UNIVERSITY OF SOUTHAMPTON

FACULTY OF ENGINEERING AND APPLIED SCIENCE

Department of Civil Engineering

THE DEVELOPMENT OF A PREDICTIVE MODEL FOR THE REMOVAL OF HELMINTH EGGS DURING RAPID SAND FILTRATION

by

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ABSTRACT

FACULTY OF ENGINEERING AND APPLIED SCIENCE

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The objective of the research was to establish whether rapid sand filtration, as a tertiary treatment process, could achieve the WHO health guidelines for the reuse of wastewaters in agriculture. These guidelines reflect the belief that helminth eggs represent the greatest threat to public health.

In order to establish the efficiency of filtration, detection procedures for helminth eggs in sewage were evaluated. The most efficient was used to establish environmental effects on percentage recovery. It was discovered that increasing the time and temperature under which settling occurred increased the efficiency of recovery. Under best conditions, recovery efficiencies were between 50 and 60%.

To investigate filtration, duplicate experiments were carried out on two identical model 125mm diameter rapid sand filters with 77cm depth of sand. 250 <u>Ascaris suum</u> eggs/l of influent were added for 22 hours. The removal efficiency in filters with effective sand sizes of 0.3, 0.4, 0.5, 0.6 and 0.7mm were investigated at a flow rate of $4m^3/m^2/h$. The flow rate was varied during the 0.5mm effective sand size runs to include $2m^3/m^2/h$ and $6m^3/m^2/h$. Experiments with anthracite/sand and sand/garnet mixed medias were also conducted.

Only the sand/garnet mixed media showed total helminth egg removal. The efficiency of the other experiments varied from 99.2% to 99.8% removal, with greater efficiencies for the smaller effective sand sizes and lower flow rates. This was equivalent to between 0.5 and 2 eggs/l of effluent. The WHO guideline value is less than one egg/l of effluent and it was shown that an effective sand size of 0.4mm or under is required to achieve this under the criteria used.

It was established that interception and sedimentation are the determining removal mechanism for helminth eggs. Decreased effective sand size and flow rates improved the efficiency of these processes.

From the results a predictive model was developed which determines the numbers of helminth eggs in the effluent using the initial parameters. It was concluded that rapid sand filtration can obtain the WHO guidelines for reuse provided certain operational criteria are met, and maintained between backwashing.

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GLOSSARY

Symbol Definition

<u>Chapter</u> <u>4</u>

a	Sphere radius	m
°,	Concentration of ion i	mol/l
c	Concentration of suspension	-
C	Inlet concentration of filtration	-
d	Particle diameter	m
D	Grain diameter	Ш
е	Elementary charge (1.60 x 10 ⁻¹⁹)	С
Е	Inertia mechanism (dimensionless group)	-
g	Acceleration due to gravity (9.81)	m/s²
G	Shear gradient	1/s
h	Planks constant (6.63 x 10 ⁻³⁴)	J/s
H	Head loss	m
I	Interception mechanism (dimensionless group)	-
I	Height of expanded bed	m
L	Distance from the inlet face of the medium	m
k	Boltzmann's constant (1.38 x 10 ^{–23})	J∕ ^o K
k ₁	Initial pressure drop through a filter	N/m ²
k ₂	Boucher filterability index	1/m
k s	The initial head loss at surface	m
k _t	Rate constant of surface head loss	1/s
n	Number of ions per unit volume	m ⁻³
Р	Peclet number (diffusion mechanism)	-
Pa	Pressure difference across tube or plug	N/m²
ΔP	Pressure drop across a filter	N/m ²
∆pw	Pressure drop across a fluidised bed	N/m ²
R	Reynolds number (hydrodynamic mechanism)	-
S	Specific surface of filter pores	1/m
t	time of filtration	-
Т	Absolute temperature	о _К

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U	velocity of liquid at infinite distance	
	from sphere	m/s
v	Approach velocity of filtration	m/s
v.	Interstitial velocity of liquid	m/s
vn	velocity of particle relative to liquid	m/s
v s	Stokes settling velocity	m/s
v	Volume of liquid strained per unit face area	m
У	dimensionless potential (=ze\/kT)	-
Уm	minimum of y	
z _i	Valence of ion i	-
–		
α	Exponent of interception group	-
β	Exponent of diffusion group	-
β _a	Bulking factor for deposits	_
Ŷ	Exponent of gravity group	-
δ	Exponent of hydrodynamic group	-
3	Porosity of expanded bed	-
3	initial porosity	-
ĸ	Debye-huckel parameter	1/m
μ	dynamic viscosity of liquid	Kg/m/s
λ	Filter coefficient	-
λ	initial filter coefficient	-
σ	Specific porosity	-
Φ	Overall potential difference between phases	V
х	chi potential	V
Ψ	Surface potential	V
υ	kinematic velocity	m/s
ρ	density of liquid	Kg/m ³
٩ _f	Fluid mass density	Kg/m³
ρ _s	density of particles	Kg/m³
٨	Efficiency of particle retention	-
Ω	angular velocity of rotating particle	rad/s

<u>Others</u>

Symbol	definition	Units
a	ln A (constant for line of pattern of removal)	-
ab	Product of a and b	-
А	Intersection of calculated straight line	-
b	Gradient of calculated straight line	-
с	Arbitrary constant	-
С	Concentration of eggs in influent	eggs/l
e	Exponential (2.718)	-
Е	Effective sand size	mm
Q	Flow rate	$m^3/m^2/h$
k	Arbitrary constant	-
Ν	Number of eggs per centimetre of column at depth X	-
N	Number of eggs in first centimetre of sand	-
p	Arbitrary constant	-
t	Time	hours
u	Uniformity coefficient	mm
Х	Depth of sand	cm

CHAPTER 1:RESEARCH AIM

There is now great importance being placed on the reuse of wastewater in agriculture and aquaculture especially in arid and semi-arid areas. Therefore, problems associated with such practices come to the fore. One of the most important implications in the reuse of wastewaters is the protection of the health of those who may come into contact with These include the producers, the consumers and the wastewater. possibly the local community. Helminth diseases have been established the most important health issue (WHO Tech report 778, 1989) and, as with this in mind, the necessity for tertiary treatments to obtain sufficient removal rates of helminth eggs is required. The tertiary treatments discussed in the above report include chlorination, lagoon treatment and rapid sand filtration. Chlorination is impractcal as it has no effect on helminth eggs and lagoon treatment was suggested as workable but in some cases impractical because of space. Rapid sand filtration is already used in several countries to accomplish further removal of suspended solids after conventional biological or chemical treatment, but little is known about its ability to remove pathogenic Present experience with its use suggests that the micro-organisms. removal of helminth eggs in a well functioning filter may be substantial. However the WHO technical report 778 (1989) states:-

"More research is needed to determine the actual performance of rapid sand filtration in helminth egg removal and to provide design guidelines for its use in the tertiary treatment of wastewater."

This statement is, in effect, the research aim. To enable the aim to be achieved a subsequent literature review revealed several major areas of background theory: the reuse of sewage effluents for irrigation; the health aspects of reuse; and the theory of filtration as a process for removing impurities. These areas will be discussed with emphasis placed on the areas that will aid the eventual creation of design guidelines for the removal of helminth eggs during rapid sand filtration.

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CHAPTER 2: THE REUSE OF WASTEWATERS FOR IRRIGATION

The filtration of wastewater is required to remove pathogens that devalue an otherwise valuable commodity. This value can be measured both in terms of relieving over burdened fresh water supplies and through the addition of nutrients to a crop. The value of enhancing crop production has been recognised in many countries including India, China and more recently in the Middle East. Rapid economic growth during the last decade in the ESCWA (Economic and Social Commission for Western Asia) region has also brought about a growing awareness of the necessity to utilise scarce resources more efficiently. As a result non-conventional sources of water such as sewage effluents are being used.

In Jordan the estimated total resource is around 1100 million cubic meters (MCM)/year (surface water plus ground water extraction). By the year 2000 it is projected that the water demand will be at this level (Musa Nassar 1988). Therefore in some regions the need to develop non-conventional resources is an impending problem. However, the use of wastewaters must be approached with caution. Current concern about environmental quality demands that a responsible and integrated approach be adopted so health risks and costs can be minimised. The health risks associated with the reuse of sewage effluents in agriculture is possibly the greatest concern with this practice and will be discussed later. Other associated environmental hazards include contamination of groundwater supply and accumulation of heavy metals and toxic organics in the surface soil and water. The are salinity, major agricultural concerns reduction in soil permeability and specific ion toxicity (Pescod and Arar 1988).

