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FARASITES IN SOIL/SLUDGE SYSTEMS*

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INTRODUCTION

Parasitism has been defined as "an association between two specifically distinct organisms in which the dependence of the parasite on its host is a metabolic one involving mutual exchange of substances. This dependence is the result of a loss by the parasite of genetic information".¹ This paper is structured in such a way that the discussion narrows from general parasitology to parasites of importance in sewage sludge, and further to the effects of specific treatment parameters (heat and ionizing radiation) on the eggs of an important parasite found in sludges, those of the Ascaris species.

ANIMAL PARASITES

It is a fact that most animals have on or within their bodies several species of parasites. In fact, there are more kinds and numbers of animal parasites than there are animals. The major groups of animal parasites are found among the Protista (such as the protozoa), the helminths (flatworms and roundworms) and the arthropods (e.g., insects such as the mosquito). The parasitic relationship can range from permanent, as is the case for tapeworms, to very temporary, as with the mosquito. In most cases the relationship is not particularly detrimental to the host. In fact, a host may gain from the exchange of chemical substances with a particular parasite. In addition, the presence of one species of parasite in a host often prevents the establishment of another species which is potentially injurious.

There are many causes of parasitic diseases, however. One of these is competition for nutrients or other materials by parasites. Another is the effect of parasitic toxins or secretions on the host. Tissue changes may be induced by parasites; for example, some shistosomes have been associated with intestinal tumor development in man.²

Tissue damage may be important, especially where large numbers of parasites are involved. An example is the damage to the human lung during the migration of Ascaris lumbricoides larvae; infected hosts commonly suffer from asthma. Mechanical interference can result from parasitic infection, as is the case with blockages of the intestine and bile ducts by Ascaris worms. Although disease can result from abnormal behavior of a particular parasite, as happens when Ascaris larvae migrate into the heart or brain of the host, most of the time parasitic disease is a function of parasite density.

PARASITES IN SEWAGE AND SLUDGES

Parasites which are to be considered in discussions of application of municipal sludges or sludge products to "accessible" land must satisfy several criteria. Firstly, they must either be intestinal parasites or release eggs or cysts into the intestine to be excreted in the feces of the host. Many protozoan parasites, such as Entamoeba sp., and many flatworms, such as the schistosomes, satisfy this condition, as do nematodes such as Ascaris sp.

Secondly, the parasite eggs or cysts must be capable of infecting the appropriate host. For example, in the case of the schistosomes (blood flukes whose eggs reach the intestine through ulceration) an intermediate snail host is required. Thus, while schistosomiasis affects 200 million people in 71 countries,³ and eggs are introduced into sewage by infected individuals, application of sludges to land in the continental United States poses little hazard since the intermediate snail host is absent.

Thirdly, parasites of primary concern are those which are pathogenic, i.e., disease-causing. Entamoeba histolytica is a protozoan parasite of man which can invade not only the intestine, but other soft tissues.

Extra-intestinal abscesses may occur in the liver (most often), the lungs, or, rarely, the brain. Approximately 600 million people were infected in 1961;⁴ more males than females are infected, but the more serious or fatal complications occur in women and children. On the other hand, another amebic parasite, Entamoeba coli is much more prevalent worldwide (overall incidence of ~28 percent), but it does not invade tissues and is non-pathogenic.

Fourthly, to be a potential health hazard in sludges, parasite eggs or cysts must survive normal sludge treatment processes. Cysts of the pathogen Entamoeba histolytica are present in significant numbers in municipal sludges (one infected person may discharge 45 million cysts per day into the sewer). However, the cysts are killed by drying, or in an efficient anaerobic digestion process. Ascaris lumbricoides ova, on the other hand, remain viable through digestion or lime treatment and can survive on soil for long periods of time.^{5,6}

A recent review paper⁷ on the potential for parasitic disease transmission with land application of sewage effluents and sludges discusses the wide range of parasitic species present in sewage sludges. Hays' literature review concluded that the most prevalent human parasitic eggs and cysts found in sewage in temperate regions are those listed in Table 1.

TABLE 1.
PARASITIC EGGS AND CYSTS PREVALENT IN SLUDGE
IN TEMPERATE REGIONS⁷

SPECIES	PHYLUM	COMMON NAME
<u>Ascaris lumbricoides</u>	nematode	-----
<u>Trichuris trichiura</u>	nematode	"whipworm"
<u>Hymenolepis</u> sp.	platyhelminths	"dwarf tapeworm"
<u>Taenia saginatus</u>	platyhelminth	"beef tapeworm"
<u>Enterobius vermicularis</u>	nematode	"pinworm"
<u>Entamoeba</u> sp.	protozoa	-----

Possibly the most resistant parasite eggs or cysts commonly found in sewage sludge are the ova of Ascaris sp. They are resistant to desiccation,⁸ chemicals,⁹ anaerobic digestion, and relative to other parasite eggs, are resistant to heat inactivation.^{10,11} Since these ova are potentially pathogenic, ubiquitous, and relatively resistant to most forms of treatment, they were chosen as an "indicator" species for inactivation studies, discussion of which constitutes the remainder of this paper.

ASCARIS OVA: INACTIVATION STUDIES

These roundworms reach lengths of 15 to 36 cm. They reside in the small intestine of the host where the female can produce 200,000 eggs per day which are released in the feces of the host, resulting in significant contamination of sewage. These eggs settle readily in a primary wastewater clarifier, so that they are concentrated in the sludge. The population in sludge is still low, however, relative to the numbers required for inactivation rate determinations where reductions by several factors of ten of viable ova must be measured. For this reason, the experimental results reported herein have all been obtained using samples seeded with relatively large numbers of fertilized, non-embryonated ova taken directly from the uteri of worms.

