Disinfection of Sewage Sludge with Gamma Radiation

L.N. MELMED AND D.K. COMNINOS
[CITY HEALTH DEPARTMENT, LABORATORY AND TECHNICAL SERVICES BRANCH, PO BOX 1477, JOHANNESBURG 2000]

Abstract

Disinfection of sewage sludge by means of gamma radiation to render it suitable for unrestricted reuse as a fertiliser or soil conditioner in the urban environment, was investigated. Inactivation of *Ascaris lumbricoides* ova was used as the criterion of disinfection.

It was found that a total radiation dose of 1 kGy effectively reduced the development of potentially infective larvae in a sludge containing 20% solids, by 99%. The 1% of larvae developing after radiation were infective to white mice. Higher doses of radiation up to 10 kGy did not achieve a 100% kill.

Complete inactivation could be obtained when 0,5 kGy radiation was applied at 50°C to a sludge containing 3% solids and when 0,4 kGy radiation was applied at 55°C to a sludge with 20% solids.

Introduction

The recycling of sewage sludge as a valuable resource is being given increasing attention throughout the world. In the Republic of South Africa, the Health Act No 63 of 1977 makes provision for promulgation of regulations to control the disposal of sewage sludge and consequently standards to permit safe reuse are earnestly being sought. Conventional sludge treatment by anaerobic digestion or aerobic stabilisation does not inactivate all pathogenic bacteria, virus and parasite ova, including *Ascaris lumbricoides* which can survive for 7 years or more in soil (Muller, 1975). The incidence of Ascariasis in the Republic, is very high, therefore it becomes essential to disinfect sludge before it can be released for unrestricted reuse in the urban environment.

Disinfection of sludge can be accomplished successfully by pasteurisation at 70°C for 30 minutes (Stern, 1974) and it is claimed that similar disinfection can be attained by a 3 kGy dose of gamma radiation with ^{60}Co (Süss *et al.*, 1974), 4 kGy of high energy electron irradiation (Trump *et al.*, 1975) and thermoradiation with 0,4 kGy at 47°C (Sivinski, 1975).

To test these claims under local conditions, experiments aimed at sludge disinfection by ionising radiation, thermoradiation and radiation combined with oxygenation were carried out, using sludge from the Johannesburg Olifantsvlei Wastewater Purification Works, which receives domestic sewage from Soweto. Sewage treatment at Olifantsvlei is either biological filtration with mesophilic digestion of the solids or alternately extended aeration activated sludge treatment of the unsettled raw sewage. The waste activated sludge is thickened by dissolved air flotation and then belt pressing. The *Ascaris* ova content of both the digested and waste activated sludge averages 3 000 eggs per gram (dry). Inhibition of development of Ascaris ova to the potentially infective larvae stage on embryonation, was used as an indication of successful disinfection.

Irradiation of the experimental material was carried out by staff of the Radiation Technology Division of the Atomic Energy Board at Pelindaba and viability studies were then conducted at the Johannesburg City Health Department Laboratory. When it was decided to test larvae which had developed subsequent to radiation for the ability to infect susceptible animals, these studies were done in the parasitology laboratory of the Onderstepoort Veterinary Research Institute.

The objectives of the experiment were:-

1 To establish the lowest dose of gamma radiation which would prevent embryonation of *Ascaris lumbricoides* ova in sludge.

2 To determine whether eggs containing embryonated *Ascaris* larvae differ from unembryonated ova in sensitivity to gamma radiation.

3 To ascertain whether a synergistic effect could be obtained when radiation was applied simultaneously with heat (thermoradiation) or oxygen.