There is now a good deal of evidence to suggest that groundwater contamination is unlikely to occur in well managed reuse systems (Montgomery 1985, Baxter and Clark 1984, Hartman et al 1980). Indeed Jerome Esmay (1988) has suggested reuse as a treatment involving primary effluent application to crops. There was some evidence to

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suggest that with the system of rapid infiltration the percolation of primary effluents produced a water that was of a higher quality than that of percolated secondary effluents. The organic material present in primary effluent is used as a source of carbon for denitrification, minimising the discharge of nitrate into the underlying groundwater. Secondary treatment removes most organics and the nitrified wastewater undergoes minimal denitrification before infiltrating to an underlying aquifer. Indeed it is now well known that the application of large amounts of inorganic fertiliser can cause large nitrate increases in the underlying groundwater. The East Anglia experience in the United Kingdom is a example where applications of inorganic fertilisers have increased the nitrate concentration in the drinking water of the area to possibly dangerous levels; and certainly above the Government guidelines of water quality (HMSO 1989).

Montgomery (1985) discussed the UK experience concerning groundwater recharge of partially treated sewage. In Winchester (Montgomery, Beard and Baxter 1984) the removal of inorganic nitrogen was mostly more than 50%, and sometimes as much as 90%, by allowing sewage treated by sedimentation only to soak away via a series of trenches and lagoons after being pumped to the top of St. Catherine's Hill. The removal of the biological oxygen demand (BOD) through the unsaturated zone (varying from 4-36 metres in depth) was similar to that of conventional treatment. Overall extensive purification occurs which offers total protection from pollution risk to the water supply intake on the River Itchen three kilometres below Winchester. Dichlorobenzene (DCB) was the principle identifiable organic compound derived from sewage to escape removal in the underlying chalk. DCB seems to have the ability to travel large distances dissolved in water and at Whitchurch it was found in groundwater 400m from the recharge site. Toxic heavy metals tend to be associated with the suspended solids of sewage (El-Nennah and El-Kobbia 1983) and thus would not effect recharge. This implies that the reuse of sludge in agriculture is of more concern to heavy metal contamination than the reuse of effluents.

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The mineral content of the sewage effluent may adversely effect recharge. Calcium, magnesium, sodium, chloride and sulphate all tend to be conserved during artificial recharge. This could restrict the use of recharged effluents for irrigation in arid and semi-arid climates in the long term by increasing salinity to prohibitive levels (Montgomery 1985).

The distance travelled by bacterial indicators of pollution tends to be only a few metres in sand, and tens of metres in crystalline bed rock with a maximum recorded value of 183 metres for faecal streptococci through silty sand and gravel (Frankenberger 1984). It has been observed that the removal of micro-organisms from percolating water is inversely proportional to the soil particle size (Pescod and Arar 1985). Overall Coliform bacteria and viruses are well removed during groundwater recharge (Montgomery 1985).

With soil-aquifer treatment producing high quality effluent for groundwater recharge, it seems that the contamination of groundwater supply through the reuse of sewage effluents in irrigation is certainly controllable and may not be the problem suggested by Pescod and Arar (1985). Recharge is likely to be of more importance in arid areas where there tends to be less groundwater and therefore lower dilution. A well managed programme should prevent any serious environmental problem from the contamination of groundwater.

Salinity can be a major problem when using irrigation waters. When soluble salts in the soil are transported by water their concentrations increase as the water is removed through evaporation and transpiration. Desiccation of surface soil by these two processes creates a gradient which produces an upward flow of salts. This is a process by which many soils become salinised, especially if the water table is close to the soil surface.

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Salinity is reduced by creating a net downward flow of water which transports the soluble salts down through the soil. This is the process of leaching. An equilibrium occurs between the cations in solution and those absorbed on to the exchange complex of the soil. An excessive amount of sodium in the complex of the soil is detrimental to the physical status of the soil, and may even be toxic to plants. In general, effluents will have a higher salinity than normal irrigation water, which implies that applications would need to be carefully monitored.

the application of large volumes of sewage effluents in With irrigation the organic content within effluents may alter the condition of the soil. A figure of 150g/m^3 of organic matter remaining in sewage effluents about to be used in irrigation would not be abnormal (Shuval 1977). Therefore if the seasonal application was 10000m³/ha, 1500 Kg/ha of organic matter would be added to the soil. If the receiving soil is heavy the addition of such quantities of organic matter may clog pores, create a surface crust and decrease the infiltration rate of the water. If the capillaries deeper in the soil become clogged anaerobic conditions may prevail causing a slow rate of This will create a decrease in soil permeability, and decomposition. if this persists ploughing may be necessary to recreate aerobic If, however, the organic matter is added to sandy soils conditions. this will make the soil heavier and improve its quality for agriculture.

The problems of using treated effluents in irrigation have been well discussed (Shuval 1977, Pescod and Arar 1985, Biswas and Arar 1988) and the difficulties may seem daunting. However, sewage effluents have been used indirectly through surface water contamination for many years. This, added to the fact that non-conventional water resources are becoming more important, means wastewater management is required to utilise this valuable resource. Many of the engineering problems can be solved by good management techniques, and many countries are already using or planning to apply the direct reuse of wastewaters.

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West Germany began to use wastewater from the middle of this century because of a shortage of fertilisers. The wastewater was used to improve the condition and productivity of light soils. Initially raw sewage was used, but pre-treatment was introduced in the mid 1960's. The effluents were applied using mechanical sprinklers. Sludge disposal, accumulation of heavy metals and high concentrations of nitrates in groundwater are now causing problems in this system. The Wastewater Utilisation Association is now designing a pre-treatment plant incorporating nitrogen removal to over come the difficulties. It should however, be noted that the experiences in the climate of Northern Germany might not be transferable to arid climatic regions.

In Egypt the acute shortage of water and low organic content of the soils necessitated the use of sewage for crop production. El Gabal El Asfar citrus farms have been using primary effluents from Cairo since Changes in the soil have been observed during the last 50 years 1910. but most of them have been considered favourable. These include increased organic matter, clay content and cation exchange capacity of the soil. Increases in copper, chromium, lead and boron have been noticed but they have not yet reached levels that effect yield. Sewage effluents have also been used at Abun Rawash farms and in In all cases sewage effluent irrigation has produced high Alexandria. yields, especially in olives. Higher yields of wheat, beans and alfalfa have been noted, with a higher accumulation of heavy metals in the leaves of the plant species studied being observed. In general sewage effluent is considered as a good source of irrigation water to increase agricultural production under controlled management.

In Jordan the use of wastewater is considered important for the general economic welfare of the country, as well as a source of irrigation water. According to estimates the use of sewage effluents could increase yields to produce 38% of the 1984 imported amounts of barley and alfalfa. Currently wastewater is only used through the deliberate mixing with surface waters during storage in King Talal Reservoir.

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Reuse also occurs in France, Portugal, most of Asia, Mexico, USA, Saudia Arabia, Syria and many other countries especially in arid or semi-arid regions.

Keeping in mind that the Middle East is importing 50% of its agricultural needs, it is imperative for this region that food production is increased through irrigated agriculture. As water is already scarce the reuse of wastewaters becomes more important. This has the added advantage of minimising the need for the addition of fertilisers. As it has been established that the reuse of wastewater is essential, the associated problems must be recognised and controlled. Perhaps the most important problem with wastewater reuse is the potential of transmitting pathogenic organisms at a much faster rate and to a wider population than previously at risk. Therefore the health risks must be established and solutions found.

2.1 HEALTH IMPLICATIONS OF THE REUSE OF WASTEWATER FOR IRRIGATION

For the pathogens within wastewater to provide an actual risk to public health during reuse several criteria need to be fulfilled. The first is that an infective dose of an excreted pathogen reaches the field, or that the pathogen is able to multiply in the field to the level of an infected dose. Second, the infective dose needs to reach the human host so that the host becomes infected. For actual risk to occur the infection needs to cause disease or the further transmission If this last criteria is not fulfilled then the of the pathogen. addition of wastewater only creates a potential risk. It is possible for a potential risk to occur without resulting in an actual risk. This is due to the effects of other factors including infective dose, human behaviour and human immunity (Pescod 1988). These factors will be discussed during the description of the pathogens associated with wastewater.

