

dumbbell-shaped (Fig. 4). A few nuclei show karyorrhexis. The external ovariolar sheath breaks at many places. Granular fibrous degeneration could not be observed even after 23 days of treatment.

The experiment clearly shows that hempa's action resembles that of apholate, metepa and thiotepa as described by Tandon and Bhargava² in producing vacuolar degeneration and breaking the interfollicular tissue after thinning. The effect, however, is very slow and does not affect all the oocytes and ovariolar. It differs from all the three chemicals, in not producing granular and fibrous degeneration, possibly due to its different chemical nature. The experiment shows that hempa damages the nuclei more than cytoplasm. It is also interesting to note that only mature oocytes are damaged to a larger extent. Therefore, the possibility of overcoming the effect of chemosterilant cannot be ruled out. LaBrecque *et al.*³ also mentioned the recovery of fertility after hempa treatment in female houseflies. Morgan⁴ also expressed his doubt that the affected cells may be able to overcome the effect.

According to Lüscher and Engelmann⁵ and Scharrer and von Harnack⁶, a decrease in the amount of cytoplasm, presence of vacuoles and densely packed nuclei with clumped chromatin mass are indications of inactivity and degeneration of corpora allata. Exactly the same picture was obtained by Bhargava and Mathur¹ after hempa treatment. These authors reported that corpora allata show early signs of degeneration near about 48 h after the treatment when vacuoles appear in the peripheral cytoplasm (Fig. 7) and nuclei tend to move towards the centre. Almost at the same time the oocytes also show early signs of degeneration. Prolonged treatment increases the number of vacuoles (Figs. 8, 9 and 10) and the nuclei undergo pycnosis in corpora allata. Simultaneously, the ovarian tissue shows increased degeneration. After 240 h the size of corpora allata is reduced (Fig. 10) and the mature oocytes show a complete picture of degeneration. Allatectomy has similar effects in many insects. It causes oosorption in *Schistocerca*⁷ and stops vitellogenesis in insects of various orders⁸. According to Girardie⁹ egg maturation in *Periplaneta americana* also is controlled by corpora allata. A conclusion may, therefore, be drawn from the above experiment that the damage of the ovarian structure in *Periplaneta americana* is because of the inactivity of corpora allata which in turn is induced by hempa.

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1. Bhargava, S. and Mathur, R., *Chemosterilant News*, 1975, 3(1), 13.
2. Tandon, G. N. and Bhargava, S., *Curr. Sci.*, 1977, 46(5), 133.
3. LaBrecque, G. C., Morgan, P. B., Meifert, D. W. and Fye, R. L., *J. Med. Ent.*, 1966, 3(1), 40.
4. Morgan, P. B., *Ann. Entomol. Soc. Amer.*, 1967, 60(4), 812.
5. Lüscher, M. and Engelmann, F., *Rev. Suisse Zool.*, 1955, 62, 649.
6. Scharrer, B. and von Harnack, *Biol. Bull.*, 1958, 115, 508.
7. Strong, L., *J. Insect Physiol.*, 1965, 11, 135 and 271.
8. Engelmann, F., *The Physiology of Insect Reproduction*, Pergamon Press, Oxford, 1970.
9. Girardie, A., *J. Insect Physiol.*, 1962, 8, 199.

SOME EXPERIMENTAL EVIDENCE ON THE VIABILITY OF *ASCARIS LUMBRICOIDES* OVA

Introduction

THE prevention of soil-transmitted helminthic infections is mainly a problem of night-soil disposal, which is a major health hazard in a country like India. Recently, sanitarians have evolved a special type of latrine, 'leach-pit bore-hole latrine', in order to produce acceptable manure by composting the night-soil. In view of the health hazard due to the presence of parasitic cysts and ova, people are reluctant to use the digested sludge as manure. It has been shown that even dried, and digested sludge from some sewage plants contains viable ova of *Ascaris*. Keller⁵ (1951), Gotaas⁴ (1956) and Malviya⁷ (1964) reported that, of all the parasitic cysts and ova encountered in sewage sludges, the ova of *Ascaris* were the most resistant to composting. Such resistance is mainly attributed to the egg shell which consists of five layers, namely, an outer proteinous epithelial membrane, three layers of chitinous material and an inner fibrillar lipoidal membrane. Hence, in the present investigation, *Ascaris* ova have been used as the yardstick to assess the survival of parasitic organisms present in the sewage sludges. Experiments were carried out to determine the viability of *Ascaris* ova inoculated to digested sludge samples free from parasitic cysts and ova, using laboratory models, simulating the field conditions as in the leach-pit bore-hole latrines.