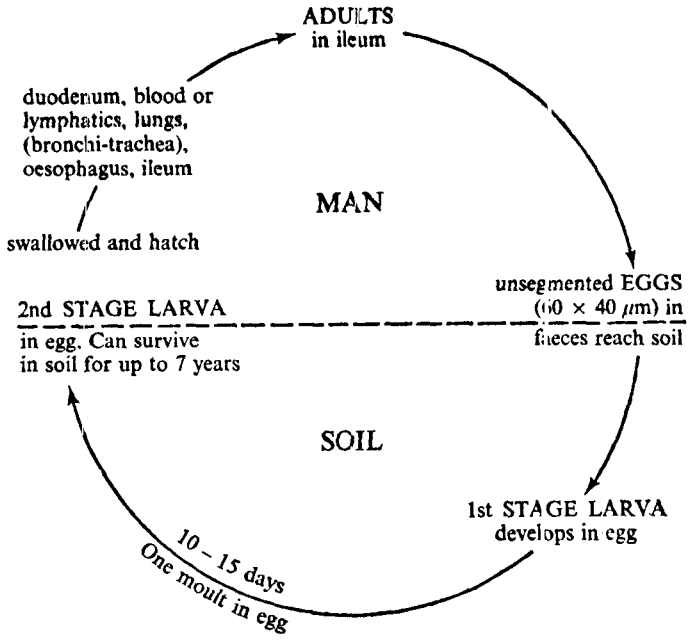


Figure 1
Life cycle of Ascaris lumbricoides *(With acknowledgement to R Muller, 1975 — "Worms and Disease")*

Experimental Procedures

Radiation

The radiation source was ^{60}Co and the dose rate used throughout the experiments was 10 kGy/h.

Controls

Untreated samples were set up with each experiment to give control counts.

Recovery and Embryonation of Ova

Treated and control sludge samples were filtered through a special Helminth filter (Visser, 1972) consisting of an inner nylon mesh bag which traps all gross material but allows the ova and particles of related size to be forced by means of a strong water jet into an outer bag where they are retained until collected into a flask through a tap at the bottom of the outer bag.

The filtrate containing the eggs was formalinised to 2% and incubated at 28°C for six weeks to allow viable eggs to embryonate.

Viability counts were carried out microscopically after embryonation; eggs which had divided or developed into larvae being counted as viable but only those producing larvae were considered to be potentially infective. The latter were regarded as significant survivors of disinfection.

Sludge Irradiation

Waste activated and digested sludge both of which had been partly dried on drying beds (approximately 30–40% solids) were reconstituted to contain 20% solids (equivalent to activated sludge after belt pressing) and filled into 1 *l* plastic containers.

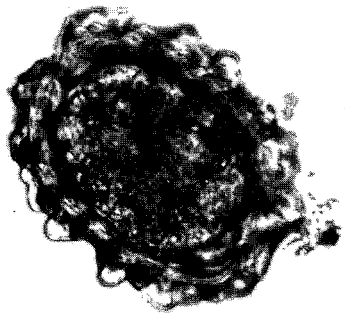
Total radiation doses ranging from 0,5 to 10 kGy at ambient temperature of approximately 22°C were applied to the activated sludge samples. The steps between radiation doses were close up to 4 kGy (0,2; 0,3 and 0,5 kGy) because this was considered to be a critical area, and 1 kGy apart between 4 kGy and 10 kGy.

The digested sludge received total radiation doses of 0,5 to 5 kGy, the first step being 0,5 kGy and 1 kGy thereafter.

