studies on endocrine control of spermatogenesis and in sterilization studies. Hence spermatogenesis during ontogeny of the testis in Opisina arenosella, a major pest of coconut palm, was studied and the results are reported here.

O. arenosella, which has eight larval instars, was maintained in the laboratory as described earlier. The testis from different larval instars, prepupa, pupa and adult were fixed and processed by routine histological methods. Phase-contrast microscopy

was also used, on teased preparations.

The larval testes of O. arenosella are a pair of kidney-shaped organs attached to the body wall on each side of the mid-dorsal line in the 5th abdominal segment. The testis of the I instar consists of only primordial germ cells. In the II instar follicles begin to differentiate and predefinitive spermatogonia are observed. The III instar testis consists of four distinct pyriform follicles. Each testis is enclosed in a peritoneal membrane. Within the testis each follicle is bound externally by a membrane, the capsula lobuli, and internally by another membrane, the tunical interna. The four follicles are covered by another membrane, the membrana communis, which is seen just beneath the peritoneal membrane. The apical cells (9-11  $\mu$ m diameter) are distinguishable by their poorly stainable cytoplasm. Predefinitive spermatogonia are arranged around the apical cells. The former undergo mitotic division and give rise to definitive spermatogonia. In the IV instar, definitive spermatogonia, through a series of mitotic divisions, give rise to a cluster of cells, which are enclosed in a cellular envelope, the cyst. The cyst contains primary spermatocytes; the V instar shows secondary spermatocytes. Spermatids are visible in the VI instar. As the spermatids elongate, the nuclei of eupyrene spermatids orientate in a band, arranged together at one end of the cyst. Eupyrene spermatozoa first occur as sperm bundles in the penultimate larval instar (figure 1). The testes of both sides fuse to form a single spherical structure during the prepupal period. The apyrene sperm bundles are distinguishable in day-1 pupa (figure 2). Thus in day-1 pupa there are both apyrene and eupyrene sperm bundles. Torsion of the fused testis occurs in, approximately, the day-6 pupa. In the newly emerged adult the major portion of the testis is occupied by sperm bundles. Though the eupyrene sperm bundles are observed in the late larval stages in this insect, the apyrene sperm bundles appear later, only in the early pupal period, as in Ostrinia nubilalis<sup>2</sup>, Porthetria dispar<sup>3</sup> and Papilio xuthus<sup>4</sup>, but unlike in Trichoplusia ni<sup>5</sup> and Heliothis virescens<sup>6</sup>, where both types of sperm bundles first appear simultaneously only in the early pupal period.

Phase-contrast studies on preparations of teased testis and seminal fluid from vas deferens, seminal vesicles and spermatophore, as well as from copulatory pouch and seminal receptacle of females after mating showed that the eupyrene and apyrene sperm bundles pass from the follicles into the vas deferens and seminal vesicles in the late papal





Figures 1 and 2. Photomicrographs of two types of sperm cysts (×600) in *Opisina arenosella* Walker.

1. Eupyrene sperm bundle, and 2. Apyrene sperm bundle.

period, during which process the apyrene sperm bundles separate into spermatozoa. Apyrene spermatozoa and eupyrene sperm bundles are observed in the vas deferens and seminal vesicles. No loose eupyrene spermatozoa are found in the male reproductive organs. In Bombyx mori<sup>7</sup> it has been observed that the apyrene sperm bundles separate while passing through the basement membrane of the testis. On the other hand, in Trichoplusia ni<sup>B</sup> and Anagasta kuhniella<sup>9</sup>, the apyrene sperm bundles separate immediately after passing from the follicle. Sequential phase-contrast studies on preparations from different regions of the male reproductive duct reveals that the apyrene spermatozoa show movement for the first time in the spermatophore, which is formed in the male ejaculatory duct. The spermatophore contains apyrene spermatozoa, eupyrene sperm bundles and secretory products of the male accessory reproductive gland. In B. mori<sup>10</sup> it has been reported that the motility of the apyrene spermatozoa is brought about by the secretory substance of the ejaculatory duct of the male. The spermatophore of O. arenosella breaks down in the copulatory pouch of the female and the contents of the spermatophore are released. With release, the spermatozoa show vigorous movement. Sequential phase-contrast studies show that the eupyrene sperm bundles of O. arenosella are separated into spermatozoa in the copulatory pouch of the semale and here they mingle with the active apyrene spermatozoa, as in B. mort<sup>11</sup>. In O. arenosella, as in B. mort<sup>10</sup>, the eupyrene spermatozoa acquire motility only when they reach the seminal receptacle. The present studies show that eupyrene sperm bundles are already differentiated in penultimate (7th) instar larva of O. arenosella and hence, for effective sterilization the insect has to be treated before this stage.

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## ENHANCEMENT OF BIOGAS PRODUCTION USING ALGAE

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THE green algae Zygogonium spp. were used to enhance the production of biogas from cow-dung. The factors that determine biogas production, viz. percentage of total solids, total organic matter<sup>1</sup>, chemical oxygen demand<sup>2</sup> and carbon to nitrogen ratio<sup>3</sup>, favour increased biogas production in