Although much work has been done on the influence of various factors, on the development of *Ascaris* ova, yet the findings of various workers regarding the viability of *Ascaris* ova in sewage sludges are at variance. Hence, an attempt has been made to assess, experimentally, the survival of *Ascaris* ova in sewage sludges using laboratory models.

Material and Methods

The viability of *Ascaris* ova in sewage sludges has been determined experimentally, by inoculating sludge samples with *Ascaris* ova and examining microscopically, as was done by Keller and Hide⁶ (1951). Fertilised *Ascaris* ova isolated from 15 adult worms were inoculated to two sets of digested sludge samples free from any intestinal parasitic cysts or ova. The samples were transferred to pyrex glass tubes (24" × 1.5" dia). The tubes were wrapped with a black paper to avoid the probable effect of sunlight and mounted vertical as shown in Fig. 1. Precautions were taken to keep the sludge moist and to maintain the field conditions.

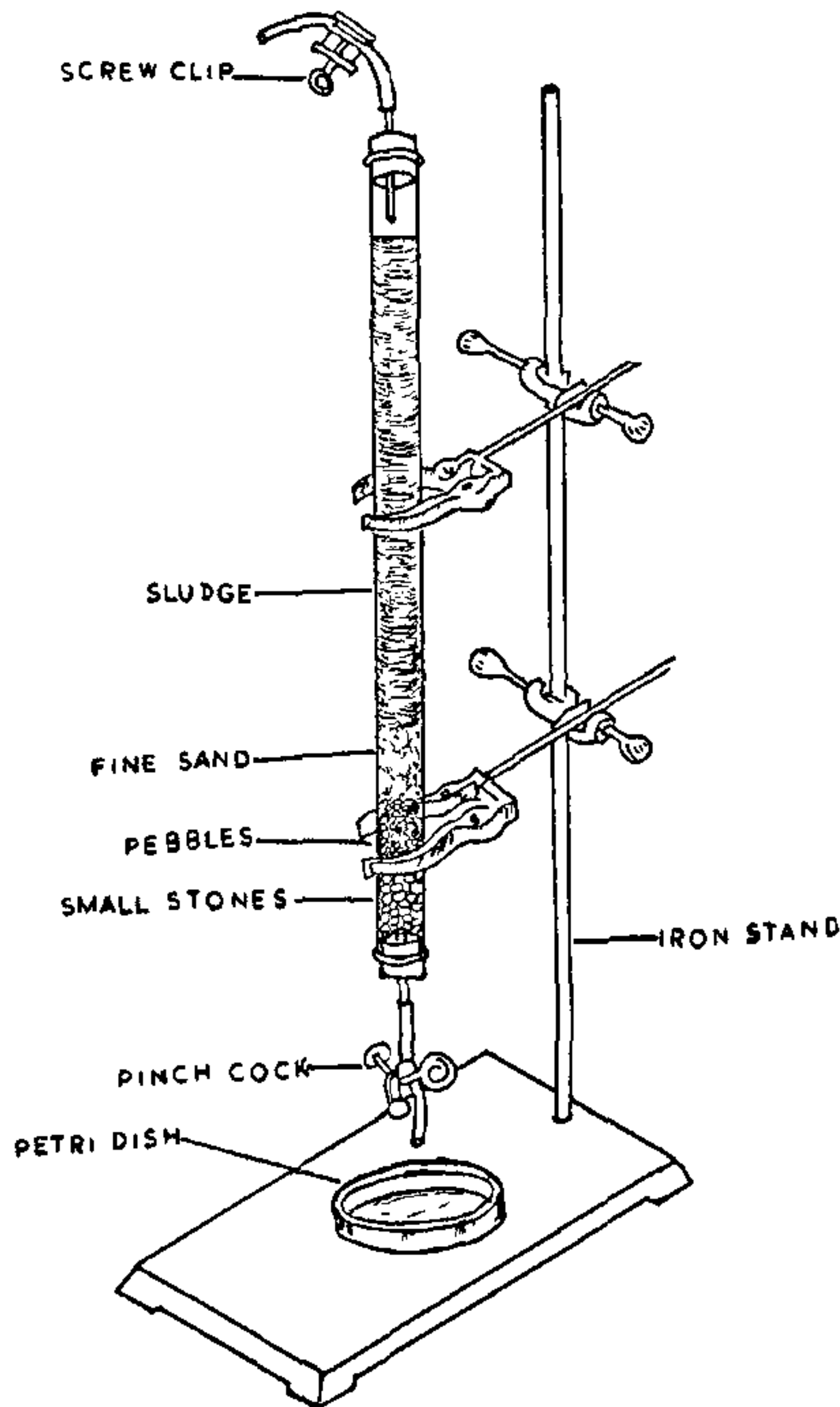


FIG. 1. Laboratory model used in the experiments.

Following the method of Malviya⁷ (1964), viability of *Ascaris* ova was determined by isolating and culturing the ova at 37° C in a medium consisting of 5% formalin in physiological saline solution. Culturing and microscopic examinations were repeated at an interval of every two weeks. To determine the viability, the criterion used by Keller⁶ (1951) was followed.

Results and Discussion

There has long been uncertainty as to the conditions which induce hatching of *Ascaris* ova. Several workers have investigated the survival or development of *Ascaris* ova in relation to different environmental

factors such as temperature, season, etc., (Obha⁹, 1923; Ogata¹⁰, 1925; Brown¹, 1927; Faust², 1939; Gartner and Muting³, 1951; Keller and Hide⁶, 1951; Gotaas⁴, 1956; Susikaran¹², 1963; Malviya⁷, 1964; Petrokova¹¹, 1967). During the present investigation, it has been observed that fertilised ova of *Ascaris*, under natural conditions, developed into active embryos within three weeks. Yoshida (1918) found that in