The actual public health risk can only be established by a comprehensive epidemiological study of the project or area. Actual risk should only be considered as the excess risk to health over and above the background levels of infection. The control of the excess risk, to prevent it or to minimise it, is the aim of the reuse With this in mind the International Reference Centre for manager. Waste Disposal (IRCWD) presented a generalised model (WHO 1989) for the protection of health during wastewater reuse. The model takes into account "excess risk", and the fact that full treatment of wastewater is not always economically possible or desirable. Other policies therefore need to be considered and these include partial treatment, crop restriction, application measures and human exposure control.

The model (Figure 2.1) consists of five concentric bands representing steps on the pathway from pathogen application on the land to the consumer. The shaded areas denote the extent of the excess risk to an individual within each section. The aim is to prevent any risk in the

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Figure 2.1:Generalised model illustrating the effect of different

KEY TO LEVEL OF CONTAMINATION(outer bands)/RISK(inner bands)



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inner two bands, thus preventing excess risk for the worker in the fields or the consumer. The model is a simplification of reality and, although it should apply to most situations, there will be some cases where it does not accurately represent the level of risk involved.

The top section of the model indicates high risk to all levels of a project when raw wastewater is applied to fields. The other extreme (regime H) shows no contamination or risk when full treatment is Full treatment is described as the treatment required to imposed. obtain the guidelines laid down by the Engelberg report in 1985 (discussed later). This is a bacterial quality of < 1000 faecal coliform per 100 ml and < 1 viable helminth egg per litre. Waste water of this quality can be obtained by several means. Waste stabilisation ponds with a total retention of 25 days are particularly efficient. The other method is to upgrade conventional secondary treatment (mechanical/biological) plants with the addition of tertiary treatment such as filtration.

Regime A in the model considers crop restriction. On its own this method is a good protective measure for the consumer, as either the crops grown are not for human consumption or they need to be cooked first. This includes a wide range of crops such as fodder, pasture, trees or root crops. Depending on the method of irrigation salad vegetables that grow well above ground (e.g. tomatoes or chillies) can also be grown. This protection method will have no effect on the worker as they will still be exposed to the pathogens present in the wastes, on the crops and in the soil.

Regime B shows the possible benefits of application measures. Careful choices can prevent any health risk reaching the crop, and therefore protection is provided for the worker and consumer. Of the three most common irrigation techniques the most effective application method is the use of localised drip or sprinkler irrigation where row crops can be grown. This method uses water efficiently, but requires water of high quality. The substantial treatment would create a water equivalent to fully treated effluent. A lower grade effluent can be

used in bubble irrigation which is a relatively new innovation. When using surface irrigation the method chosen effects the health risks involved. Border irrigation is capable of being safe for the worker in the field after large particulate matter has been removed from the Crop selection is required to insure the consumers safety. sewage. Basin irrigation requires more contact of the worker with the irrigation water, and is therefore less appropriate in reuse. It is. however, a useful method for the irrigation of orchards. Furrow irrigation is the best surface method when considering irrigation with There need be no contact with the worker as valves can wastewaters. efficiently be used, and as the crop is on raised ridges it does not come into contact with the water directly. The crop should then also be safe from contamination.

Human exposure control methods are illustrated in regime C. These methods can effect both the consumer and the worker in trying to prevent contact with the pathogens. For the worker this includes the wearing of protective clothes and increased hygiene, or even immunisation. Consumer measures include increase health education and thorough cooking. This method in practice does not achieve "safe" conditions but should create a reduced level of risk.

Regime D offers the effect of partial treatment which causes a reduction in the level of contamination. The effects vary depending on the treatment. Waste stabilisation ponds with a retention time of 8-10 days reportably remove all helminth eggs (WHO 1987), but some bacterial contamination may remain which could put the consumer at Partial treatment with conventional works can not guarantee risk. safety for the worker or consumer. When combined with crop restriction (regime E_T) partial treatment provides better protection, although when conventional treatment is used (Regime E_{TT}) total protection for the worker cannot be assured. The extra barrier of human exposure control with partial treatment (Regime F) would provide safe conditions for the worker. However it would be difficult to provide sufficient control for the consumer and therefore some risk will remain. Where no treatment is possible a combination of crop

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restriction and human exposure control (Regime G) will prevent the contamination of the consumer and reduce contamination for the worker.

The model suggests that there are three regimes which provide "safe" conditions for both the consumer and the worker. These are localised application of wastes (Regime B), partial treatment with waste stabilisation ponds combined with crop restriction (Regime E_{τ}) and full treatment. Several regimes are available which reduce the risk of contamination and it is suggested that these regimes should be used while the regimes that create "safe" conditions are being developed. The model allows the use of partially treated effluents in irrigated agriculture while using alternative measures to provide safe There are many places in the world already operating conditions. projects that exhibit the model's characteristics. In Peru the use of partially treated effluents allows agriculture in the otherwise unproductive arid coastal regions. Crop restriction also applies as cash crops such as cotton are grown. This provides protection for the consumer and reduces the risks for the worker when compared with the use of raw wastewaters. Other areas operating similar systems include Mexico, Tunisia, Guatemala and India. Those planning future schemes of waste reuse should evaluate health risks using epidemiologists Health protection measures should be targeted at concept of risk. specific exposed groups in the population to allow the safe utilisation of a valuable resource.

The definition of "safe" within the model was established from the findings of the Engelberg report (IRCWD News 23 1985). A group of experts collated the present knowledge and changed the 1973 standards to more realistic guidelines. Guidelines were chosen so not to encourage the breakdown of all treatment if standards could not be met. This often resulted in unregulated illegal irrigation of salad crops with raw wastewater which created a serious public health problem. The group of experts also considered helminth contamination for the first time. This was intentionally innovative as knowledge on the risk of helminth eggs is still scarce. The new guidelines were based on papers by Strauss (1985), and Blum and Feachem (1985).

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Strauss (1985) reviewed the papers assessing the survival of the four groups of pathogens in various waste collection mechanisms, soil and crops. All pathogens will eventually die or lose infectivity after In general the die-off is exponential with a high initial excretion. die-off in the first few hours or days reducing to a few surviving for much longer periods. Species which have the ability of multiplying outside the host in an intermediate host will still demonstrate the same die off potential within each stage of development. There were several factors established as affecting the survival of the pathogens in storage and treatment. Temperature will increase die-off as it increases, while moisture creates friendlier conditions and therefore promotes survival. The absence of essential nutrients may accelerate die-off, as will competition from other micro-organisms. Sunlight, particularly ultra-violet radiation, will accelerate die-off of all pathogens, and pH effects different pathogens in different ways.

In real world situations Strauss suggested that all non-sewerage systems of collection and storage of waste permit the survival of at least some pathogens in concentrations which could cause disease. The work on survival times was extended to that on crops and in the soil. These were compared to the times of vegetable maturation. In soil the results showed that protozoa and most bacteria die before the crop has reached maturity. Salmonella sp however can survive for up to 100 days in moist shaded areas, which is longer than many crops. Ascaris lumbricoides has the longest survival time which is greater than the maturation period of most crops. The more realistic comparison is the survival of the pathogens on the crops. This greatly reduces the survival time of the pathogens. Ascaris lumbricoides is again the longest survivor with up to 60 days. Only spinach and cucumber have a maturation time shorter than that. If contaminated wastewater is used in irrigation then the time between application and harvest should be that the survival time of the pathogens present. greater Strauss concludes by recommending that any contaminated (1985)waste application should be restricted to fallow periods or to the early phases of plant growth. This should restrict any excess transmission of pathogens.

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Blum and Feachem (1985) summarised the established and assumed risks for humans and animals on various crops irrigated with wastewater (Table 1). Of particular note is the fact that persons consuming crops and all agricultural workers have been shown to be at risk from nematodes when human waste is applied to the land as fertiliser. This led the authors of the Engelberg report to suggest that helminth disease created the greatest risk to public health. This is reinforced by the low infective dose and long survival times of this group of pathogens. Of particular importance are the hookworms and Ascaris lumbricoides. The guideline suggested was therefore a strict

Exposure	infectious disease risk from nightsoil or sludge fertilisation of:			
0P	crops for humans	crops for animals	non-consumable crops	
Persons consuming crops	V B*P^ <u>N</u>	_	-	
Persons consuming meat/milk	-	<u>B</u> <u>C</u>		
agricultura /sanitation workers at site of use	l V"В Р <u>N Т</u>	V" B P <u>N</u>	V"B P <u>N</u>	
V= excreted B= excreted P= excreted N= excreted C= excreted T= excreted -= not appli	viruses bacteria protozoa nematodes cestodes trematodes icable	<pre>V = risk supported data V*= risk suggested V^= risk reported ical conformat V"= risk inconsist by epidemiolog</pre>	by epidemiological by other data but no epidemiolg- ion ency support by ical data	

TABLE 2.1:RISK OF HUMAN INFECTION WITH THE MAJOR GROUPS OF PATHOGENS.