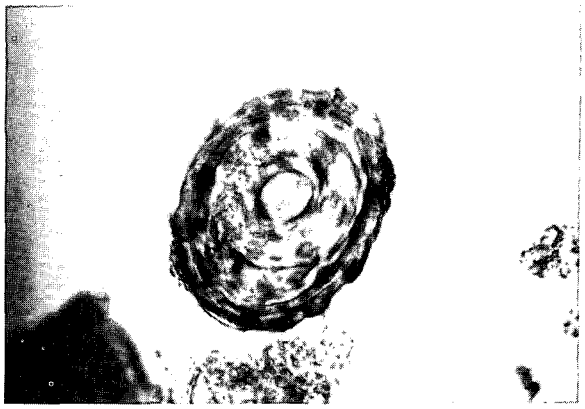
Confirmatory Tests with Animals

For these tests white mice of a strain which had been shown to be susceptible to *Ascaris lumbricoides* were fed with known quantities of stage 2 larvae obtained from plain ova suspension or seeded sludge which had been irradiated and embryonated subsequent to irradiation, or seeded sludge irradiated after embryonation. The ova were obtained by dissecting worms taken from infected children or the sewage works screens. Total radiation doses ranging from 0,3 to 4 kGy were administered at ambient temperature (approximately 22°C).

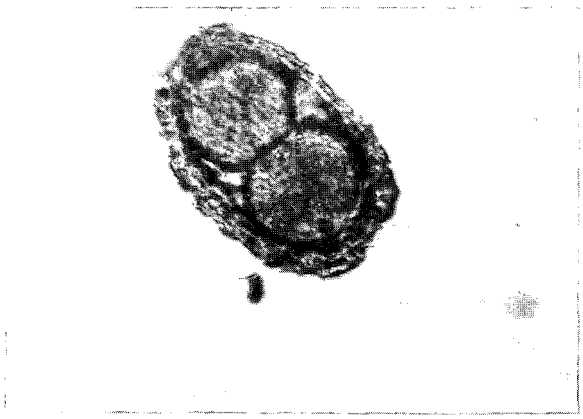
The mice were slaughtered after 4 and 8 days and liver,



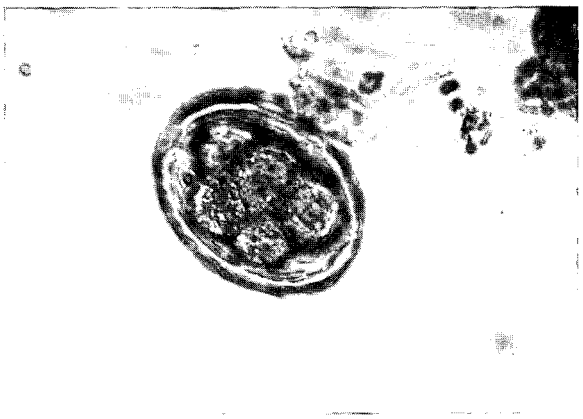
Fertile Unembryonated Egg



Egg Containing 2nd Stage Infective Larva



Embryonating Egg — 2 divisions



Embryonating Egg — morula stage

Figure 2
Embryonation of Ascaris *ova*

lungs and intestine were examined for the presence of worms which were identified microscopically as *Ascaris lumbricoides*. The ability of surviving larvae to migrate to the liver, lungs or intestine was taken as the criterion of infectivity.

Thermoradiation

Samples of waste activated sludge containing 3% or 20% solids were irradiated with total radiation doses of 0,4; 0,5 and 0,6 kGy at temperatures of approximately 22 (ambient); 40; 45; 50 and 55°C respectively.

Oxygen and Radiation

Waste activated sludge containing 3% solids was treated by bubbling oxygen through the sludge for 30 min and then irradiating while continuing oxygenation. Total radiation doses of 0,5 to 3 kGy at 0,5 kGy intervals were applied.

Results and Discussion

Sludge irradiation

The results of activated sludge irradiation are shown in Figure 3 and of digested sludge irradiation in Figure 4.

From these figures it may be seen that in the control sample of activated sludge, 85% of ova were potentially infective whilst in the digested sludge only 44% developed larvae. This would indicate that digestion damages a proportion of the eggs.