favourable environment, embryos appear within the egg shell after a fortnight. Similarly, Manalang⁸ (1927) observed that under experimental conditions, *Ascaris* ova developed into motile larva from 9 to 15 days at room temperature. Gartner and Muting³ (1951) reported that *Ascaris* ova did not survive longer than 1½ years in land irrigated with sewage. Faust² (1939) reported the longest survival period of 5 to 6 years for the ova found in contaminated soil.

In the present study, the results of microscopic examinations proved that the viability of *Ascaris* ova is 50% after 1 year; 25% after 1½ years; 12% after 2 years and practically no survival after 3 years in the digested sludge sample. These findings are in conformity with the observations of Petrokova¹¹ (1967).

Although Susikaran¹² (1963) found viable *Ascaris* ova up to 14 months in leach-pit bore-hole latrine containing digested sludge, the results of the present investigation prove that, it is not safe to utilise the sewage sludge as manure, before a minimum period of three years after closing the pit. In view of the above findings, it is suggested, that appropriate regulations are enacted banning the use of wet sewage sludge as manure without allowing for a minimum period of three years after closing the pit.

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1. Brown, H. W., *J. Parasit.*, 1927, 14, 1.
2. Faust, E. C., *Human Helminthology*, 2nd Edition, Lea and Febiger, London, 1939, pp. 780.
3. Gartner, H. and Muting, L., *Ztschr. f. Hyg. u. Infektionskr.*, 1951, 132, 244.
4. Gotaas, H. B., *Composting—Sanitary Disposal and Reclamation of Organic Wastes*. WHO Monograph Series, 1956, No. 31, pp. 205.
5. Keller, P., *J. Proc. Inst. Sew. Purif.*, 1951, Part I, p. 100.
6. —, and Hide, C. G., *South African, Med. J.*, 1951, 25, 338.
7. Malviya, H. M., *Envir. Hlth.*, 1964, 6, 63.
8. Manalang, C., *Philippine. J. Sci.*, 1927, 33, 249.
9. Obha, T., *Jap. Jour. Zoo.*, 1923, 1, 120.
10. Ogata, S., *Ann. Trop. Med. Parasit.*, 1925, 19, 301.
11. Petrokova, I., *Cslk. Hyg.*, 1967, 12, 612.
12. Susikaran, M., *Envir. Hlth.*, 1963, 5, 114.