(Source; Blum and Feachem 1985)

value of less than or equal to one viable helminth egg per litre of effluent. It has been established that waste stabilisation ponds with a retention time of more than 20 days will achieve this level (Feachem et al 1983). As stated earlier the guideline for bacteria was relaxed, but no guideline was given for protozoa or viruses. This is because they offer little excess risk due to their fast die-off rates. Viruses are also very difficult to detect. Viruses also tend to be localised and there is often high local immunity amongst the resident Therefore, as long as the waste does not travel a great population. distance before reuse, the localised immunity should offer a good protection. Also of relevance is that the treatment required to produce an effluent quality for helminths and bacteria would probably remove the majority of these two groups. Having established the guidelines (Mara and Cairncross 1989, summarised in table 2.2) it was later considered necessary to publish a rationale for these guidelines. Shuval (1988) in IRCWD News issue 24/25 discussed this.

catego	ry reuse conditions	exposed group	intestinal nematodes (per litre)	faecal coliforms (per 100ml)	wastewater treatment required
A	irrigation of crops likely to be eaten raw, sports fields, lawns	workers, consumers, public.	<u>≺</u> 1	<u><</u> 1000	WSP or equivalent treatment
В	irrigation of cereal, indu- strial or fodder crops	workers	<u>≤</u> 1	No standard recommend	WSP, 8-10 days or equivalent
С	Localised irrigation of crops in category B where there is no human contact.	None	Not applicable	Not applicable	treat as irrigation technology requires. At least primary settling

TABLE 2.2: PRESENT RECOMMENDED MICROBIOLOGICAL GUIDELINES FOR WASTEWATER USE IN AGRICULTURE

WSP= Waste Stabilisation Ponds. (adapted from WHO TR778, 1989)

The initial guidelines offered over 50 years ago by the California State Health Department of 2.2 coliforms per 100ml was established when people believed the antiseptic environment was possible. These standards were not based on an analysis of the epidemiological evidence but seemed to be a backlash against poor reuse projects creating odour and fly problems in the state. This was recognised by the group of experts in 1973 (WHO Tech report 517 1973) because they suggested more research was required, and relaxed the value to a level of 100 faecal coliform per 100ml. After several epidemiological studies including Blum and Feachem (1985) a level of 1000 faecal coliform per 100 ml was shown to be both epidemiologically sound and technically feasible. As 50% of the rivers used in unrestricted irrigation in Europe and America have this level of coliform contamination without causing serious health problems this level was assumed safe. The 1985 group did however recognise that in many developing countries the main risks from wastewater irrigation were associated with helminth diseases. Therefore a high degree of helminth removal (about 99.9%) was required to reach the strict guideline value of ≤ 1 viable egg per litre. These recommendations were developed to guide engineers and planners in the choice of treatment technologies and management options (eg crop restriction) so the proposed quality can be achieved. Once achieved there would be no necessity for continuous monitoring.

Although the guidelines in table 2.2 are based on present epidemiological evidence, Cairncross (1990) illustrated that presents methods of research in the field of epidemiology are prone to major inaccuracies. As such he concludes that a reconsideration of present evidence is undertaken so new and improved programme design and evaluation can be increased. Despite this shortfall, Shuval (1988) states that the new guidelines should provide a very high degree of public health protection. At the same time it will enable the development of wastewater recycling projects and result in multiple benefits regarding the promotion of agriculture and water conservation coupled with improved water pollution control.

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In conclusion Strauss and Blumenthal (1990) in an IRCWD executive summary on the reuse of wastewaters in agriculture suggest five global First, authorities should be encouraged to consider recommendations. new or expanding existing wastewater or excreta reuse schemes, but only if health protection measures are an integrated component of the Secondly, waste reuse should be fully integrated into scheme. strategic water resource planning; and thirdly, the careful monitoring of industrial waste streams should be undertaken to prevent crop contamination. Fourthly, all four protection measures discussed from figure 2.1 should be examined and flexibility to individual situations should be applied. The four methods of protection are wastewater treatment, crop restriction, appropriate application methods and human exposure control. To achieve this it is recommended that there is the need for suitable administrative, legislative and political support systems. Finally, the need for field level investigations with an epidemiological perspective is emphasised. Study situations should be so as to allow the effectiveness of individual health chosen protection measures or of combination of measures to be tested in avoiding actual risk from the reuse practice.

With reference to the relevance of each group of pathogenic organisms as described by the guidelines above, the health implications of the species most commonly found in wastewaters will be discussed. As there are no direct guideline values for viruses and protozoa these groups will not be discussed in subsequent sections.

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2.2.MAJOR PATHOGENIC SPECIES ASSOCIATED WITH WASTEWATER

To establish the need for design guidelines concerning the removal of helminth eggs during rapid sand filtration, the pathogenacy of the species involved require to be discussed with some comparison to other pathogencic organisms.

There are in excess of fifty forms of infections transmitted from the excreta of an infected individual to the mouth of another. This faecal-oral transmission route occurs on contaminated fingers, food, utensils, water or any other route which may allow minute quantities of infected excreta to be ingested. Some infections, notably hookworms and schistosomiasis, have the ability to penetrate the skin.

Some of the diseases transmitted in human excreta are among the chief causes of sickness and death in underdeveloped societies. Diarrhoeas, together with malnutrition, respiratory disease and endemic malaria, are among the main causes of death among small children and infants in developing countries. Cholera causes death in all age groups but, again, especially in children when it is endemic. Other diseases such as hookworm and schistosomiasis cause chronic debilitary conditions that impair the quality of life and make the individual more liable to super-imposed acute infections.

As stated earlier, it has been established in the literature (WHO TR 517,1973;Feachem et al,1983; IRCWD News No 24/25,1988) that the main groups of pathogens associated with the reuse of wastewater are bacteria and helminths. These can also be categorised according to Feachem et al (1983) with reference to their pathogenic nature. Category I infections are caused by pathogens which are infective immediately on excretion and have a low median infective dose but cannot multiply in the environment. The transmission of these pathogens occurs predominantly through direct transmission from person to person in the domestic environment.

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The organisms causing category II infections are the excreted bacteria. They are infective immediately on excretion but tend to need a higher infective dose than category I infections. Therefore they must generally be ingested in greater numbers to cause a disease. They can however multiply in the environment in substances like food or milk. This allows for longer survival times in the environment. Transmission routes created by reuse projects can be used by these species.

The diseases in category III are caused by soil transmitted intestinal nematodes which require no intermediate host. They also require a latent period in the environment before they can cause infection. The minimum infective dose is only one organism and they will be readily transmitted in reuse projects.

Category IV infections must first be ingested by an intermediate host before humans can be infected by eating under-cooked meat. These organisms may be encouraged by the irrigation of pasture with contaminated wastewater.

Category V infections are those caused by the water-based helminths that require one or two intermediate hosts. These pathogens can multiply asexually in the intermediate host. Infection is caused by the consumption of under-cooked meat or by penetrating unblemished skin. The reuse of wastewater could become a major transmission route for such organisms.

As Feachem et al (1983) has categorised the pathogens associated with wastewater in such a manner, this classification will be used in subsequent sections.

2.2.1.Excreted pathogenic bacteria

Bacteria fall into category II of Feachem's divisions. The ability of bacteria to persist in the environment poses a potential problem for irrigation projects. well wastewater For example, there are documented cases of cholera epidemics caused by the irrigation of vegetable crops with untreated wastewater (Shuval et al 1985). Indeed cholera and tyhoid are the classic water borne diseases which brought attention to the potential problem created by the improper use of Other harmful excreted bacteria include pathogenic wastewater. Escherichia coli, Leptospira sp, Salmonella sp, Shigella sp, and a recently discovered pathogenic group called the Campylobacter sp.

The transmission route for bacteria tends to be the direct faecal-oral route. Transmission through contaminated food is another possibility. The severity varies from cholera, which will cause fatality in 60% of untreated cases, to other species causing mild diarrhoea. However, as diarrhoea is one of the major killers in the developing world any infection may be important. The survival times tend to be in the region of 8-10 days, and it is temperature dependent with warmer temperatures promoting faster die off (Chang et al 1948). Pathogenic bacteria are shed in large numbers by infected individuals $(10^6-10^9/g)$ Dizon etal 1967) but the infective dose tends to be high which lowers the relevance of pathogenic bacteria as a problem in reuse projects.