After a 1 kGy dose of radiation only 0,7 to 1% of the eggs were still apparently infective except for one sample of activated

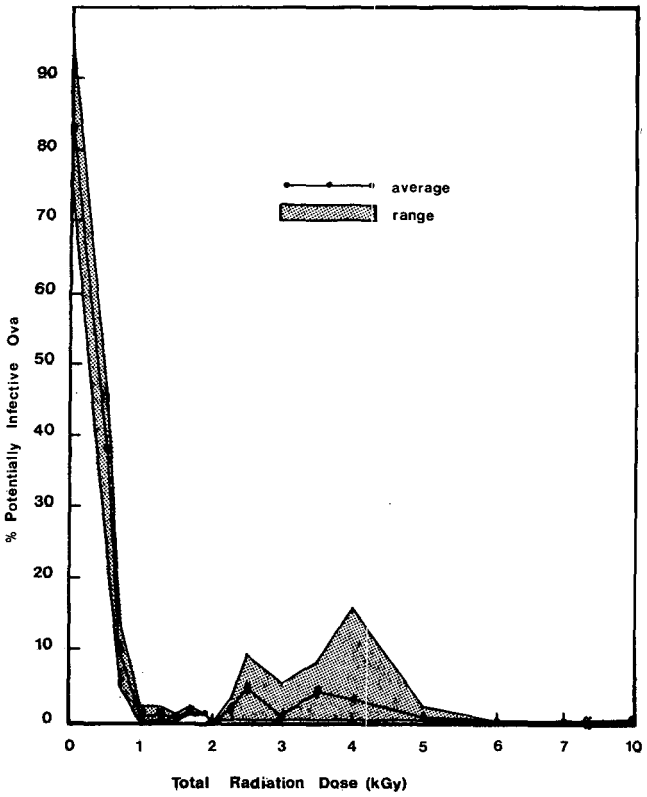


Figure 3
Effect of ^{60}Co *on* Ascaris lumbricoides *ova in activated sludge*

sludge which showed an exceptionally high survival rate and is responsible for the spread of results shown in Figure 3. At a total radiation dose of 5 kGy in both the activated and digested sludges, 0,1 to 0,5% of eggs still developed larvae and in the activated sludge 0,4 to 1,4% of eggs produced larvae after total radiation doses up to 10 kGy. From these results a total radiation dose of 1 kGy appeared to be a critical treatment level.

Taking into consideration the original load of 2 000 — 3 000 ova per gram dry Olifantsvlei sludge, excluding the exceptional sample, the average number of larvae surviving a 1 kGy dose of irradiation would be between 15 — 30/g dry. Higher doses of total radiation did not appear to be advantageous. Consequent to these findings it became essential to establish whether the surviving larvae could infect a susceptible host. Mouse feeding tests were accordingly undertaken in the manner suggested by the work of Verster *et al* (1976) where it was shown that pig carcasses infected with cysticerci of *Taenia solium* could probably be rendered safe for human consumption by treatment with 0,2—0,6 kGy of gamma radiation. This did not kill the cysticerci but rendered them incapable of infection.

A summary of the results of these tests appears in Table 1. The detailed results are shown in Table 2(a), (b) and (c).

For interpretation of the results of animal feeding tests, cognisance must be taken of the fact that since white mice are an unnatural host for *Ascaris lumbricoides,* they were fed with much larger numbers of irradiated larvae than would normally survive in irradiated Olifantsvlei sludge.