Materials and Methods

The eggs were obtained from pregnant female Ascaris suum worms purchased from Carolina Biological Supply Co. The procedures used for "seeding", radiation or heat treatment, and counting have been described previously.¹² The parameter used to measure inactivation was the ability of fertilized eggs to embryonate after treatment. With no treatment, approximately 90 to 95 percent of the ova will embryonate

in 21 days, so that the "embryonation ratio" is the percent embryonation following treatment relative to that of the untreated eggs.

When liquid sludge was used as the inactivation medium, a modification¹³ of the ether-formalin flotation technique¹⁴ was used. When composted sludge (furnished by SEA/USDA, Beltsville, MD) was the medium, the zinc-sulfate separation technique¹⁵ proved more efficient for recovery of the ova.

Results and Discussion

Bacteriological considerations have led to the determination that at least 300 kilorads will be required for radiation treatment of sewage sludge. Previously published data^{12,16} on the effects of ionizing radiation on Ascaris eggs in seeded liquid systems have shown that in all liquid media tested, including digested sludge, 150 kilorads of absorbed dose is sufficient to prevent embryonation in more than 99.9 percent of the ova present. Recent preliminary data indicate that 150 kilorads delivered to seeded composted sludge is sufficient to prevent embryonation in more than 99 percent of the eggs. Thus, it appears that the dose required for bacterial inactivation (> 300 kilorads) should sufficiently reduce the number of viable Ascaris ova in liquid and dried/composted sludge systems.

Figure 1 shows the effects of heat on the viability of Ascaris eggs in water.^{12,17} Moderate temperatures (>50°C) are effective in preventing embryonation of these eggs in liquid systems. Figure 2 shows the effects of heat on Ascaris eggs added to composted sewage sludge. There appears to be approximately the same threshold

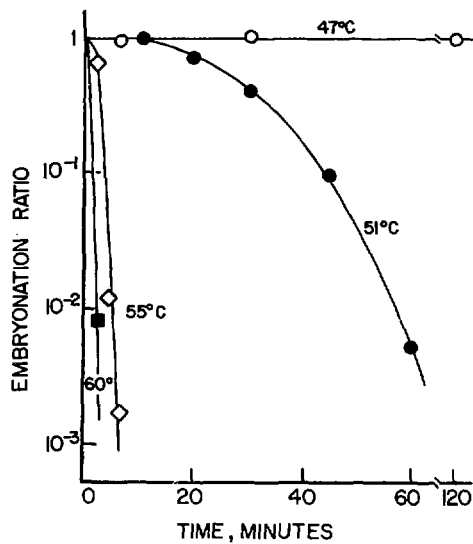


Figure 1. Heat inactivation of *Ascaris* eggs in water

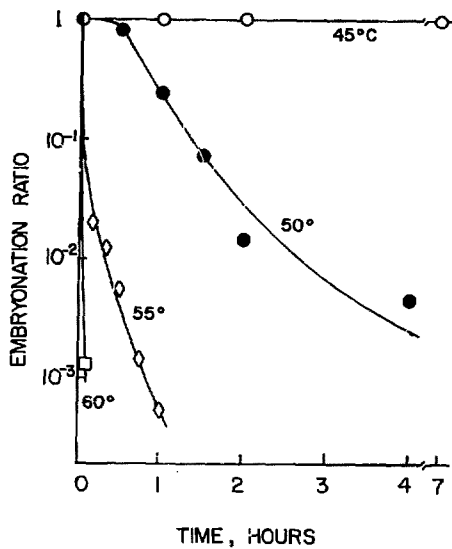


Figure 2. Heat inactivation of *Ascaris* eggs in composted sludge (60 percent solids).

temperature ($\sim 50^{\circ}\text{C}$) for inactivation of the eggs in compost as in liquid systems. However, a comparison of the time scales over which the two sets of experiments were run shows the increased resistance of Ascaris eggs in the composted sludge. Nevertheless, an efficient composting operation, wherein all of the material undergoing composting reaches temperatures in excess of 55°C for relatively long times, would be expected to eliminate viable parasite ova in the finished product.

Conclusions

Studies reported herein have shown that a treatment of 55°C for 1 hour or more sufficiently reduces the number of viable Ascaris eggs in seeded sludge systems. An absorbed dose of 300 kilorads is more than adequate for the same purpose. However, before an unequivocal statement can be made about the effectiveness of either of these treatments in reducing viable ova in "real" systems, certain qualifying factors must be investigated. There are conflicting reports on the radiation sensitivities of Ascaris eggs in different stages of development.^{18,19,20} Also, irradiation of composted sludge using an electron-beam (which, for all practical purposes, is equivalent to gamma irradiation for a given absorbed dose) was unsuccessful in rendering all naturally-occurring Ascaris ova non-viable, even at 300 kilorads.²¹ The significant differences in radiation and heat sensitivities of Ascaris eggs in compost vs liquid systems points out the need to further investigate the effects of moisture levels on these sensitivities. This becomes especially important in the case of treating air-dried sludges to reduce pathogens. It appears, in any event, that efficient composting is effective in preventing embryonation of parasite ova in sewage sludge.

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