It has been known for a long time that many kinds of non-pathogenic bacteria are also excreted by man and this knowledge has led to excreted bacteria being used as certain indicators of faecal pollution. Species such as E.coli, faecal streptococci and bifidobacteria can provide useful monitoring standards of bacterial progress in reuse projects. Despite the many types of excreted bacterial pathogens this group does not cause the major concern to a reuse project manager. This is mainly due to the high infective dose required to cause infection and the general inability of the organisms to proliferate in the environment. Bacteria do however merit great

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caution and need careful monitoring within a reuse project. As such the levels of <u>E.coli</u> in sewage effluents have a guideline safe value of < 1000 faecal coliform (geometric mean number per 100 ml) established by the WHO (1989). <u>E.coli</u> are chosen because they are excreted in large numbers in most regions of the world and they are incapable of proliferation in the natural environment. Pathogenic <u>E.coli</u> have only been identified in the past 30 years, and many individuals are not susceptible to its disease. This makes the commensal forms of <u>E.coli</u> ideal indicators for bacterial contamination of effluent in general.

2.2.2.Excreted pathogenic helminths

As the problem of excreted pathogenic helminths has been shown to be the most serious when considering reuse, this section will go into detail concerning the wastewater associated helminthic diseases. An understanding of helminthic diseases is important in order to study the effectiveness of rapid sand filtration during the removal of their eggs.

The species of helminth which infect humans fall into all but one of the categories described by Feachem et al (1983). The only category without helminth is category II which include species that can multiply outside the host in the environment. As Feachem et al has categorised the pathogens they shall be discussed according to this classification.

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CATEGORY I HELMINTHS

These are pathogens that are infective immediately on excretion, have a low infective dose and are unable to multiply in the environment. The major helminth species within this group are <u>Enterobius</u> vermicularis (pinworm) and Hymenolepis nana (dwarf tapeworm).

<u>E.vermicularis</u> is the most common of the intestinal parasites discussed in this section. It is however of very minor public health importance as the disease it causes, enterobiasis, shows slight symptoms or none at all. Although there are estimated to be 1000 million cases worldwide the minor importance means there is little concern about the eggs during the reuse of wastewaters. Eggs are, however, regularly found in sewage samples (Dunn 1988). The female worm is 8-13mm long and contains about 10,000 eggs. The male is smaller at 2-5mm long. The eggs measure 50-60 μ m by 20-30 μ m. It is known that man is the only reservoir and that susceptibility is general.

The other category I helminth is the cestode <u>Hymenolepis nana</u>. This species is unusual among the cestodes (category IV) as it does not require an intermediate host; the eggs are immediately infective if ingested by the new host. Like <u>E.vermicularis</u>, <u>H.nana</u> is of little public health significance, although it can be common in local areas and is considered serious by clinicians. A light infection will normally have light or minor symptoms. The occurrence is world-wide, especially in warm climates, and is common in areas of overcrowding or institutions. Due to the small size of the eggs (30-47 microns) its inactivation by sewage treatment is lower than other species. Waste stabilisation ponds with a retention time of over 20 days should remove all eggs.

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CATEGORY III HELMINTHS

This category contains some of the most important human helminths. These are <u>Ascaris lumbricoides</u> and the hookworms <u>Ancylostoma duodenale</u> and <u>Necator americanus</u>. These are soil transmitted intestinal nematodes that require no intermediate host. Their eggs require a latent period of development in the environment before they become infective. They cannot however multiply outside the host.

The hookworms are of major importance because of the seriousness of the disease and the number of people thought to be infected. Stoll (1962) estimated that 21% of the world's population carry hookworm. In 1962 this represented 630 million people. More recent estimates put the number at around 800 million. In 1962 the estimated daily blood loss through hookworm infections would be equivalent to the total blood in 1.5 million people (Banwell and Schad 1978). This indicates the enormous burden of human ill health caused by hookworm throughout the world.

The occurrence of the hookworms is now worldwide. <u>N.americanus</u> infects the area south of 20° north and <u>A.duodenale</u> infects the area north of 20° north. The worms spread with human migration but the present maps still lack information on the relative importance of the species or its intensity. These would need to be determined before control intervention can occur. A knowledge of the life cycle is necessary in order to understand the possibility of filtration being a process for breaking the transmission route.

The adult worms live in the small intestine. They are nematodes which are either off-white or rusty in colour depending on whether the blood is visible. <u>A.duodenale</u> is larger than <u>N.americanus</u> with the males measuring between 5 and 10 mm and the females between 10 and 18mm. The females are prolific eggs layers and lay the eggs in the lumen of the intestine. Estimates of fecundity vary but commonly cited figures are 10,000/female/day for <u>N.americanus</u> and twice that for <u>A.duodenale</u>.

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Fecundity is density dependent (Hill 1926) and therefore varies largely. The eggs of the two species are very similar and difficult to distinguish. Those of <u>N.americanus</u> are slightly larger measuring $64-76 \times 36-40$ microns. <u>A.duodenale</u> eggs measure $56-60 \times 36-40$ microns.

The eggs leave the host in the faeces and, if environmental conditions are suitable, will embryonate and hatch within 24 hours. The eggs will not hatch when in sewage but remain infective after coventional treatment. The first stage larva, referred to as a rhabditiform, feeds on the faecal microflora and other organic particles of the soil. The larvae grows and moults to become a larger second stage larva that resembles the first. The third stage larva is obtained after a partial moult and the ensheathed larva is the infective stage, usually referred to as the filariform. If the eggs can be prevented from reaching the soil the transmission created by the reuse of effluents will be eliminated.

The larvae of both species are considered efficient skin penetrators and do not need a skin blemish to obtain entry to the body. Following infection the larvae migrate through the blood circulation system to the lungs where they enter the alveoli and move via the respiratory tract to the pharynx. Here they are swallowed and complete their development in the intestine. If the larvae have gained access to the host through the mouth the complete development occurs in the gut.

Many of the control measures for hookworm apply equally to the control measures of <u>Ascaris</u> <u>sp</u> (discussed later). The major difference is that the hookworm adults tend to live much longer in the gut than <u>Ascaris</u> <u>sp</u> and therefore any control measures will take longer to make a noticeable difference.

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It has been shown (Moore et al 1965) that the single most influential factor in the control of hookworm is the wearing of footwear. In rural Costa Rica females who wore shoes had a 13% prevalence compared to 29% with those who occasionally wore shoes and 39% with those who never wore shoes. The universal wearing of shoes, however, is not practicable for economic as well as customary reasons. Available evidence also indicates that excreta disposal programmes will fail to control hookworm infections on their own. This is due to the behavioural attitudes which will produce focal areas of infection such as young children defaecating in and around the house and agricultural workers defecating in the fields. These programs are valuable however, when used in conjunction with mass chemotherapy, intensive health education and the development of basic health services. There can be no doubt that the improvement in hygiene and excreta disposal greatly reduce the prevalence and intensity of hookworm would infections.

The fate of hookworm eggs during sewage treatment is again similar to Ascaris sp. The differences are that hookworm eggs are less dense (1.055 compared to 1.11) and that they may hatch during treatment. Being less dense implies they will be less likely to be removed as efficiently as Ascaris sp eggs in the sedimentation process. This to be the major process by which eggs are removed in tends conventional sewage treatment. If the eggs hatch the larvae will be associated with the fluid part of the sewage and therefore will not settle (Cram 1943). Sedimentation in conventional works occurs at ambient temperature so the eggs are not killed, just transferred to the solid fraction for disposal by other means. Other techniques vary in efficiency but Cram (1943) states that filtration or land lagooning Effluent chlorination will have will remove any remaining eggs. little effect on hookworm eggs. The most effective way of destroying the eggs in sludge is by heating above 60° C.

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The helminth which singularly infects the highest number of people worldwide is <u>Ascaris lumbricoides</u> (round worm). The prevalence of ascariasis, the disease caused by <u>A.lumbricoides</u>, is commonly quoted at around 25% of the world's population, or approximately 1000 million people. The highest estimate published by Peters (1978) suggested that 32% of a global population of 4000 million were infected. This would give a figure of 1269 million cases. This is obviously a significant proportion of the population. Crompton (1988) has suggested that the prevalence is increasing in more countries than it is decreasing.