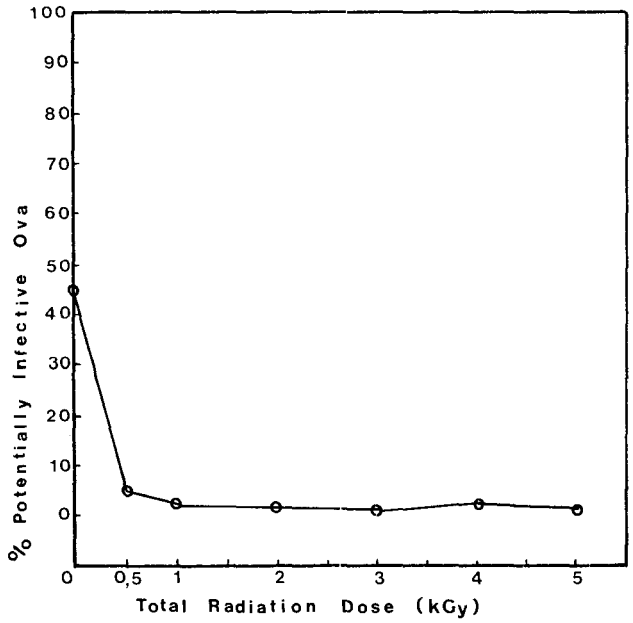


Figure 4
Effect of ^{60}Co *on* Ascaris lumbricoides *ova in digested sludge*

TABLE 1
INFECTION OF WHITE MICE WITH *ASCARIS LUMBRICOIDES* LARVAE WHICH SURVIVED GAMMA RADIATION

Total Radiation Dose kGy	Larvae recovered from Mice Treatment of ova		
	Suspended in distilled water	Seeded into sludge	Embryonated larvae irradiated in sludge
Nil (control)	+	+	+
0,3	+		
0,5	+	+	
0,7	+	+	
1,0	+	+	—
1,3	—	+	
1,5		+	
1,7		+	
1,9		—	
2,0		—	
2,1		—	+
2,3		—	
2,5		—	
3,0		—	+
3,5		—	
4,0		—	+

Nevertheless migration of larvae in the mouse can be taken as a certain indication that infection of the natural human host would occur with larvae surviving low radiation doses, e.g. 0,5 — 1,0 kGy. Additionally, due to the time-consuming nature of these investigations it was only possible to carry out a very limited number of experiments, so that the results presented cannot be taken as conclusive but only indicative of trends.

Table 1 shows that ova suspended in distilled water were inactivated by a total radiation dose of 1,3 kGy whereas those seeded into sludge required doses in excess of 1,9 kGy to inhibit infection. This would indicate that the sludge provides a shielding effect against radiation. When sludge was inoculated with fully embryonated larvae and then irradiated, some of the larvae were resistant to a radiation dose of 4,0 kGy.

TABLE 2(a)
INFECTION OF WHITE MICE WITH *ASCARIS LUMBRICOIDES* LARVAE WHICH SURVIVED GAMMA RADIATION

Date	Source of Ova	Radiation Dose kGy	% Potentially Infective Ova	No fed/mouse	No of larvae recovered
January 1977	Worms from Child A	Nil	81,0	460	19
		0,3	24,0	430	23
		0,5	3,0	530	3
		0,7	2,0	180	2
April 1977	Worms from Child B	Nil	46,0	1 000	74
		0,3	27,0	1 100	42
		0,7	6,0	8 500	46
		1,0	4,0	2 100	8
		1,3	1,0	460	nil

TABLE 2(b)
INFECTION OF WHITE MICE WITH *ASCARIS LUMBRICOIDES* LARVAE WHICH SURVIVED GAMMA RADIATION

Date	Source of Ova	Radiation Dose kGy	% Potentially Infective Ova	No fed/mouse	No of larvae recovered
April 1977	Infected Child B	Nil	45,0	1 000	11
		1,0	2,6	14 000	30
		1,3	0,3	1 700	15
November 1977	Worms from	Nil	41,0	1 000	236
	Sewage Works	1,0	3,0	1 400	19
	screens	1,3	nil	*	nil
		1,5	nil	*	nil
		1,7	nil	*	nil
		1,9	nil	*	nil
		2,1	nil	*	nil
		2,3	nil	*	nil
		2,5	nil	*	nil
December 1977	Worms from Sewage	Nil	86,0	1 200	188
	Works screens	1,0	7,0	1 500	44
		1,3	0,6	120	nil
		1,5	nil	*	nil
		1,7	nil	*	nil
		1,9	nil	*	nil
		2,1	nil	*	nil
		2,3	nil	*	nil
		2,5	nil	*	nil
		3,0	nil	*	nil
		3,5	0,2	34	nil
		4,0	nil	*	nil
June 1978	Worms from Child D	Nil	83,0	840	76
		0,5	29,0	810	21
		0,7	2,0	250	2
		1,0	nil	*	7
		1,3	0,7	90	2
		1,5	nil	*	9
		1,7	nil	*	4

