



**Figures 1 and 2.** Photomicrographs of two types of sperm cysts ( $\times 600$ ) 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>8</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 female and here they mingle with the active apyrene spermatozoa, as in *B. mori*<sup>11</sup>. In *O. arenosella*, as in *B. mori*<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 *Zygonium* 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

**Table 1** Effect of addition of algal slurry to cow-dung on biogas production

Bottle	Starter (ml)	Fresh cow-dung slurry (ml)	Algal slurry (ml)	Total volume of biogas evolved (ml)	Duration of biogas production (days)
I	100	260	0	4600	26
II	100	260	150	7859	31
III	100	260	300	10893	42
IV	100	260	450	14183	53

digesters with the algae as higher amount of digestible organic matter is present in these digesters than in those containing cow-dung alone.

The cow-dung slurry was maintained under anaerobic conditions and the pH was noted daily. After four weeks, the slurry contains large numbers of methanogenic bacteria, which convert all organic matter to methane under anaerobic conditions. At this stage the cow-dung slurry is called starter. A small amount of starter was mixed with fresh slurries of cow-dung and algae in different proportions in batch digesters. The digesters were kept at room temperature ( $25 \pm 2^\circ\text{C}$ ) in anaerobic conditions and the volume of biogas evolved (measured as volume of water displaced) was noted daily. The results are presented in table 1.

The amount of biogas produced from algae is twice that obtained from cow-dung (on dry weight basis 1 g of dry algae produces 344 ml whereas 1 g of dry cow-dung produces 179 ml). The duration of gas evolution increased with increase in the proportion of algal slurry. The calorific value of the gas was  $4800 \text{ kcal/m}^3$  and the percentage of methane was 55.43.

It can therefore be suggested that algae can be added to cow-dung in biogas plants, especially during winter, when production of gas from cow-dung alone is low. The digested sludge can be used as a good manure.

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## IMMOBILIZATION OF EXTRACELLULAR LIPASE PRODUCED BY *ASPERGILLUS JAPONICUS* IN RESPONSE TO PLANT LATEX

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THE power of enzymes as catalysts for organic chemical reactions is diverse and remarkable. Among such biocatalysts are extracellular microbial lipases (glycerol ester hydrolase EC 3.1.1.3), classified among the hydrolases; lipases catalyse the hydrolysis of fats and oils to give diacylglycerols, monoacylglycerols, glycerols and fatty acids<sup>1</sup>. They have been used in hydrolysis of oils and fats<sup>2</sup>, flavour development in dairy products<sup>3</sup>, ripening of cheddar cheese and improving the flavour of processed and blue cheese<sup>4-7</sup>. The reversibility of the lipase reaction allows use of the enzyme as a catalyst in the formation of esters from alcohols and fatty acids<sup>8,9</sup>, and also to change the physical properties of mixtures of fats and oils by altering the distribution of fatty acyl groups among triglycerides.

In our laboratory, a hydrocarbon-degrading strain of *Aspergillus japonicus* was found to produce extracellular lipase<sup>10</sup>. Induction of lipase was possible on an easily available and cheap carbon source, like latex of *Calotropis gigantea*. The enzyme was purified 77-fold in two steps of purification. The lipase (molecular weight 156,000) was stable at room temperature for a month<sup>10</sup>. Keeping in view the extensive applications of lipases, and to further its industrial applicability, we report the immobilization of latex-induced lipase from *A. japonicus* and the properties of the immobilized enzyme.

### Immobilization of lipase on Dowex<sup>(R)</sup>

The growth conditions for *A. japonicus* and