About 85% of infections are symptomless, although the presence of a few worms is potentially dangerous. The presence of <u>Ascaris sp</u> is particularly important in undernourished communities. It has been estimated that a child with 26 worms may lose 10% of his or her total daily intake of protein (Feachem et al 1983). Crompton (1985) has stated that the global distribution of protein-energy malnutrition (PEM) coincides indistinguishably with that of ascariasis. Other symptoms include pneumonitis with cough, fever, digestive disorders, and bowel obstruction leading to death in children, and migrations of the adult worms to the liver, gall bladder or appendix.

The adult female, which measures approximately 250mm (the males measure 150mm) produces about 200,000 eggs a day. This implies that 9 x 10^{14} eggs contaminate the earth daily. The eggs are ovoid and measure 45-70 microns by 35-50 μ m, and are extremely well protected from adverse conditions by a thick proteinous wall. Some retain their viability for several years with a maximum recorded 14 years in one case (Crompton 1985). The infective eggs are ingested on hands, food, utensils etc. and swallowed. The larvae hatch in the small intestine, pass through the gut wall and into the liver. From there they enter the blood stream and migrate to the lungs were they reach the alimentary tract and travel to the small intestine. Here they develop and mature and the females produce eggs on mating. An adult can survive for 10-18 months.

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The long survival times of <u>Ascaris sp</u> eggs means that they are common in the environment of endemic areas. The numbers in sewage can be large. Bhaskaran et al (1956) implied concentrations of between 200-2000 eggs per litre in sewage from Calcutta. Even in countries thought to have low incidence of ascariasis loads have been found at 200 eggs per litre (Dunn 1988). Their survival in the soil has been recorded by Kirpichnikov (1963) to be a maximum of seven years. Survival is promoted by cool, moist, shaded soil and by being under the surface rather than on it. Exposure to sunlight and desiccation reduces the survival times considerably. The possibility of long survival times is of great importance when irrigation with wastewaters is considered.

The contamination of crops is an important transmission route in some communities where family hygiene is of a high standard. There are many examples of <u>Ascaris sp</u> contamination of vegetables irrigated with raw sewage infecting the consumer. It is arguable that <u>A.lumbricoides</u> is the most important pathogen concerned with the reuse of wastewater due to the long survival times of the eggs. Therefore their removal by rapid sand filtration before reuse is considered very important.

Conventional treatment is able to remove the majority of eggs mainly through sedimentation. It has been reported that 35-90% are removed by primary sedimentation and 90-99% by a complete trickling filter plant (Cram 1943, Feachem et al 1983). The removal of the eggs does not destroy them, merely concentrates them in the sludge. The most effective way to destroy them is by heating to $60^{\circ}C$ for a few hours.

It is claimed that tertiary treatment in the form of filtration should remove <u>Ascaris sp</u> eggs from sewage effluents (Feachem et al 1983), although there is no evidence given for this. Waste stabilisation ponds with a retention time of at least 20 days will remove the majority of <u>Ascaris sp</u> eggs from the effluent. The eggs settle to the sludge layer where they die after a few months. A comprehensive literature review of <u>Ascaris sp</u> and other pathogens is given by Feachem et al (1983).

The third major helminth in this category is Trichuris trichiura, commonly known as whipworm. Whipworm is similar to Ascaris sp and hookworms because it has a cosmopolitan distribution and produces severe clinical consequences in heavily infected individuals. It is often associated with Ascaris because they are usually endemic in the the life cycles, mode same areas and of transmission and epidemiologies are similar. Trichuriasis occurs throughout the world is very common in warm humid climates. Local prevalences of and 50-99% have been reported, although 25-40% is more common.

The adult female worms measure 25-50mm in length and the males 30-45mm. Females lay between 2,000-10,000 lemon shaped eggs per day, 50-55 µm long and 22 µm wide. Once discharged into the environment they take 2-5 weeks to develop into the infective stage when under moist, warm conditions. The eggs are less resistant than <u>Ascaris sp</u> eggs, but can survive for several months. The eggs are ingested on hands, food or soil and hatch in the small intestine. Development takes about two months and the adult may survive for 3-5 years.

The fourth major parasitic helminth in category III is <u>Strongyloides</u> <u>stercoralis</u>. The infection is very similar to hookworm, but the life cycle varies considerably. Infection is often symptomless, but any infection is potentially dangerous, especially in the malnourished. When the body's immune responses are low the larva may attack most of the body organs, which is usually fatal. The adult worms are only 2-2.5mm long living embedded in the mucosa of the small intestine. The female worm can produce eggs asexually and may lay several dozen measuring 50-60 μ m by 30-35 μ m. The eggs are seldom seen, as they hatch in the gut, and the larvae are passed in the faeces. As such the removal of the eggs by rapid sand filtration will not occur.

Category III helminths contain the majority of the helminth species which are of most concern to public health during the reuse of sewage effluents. This is because of the long survival times of the eggs and the ability of the hookworm larva to penetrate the skin. Both aspects provide difficulties for the control of the disease.

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CATEGORY IV HELMINTHS

For the successful transmission of viable eggs in this category they must first be ingested by a pig or cow. Humans then become infected by eating the undercooked meat of infected animals. Therefore transmission can be aided if contaminated waters are used to irrigated pastures. The category is mainly concerned with the tapeworms of the genus <u>Taenia</u>. Although the infection of Taeniasis, caused by <u>Taenia</u> <u>sp</u>, may be common in some areas of the world the disease is not a major public health problem. It is however a major veterinary problem as cattle and pigs infected with the worm are unfit for human consumption. This produces considerable financial loss and wastage in endemic areas.

The adult worm attaches itself to the wall of the small intestine and <u>T.solium</u> grows to 2-4 meters in length. <u>T.saginata</u> can grow to 15 meters in length but is normally 6-10 meters. Typically the disease is symptomless although Cysticercosis can be serious. This describes the infection caused by the larva of the species. The adult tapeworm passes about 8×10^5 to 1×10^6 eggs a day inside gravid segments. The eggs measure 30-70 µm in diameter.

Once in the environment the survival of the eggs varies from 30-200 days depending on the conditions. Their inactivation by sewage treatment is similar to <u>Ascaris sp.</u> However, the eggs may not be detectable if they are still present in their proglotids.

Although the category is of little public health importance the loss to the farmer and therefore his livelihood may be appreciable. This implies that tapeworms must be monitored and controlled in any reuse project or pasture land should not be irrigated with wastewater in endemic areas.

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CATEGORY V HELMINTHS

Category V infections are all caused by water-based helminths that require one or two intermediate hosts. They include diseases that have the potential of multiplying many times outside of the major Possibly the most significant disease to sanitary engineers is host. included in this group. That is Schistosomiasis, which is one of the few diseases increasing due to irrigation practise. It is caused by from the genus Schistosoma. One species, S.haematobium, worms inhabits the veins around the bladder, whereas the other major species live in the portal venous system that transport blood from the intestine to the liver. These are S.mansoni and S.japonicum. S.intercalatum is important in localised areas of West Africa.

The range of disease produced by infection is great. For many years cause of death has been reported as brain damage, liver or kidney failure when this had resulted from Schistosoma sp infection. The adult worm in the veins give rise to few disorders; it is the eggs which create the problem. These may number hundreds per female per day or, in the case of S.japonicum, thousands. A proportion of the eggs (about 20%) escape the body in the urine or faeces and are responsible for transmission to other humans, and may cause some damage as they pass through the body. The majority of the eggs are retained within the body. They may get stuck in the blood vessels of the liver or the kidneys, the brain, or occasionally the spinal chord. The blockage causd by the eggs induces a chronic inflammatory response around it and the egg gradually becomes calcified. The retained eggs S.haematobium may block the passage of the urine from the kidneys of and the resultant back pressure may destroy the kidney. The eggs that pass in the urine will often damage the tissues, and this produces blood in the urine.

The intestinal Schistosomes cause bleeding into the bowel, and in heavy infections bouts of dysentery with the passage of blood may occur. In the worst cases impacted eggs in the liver may cause back pressure on the blood which results in the creation of a series of

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bypasses that return the blood to the heart without first passing through the liver. The eggs cause the problem due to their size (140µm in length) and the presence of a large lateral spine. This may also be a reason way the eggs may take a few days to pass out of the body. Once the eggs leave the body they are mature and are ready to hatch. The egg hatches when the osmotic pressure of the surrounding environment falls. This causes a problem during rapid sand filtration as the resulting miracidium has the ability to move through a filter (Jones et al 1947) thus creating a very low removal efficiency.