*Numbers could not be estimated becausae there were less than 0,3% potentially infective ova in the 300 ova counted for this sample

TABLE 2(c)
INFECTION OF WHITE MICE WITH *ASCARIS LUMBRICOIDES* LARVAE WHICH SURVIVED GAMMA RADIATION

Date	Source of Ova	Radiation Dose kGy	% Potentially Infective Ova	No fed/mouse	No of larvae recovered
December 1977	Worms from Sewage Works screens	Nil	86,0	1 200	188
		1,0		*	nil
		3,0		*	2
		4,0		*	nil
April 1978	Worms from Child C	Nil	92,0	1 000	501
		1,0		*	nil
		2,0		*	nil
		3,0		*	nil
		4,0		*	nil
June 1978	Worms from Child D	Nil	83,0	840	76
		1,0		*	nil
		2,0		*	37
		3,0		*	4
		4,0		*	3

*Numbers of infective larvae could not be estimated because infective and non-infective larvae all had the same appearance microscopically

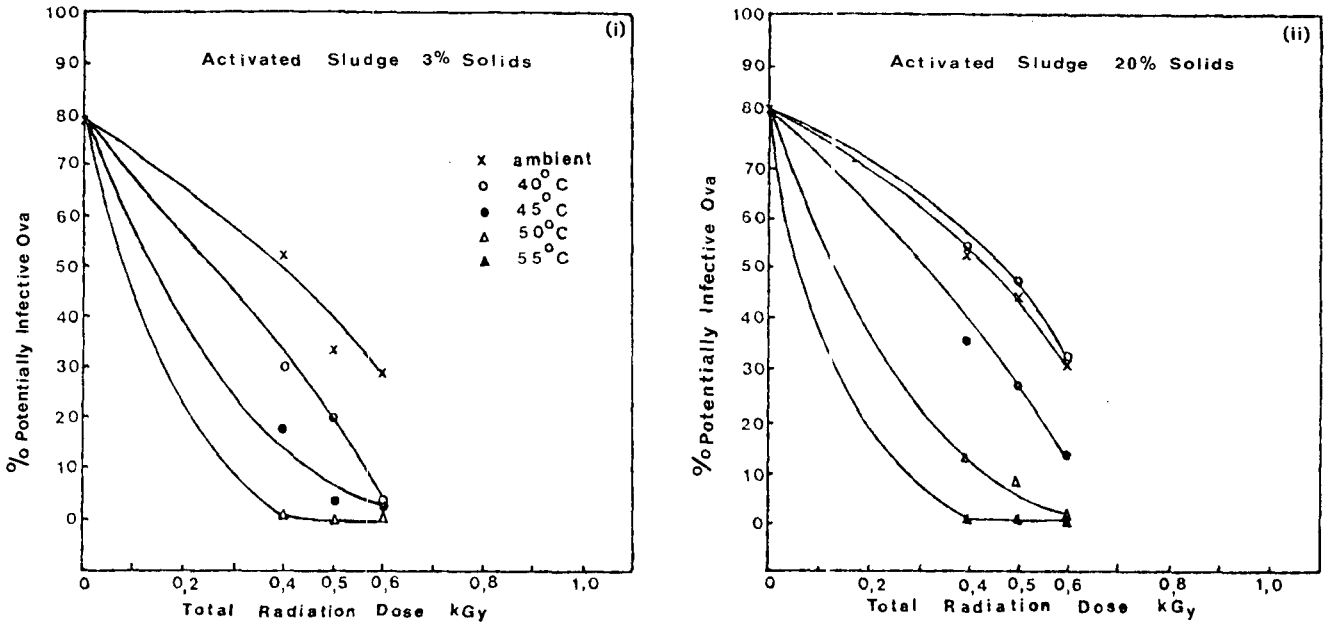


Figure 5
Thermoradiation: Effect of radiation applied at various temperatures on ascaris lumbricoides *ova*

Synergistic Effects

Thermoradiation

Figures 5(i) and (ii) depict the results of thermoradiation.