The miracidium have up to six hours to find a suitable snail host. The main species of snail are Balinus sp for S.haematobium, Biomphalaria sp for S.mansoni and Oncomelaria sp or Tricula sp for S.japonicum. On encountering a suitable snail the larva penetrates and develops for one to three months, undergoing massive asexual reproduction. The aquatic larvae called cercariae are then released. These may number thousands for each egg reaching a snail host. The cercariae have 48 hours in the water, swimming to the surface and sinking during rest periods, to find human skin. On encountering the skin the larvae penetrate quickly and migrate to the lungs. Here they develop for some days before they move to the portal venous system of the liver (or the bladder in the case of S.haematabium) to mature and pair. They then migrate to the intestinal and vesicle blood vessels to release the eggs. It has been recorded that adult worms have survived for 30 years in the gut but 3-6 years are more common life spans. The long survival times of the adults make control difficult.

The main aspect of the control of Schistosoma has been concentrated on the control of the intermediate host. Molluscicidig has a long history, starting from copper sulphate, but now niclosamide and tritylmorpholine are the only ones in use. They are relatively non-toxic to man although they may harm fish and destroy some non-target invertebrates. Snail resistance seems minimal despite long periods of application. Dosage does, however, need careful control and is improved where irrigation flows can be managed.

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The irrigation canals also provide an area for control. By reducing the valid habitat for the snails with concrete linings snail populations will decrease. Water speeds can also be used to control snails. Jewsbury (1985) showed that shell height velocities of 33 cms^{-1} were enough to immobilise the aquatic snail, and 65 cms^{-1} would dislodge it. Field observations suggest that waters travelling over 35 cms^{-1} are generally free of snails (Jobin 1964). It has also been shown that the peak infections of miracidia in snails occurs at velocities of $30-40 \text{ cms}^{-1}$. Below this fewer encounters between the cercariae and snail occur, and above this turbulence prevents penetration of the snail.

It is now suggested that control programmes should be integrated to include mass chemotherapy, mollusciciding and improvements in water supply and sanitation to prevent the eggs reaching the water.

Other species within this category are the liver flukes of which only <u>Paragonimus</u> <u>westermani</u> is of any public health significance. It is a disease of the Far East where crabs or crayfish are eaten raw. The infection is of the lungs, and sometimes the brain, and can be serious. Its life cycle is similar to <u>Schistosoma sp</u> in the respect of needing an aquatic snail as an intermediate host to asexually reproduce. The released cercariae need to find a shellfish to penetrate which is then eaten raw by the primary host to cause an infection.

The variety and, in some cases, the severity of helminth diseases show the need for the effective removal of their eggs from wastewaters before application as irrigation water. Therefore the size and shape of the major helminth eggs associated with wastewater warrent further discussion.

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2.2.3. The eggs of the major wastewater associated helminths

Scale drawings of the major helminth eggs are shown in figure 2.2. This is an artists impression of a typical egg. For a more extensive atlas of helminth eggs in various stages of development see Spencer and Monroe (1982).

<u>Clonorchis</u> <u>sinensis</u> is a species of trematode that occurs in the far east of China, Hong Kong, Japan and Vietnam. The eggs are relatively small, measuring 23-35 by $10-20\mu m$. They are characteristically pyriform in shape, with a prominent operculum (an obvious break in the egg wall which will allow the larva to hatch) which identifies the eggs from other species of trematode. It is now established that 30 million people in eastern Asia are infected with this parasite (figures based on Stoll 1947). The eggs can survive for up to three months.

<u>Taenia</u> <u>solium</u> and <u>T.saginata</u> are the tapeworms that can not be distinguished by their eggs. The eggs are spherical measuring $30-50\mu$ m diameter. They have a distinctive dark brown thick wall, and the egg is sometimes surrounded by a hyaline embryonic membrane often referred to as a mucous sheath. When present this aids identification. The survival times of the eggs is in the region of 95-116 days.

<u>Hymenolepis</u> <u>nana</u> is another cestode, and can be spherical or ovoidal in shape, and are slightly larger than <u>Taenia spp</u> but without a thick dark wall. The outer shell consists of hyaline, and this large clear area is the major distinguishing feature. The survival times are similar to those of Taenia spp.

<u>Trichuris</u> <u>trichiura</u> have very distinctive eggs being elongated and obviously bipolar. The size is 30 by 50µm and is easily identified in sewage samples. The eggs have a natural yellow-brown colour and a clear triple shell. More commonly known as whipworm, the species is widely spread throughout the the warm, moist climates of the world. Survival times tend to be slightly shorter than those of Ascaris.

Figure 2.2: Scale drawings of the major waste water associated

helminth eggs (From Ayres 1989)



Clonorchis sinensis



Taenia solium and Taenia saginata



Hymenolepis nana



Trichuris trich:ura







Enterobius vermicularis

Hookworm

Ascaris lumbricoides (ferfilised) (unfer

bricoides (unfertilised)



Diphyllobothrium latum



Schistosoma japonicum



Hymenolepis diminuta



0 10 20 30 40 50 µm

Paragonimus westermani



Schistosoma mansoni



Schistosoma haematobium

<u>Enterobius</u> <u>vermicularis</u> is a very common lightly pathogenic pinworm. It is thought that every child in temperate climates have been infected at one time (Feachem et al 1983). The eggs are opaque and measure 50-60µm by 20-30µm with a distinctive lopsided oval shape. The larva is usually visible in the egg. Although they are regularly present in effluents they are not of a major public health problem because of the low pathogenicy. The eggs can survive for up to eight weeks.

The eggs of the hookworms, <u>Necator sp</u> and <u>Ancylostoma sp</u>, are difficult to differentiate. There is a clear space between the thin outer membrane and the developing embryo which is not stained by iodine. The eggs range from 55-65µm by 40µm in size and are regularly present in the sewage of infected areas. They pose a major public health problem partly because the larva have the ability to penetrate unblemished skin. It is thought that 600-800 million people world-wide are infected (Feachem et al 1983). The eggs will hatch in 9-17 days and the larva can survive for up to 90 days.

Ascaris lumbricoides is the largest of the common species found in It is easy to observe the difference between fertilized and sewage. unfertilized eggs. The fertilized eggs often have an albuminoid external layer and are almost circular with the embryo clearly visible. The size is 45-70µm by 35-50µm. Unfertilised eggs are more elongated and rarely have the albuminoid layer. The wall of Ascaris eggs is extremely resistant to external environmental factors. This enables the eggs to remain viable for long periods of time. The maximum recorded survival time is 14 years (Crompton 1985) which helps make Ascaris a major public health hazard. It is thought that 1000 million people are infected throughout the world (Crompton 1985). The size, frequency and global distribution of Ascaris make it a good indicator organism of potential health problems.

<u>Diphyllobothrium latum</u> is the fish tapeworm which can infect humans if raw fish is eaten. It is particularly common in temperate countries with such diets. Prevalence may be 10-30% in local areas. The eggs measure 55-80µm by 40-60µm, ovoidal, and have a prominent large operculum. They are not a major public health hazard. The eggs will develop in 11-15 days and the larva will die within a week if a suitable host is not found.

<u>Hymenolepis</u> <u>diminuta</u> is the rat tapeworm, and is not frequently diagnosed in humans. The eggs are spherical, $47-50\mu m$ in diameter and have a yellowish brown colour. They are of very little public health importance.

<u>Paragonimus</u> westermani have large eggs $80-110 \ \mu m$ by $50-60 \ \mu m$ with a very obvious operculum at one end. The trematode generally infects man when raw shellfish is eaten. The eggs can survive for long periods although the miracidia normally hatch in three weeks and survive 24 hours.

The eggs of <u>Schistosoma</u> <u>sp</u> are very large and distinctive. The species of <u>S.mansoni</u> and <u>S.haematabium</u> may be up to 140 μ m long and have a characteristic lateral spine. This makes it difficult for the eggs to pass out of the infective host. The proportion of eggs leaving the host may be as little as 20%. The eggs hatch when they reach water due to the change in osmotic pressure. If the eggs are removed during sewage treatment it is possible that the miracidia will hatch and swim through the treatment processes. This, and the ability of the cerariae to penetrate unblemished skin, make <u>Schistosoma sp</u> a major public health problem when considering the reuse of wastewater in irrigation.

Of the species discussed here <u>Ascaris</u> <u>sp</u> fulfils the role as the best indicator organism. This is helped by the fact that the eggs of the pig roundworm, <u>Ascaris</u> <u>suum</u>, are virtually identical to those of the human infection but are not pathogenic to man. Therefore the eggs of A.suum were used in the laboratory tests.