In a sludge with 3% solids, total inactivation of *Ascaris* ova was achieved by 0,5 kGy radiation at 50 °C. In a sludge with 20 % solids, the combination of 0,4 kGy at 55 °C was required. In both cases synergism is demonstrated because irradiation with 0,5 kGy at ambient temperature left a residual of 34% potentially infective ova and 0,4 kGy left 52 %. The times taken to administer 0,4 kGy and 0,5 kGy doses of radiation at 10 kGy/h are 2,4 and 3,0 min respectively, and Brannen *et al.* (1975) showed that embryonation was unaffected by 10 min exposure to 51 °C and 55 °C total inactivation would require more than 6 min. Brandon (1978) found that in composted sludge of 60 % solids a treatment at 55 °C for 1 h or more was required to sufficiently reduce the number of viable *Ascaris* ova.

Oxygen and Radiation

In Figure 6 it is shown that after a 0,5 kGy dose of radiation 34 % of ova still embryonate whereas when 0,5 kGy of radiation is applied during oxygenation only 3,5 % of ova produced larvae. At 1,5 kGy with or without oxygenation none of the ova were potentially infective. This would indicate a possibility of synergism but a much more detailed study would be required to prove the point.

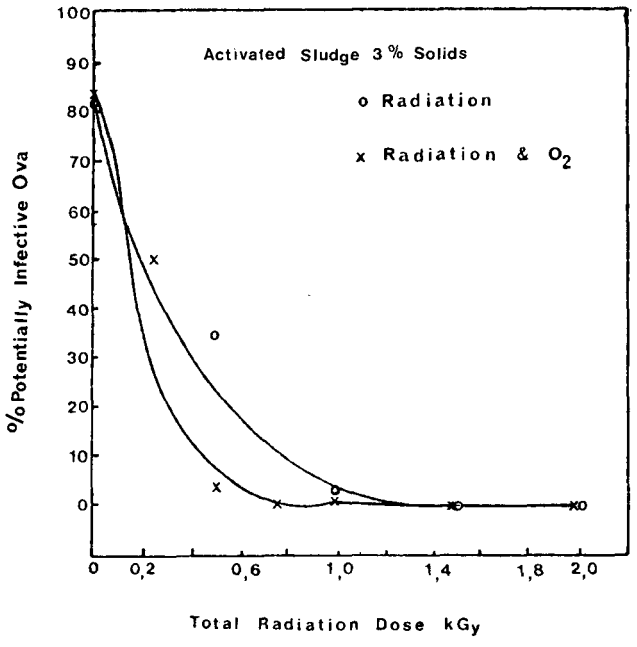


Figure 6
Effect of radiation applied simultaneously with oxygen on Ascaris lumbricoides ova

Conclusions

Gamma radiation at a level of 1,0 kGy effectively reduces the number of potentially infective larvae developing in activated and digested sludge by 99 %. Doses up to 10 kGy do not achieve 100 % inactivation. Larvae which survive radiation can infect white mice so that it must be assumed that they will be infective for the human host. However, it is not known whether the "bio-negative" effect referred to by Varga (1973) would be operative making the worms produced from irradiated ova sterile.

Larvated *Ascaris lumbricoides* ova are more resistant to gamma radiation with ^{60}Co than unsegmented eggs.

A synergistic effect is obtained when 0,5 kGy of radiation is applied to 3 % solids sludge at 50 °C but at higher solids content more intensive treatment would be required.

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