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CHAPTER 3: DETECTION OF HELMINTH EGGS IN SEWAGE

3.1. INTRODUCTION AND METHODS

Initially work on the detection of helminth eggs in sewage was undertaken to establish the possibility of using such techniques for sampling the filter during the experimental runs. The reliability of the available detection procedures therefore needed to be assessed. The necessity of reliable detection has been confirmed by recent publications for guidelines of helminth egg numbers in effluents to be used in irrigation (WHO TR778 1989). Professor Schwartzbrod of the University of Nancy, France has evaluated twenty techniques available within the literature. Of these, two were described as efficient. These two techniques have been evaluated by the author in Britain and and their efficiencies were recorded as well as their Jordan susceptibility to environmental conditions. One technique is based on sedimentation whilst the other relies on flotation. The procedures for these techniques are shown in figures 3.1 and 3.2.

The flotation technique relies upon a few fundamental assumptions. The first assumption is that all the eggs in the litre of sample reach the sediment within the allowed overnight settling time. The size and density of the eggs assures that this is a safe assumption. The next assumption is that the addition of sodium nitrate has the ability to penetrate the initial centrifuged pellet in step iv of the process and allow the flotation of the eggs as distinct entities to the organic matter in which they are contained. There was a problem in obtaining a clean supernatant at step v. In carrying out this procedure the most difficult part was the removal of the supernatant at any stage without disturbing the sediment. Otherwise the technique is easy and repeatable.

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Figure 3.1: Technique 1 (Flotation, WHO 1989)

Procedure

i) A grab sample of sewage is taken back to the laboratory and one litre is placed in a beaker and allowed to settle overnight. In the U.K. the samples were seeded with known numbers of <u>Ascaris suum</u> before settling. The numbers of eggs added were 2000, 3000 and 4000 eggs per litre.

ii) The supernatant is removed without disturbing the sediment and discarded. About 100ml of sludge remains.

iii) The sludge is centrifuged in four tubes at 1000rpm for 10 minutes and the supernatant discarded.

iv) 3ml of saturated sodium nitrate was added to each tube. This was centrifuged at 2500rpm for three minutes. The supernatant was decanted into a conical flask containing one litre of clean water. This was repeated three times.

v) The conical flask was left to settle for a two hours before the supernatant was discarded leaving the bottom undisturbed. The remaining liquid was centrifuged at 2500rpm for six minutes.

vi) The bottom 1ml of sample was placed in a MacMasters microscope slide and enumerated. The number of eggs counted is equal to the number of eggs present in the initial sample of one litre.

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Figure 3.2:Technique 2 (Sedimentation, Bailenger 1979)

Procedure

i) A grab sample of sewage was taken back to the laboratory. A one litre sample was placed in a beaker and left to settle overnight.

ii) The supernatant was removed and discarded without disturbing the lower 100-200ml of sludge.

iii) The sludge was placed in four centrifuge tubes and centrifuged at 2500rpm for 15 minutes. The supernatant was discarded.

iv)An equal volume to the pellet of aceto/acetic buffer* was added and mixed.

v) Ether, to a volume twice that of the buffer, was added and the mixture was stirred for 10 minutes.

vi)The organic layer created by this step is removed and discarded.

vii) Zinc sulphate was added to a volume of roughly five times that of the remaining pellet. After the mixture had been well stirred 1ml was taken for enumeration under the microscope in a MacMasters slide. The number of eggs in the sample can then be calculated by:-

$$N = n/0.15 \times V$$

where N = the calculated number of eggs in the sample n = the number of eggs in one chamber of a MacMasters slide 0.15 = the volume in one chamber of a MacMasters slide and V = the total volume of the sample.

*Aceto/acetic buffer: Crystaline sodium acetate 5g Glacial acetic acid 3.6g Distilled water to make up to 1000ml

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The application of the sedimentation procedure also had areas where variability would occur. The amount of zinc sulphate varied in proportion to the amount of organic matter remaining after step v. The technique described by Professor Schwartzbrod suggests 5ml. This was invariably not enough to allow reasonable counting of the eggs to take place unhindered by organic matter. In general 10ml was required. If this quantity becomes too high the potential error of the technique increases. The observation and identification of the eggs is also a problem with this technique. Even after the separation of the organic matter the amount remaining is enough to hinder the counting of eggs by masking the eggs. This problem was particularly acute in Jordan where unknown species of egg were being identified.

Both techniques try to concentrate the numbers of eggs in a large sample and then remove the organic matter so counting is possible. This is achieved to varying success as will be described later.

3.2.RESULTS

The seeding of samples with known numbers of eggs was based on the reliability of counting shown in table 3.1. A known number of eggs (10,000/ml) was counted ten times and then diluted to the theorectical concentration of eggs indicated in the first column of the table. The mean count equates to the number of eggs actually observed.

dilution	mean of counts	standard
(eggs/ml) ===============	(n=10)	deviation
10000	8672	1324
5000	4793	1090
2000	2320	421
1000	861	103
500	582	107
100	96	37

Table 3.1:Reliability of seeding counts

Each dilution was counted ten times for a mean to be obtained. Although the standard deviation is reasonably large there is no overlap between samples. The means were considered reliable enough for the efficiency of the techniques to be assessed. The percentage efficiencies of the results produced i this section are based on the values shown in table 3.1. These values are similar to those established in a similar analysis by Da Lage (1990).

Table 3.2 shows the results of the work undertaken in the U.K. using technique one. The results are the number of <u>Ascaris suum</u> eggs/l found in the final count. They are also expressed as a percentage of the known number of eggs added (4000 eggs/l) based on table 3.1. As can be seen the results give very poor percentage recovery rates. As such it was decided that technique 1 should be ignored in preference to technique 2. The results obtained in the laboratory and in Jordan were therefore obtained using the sedimentation technique.

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Table 3.2: Recovery rate of technique 1

Sample	Raw		final	
	sewage		effluen	t
	No./l	%	No./l	%
А	980	25	1680	42
В	9 20	23	1320	33
С	33	2	88	4
D	840	21	1560	3 9

Table 3.3 shows the calculated percentage recovery using technique 2 of <u>Ascaris suum</u> eggs from samples of seeded raw sewage. The oven samples were kept at 35° C in an incubator. This was warmer than the overnight ambient bench temperature. As can be seen both time and temperature have a profound effect on the recovery of <u>Ascaris suum</u> eggs, although the large variability of the results should be noted.

Table 3.3:Recovery rate of technique 2 under the effects of time and temperature

Nights	Temp.	% recovery	Average
in lab		(six samples)	recovery
ONE	вепсп	44,13,48,52,30,33	3/./
	0ven	48,26,53,49,47,51	45.7
	Bench	29,24,54,54,51,42	42.3
T₩O	0ven	49,29,67,56,58,65	54.0
	Bench	58,28,62,54,54,49	50.8
THREE	0ven	70,49,68,73,91,96	74.5

Table 3.4 shows the results gained using technique 2 from raw sewage in Jordan during the summer of 1988. The cold samples were those kept in the refrigerator overnight at $6^{\circ}C$ and the warm samples were left on the bench overnight. The coldest night temperatures were $18^{\circ}C$ and the warmest day temperatures were the mid to high thirties in degress Centigrade.

Of interest in these data is the large total number of eggs present, deriving from several species. <u>A.lumbricoides</u> is the most common species in all cases with the two species of hookworm regularly present in high numbers. These numbers of eggs are present despite the providing community having a relatively high level of public health. As far as the technique was able to detect, keeping the samples in warm conditions seems to dramatically improve the recovery rate. This compares well with the work undertaken in the U.K.

The raw data for the results in table 3.4 are given in Appendix A. The two areas of note from this information are the lack of data coming from the oxidation ditch samples, and the lack of pathogenic eggs in the effluent of the sedimentation tanks. The lack of data from the oxidation ditch was due to the large amounts of organic matter present in such samples which the technique was unable to remove efficiently. The single egg found in the sedimentation tank effluent was <u>Schistosoma</u> <u>bovis</u> which causes an animal form of schistosomasis. The eggs of this species are very large and its presence suggests that helminth eggs pathogenic to humans may also be able to pass sedimentation tanks. No helminth eggs pathogenic to humans were found in the effluent of the sedimentation tanks.

The raw data also gives a good indication of the variation between samples. If a large general sample of five litres is removed from the sewage works and mixed well before it is divided into the one litre samples, the distribution of the eggs in the one litre samples should be approximately the same. At times, notably the warm samples taken on 31/7, the variation is very large. One of the reasons suggested for this is the difficulty in counting eggs in the samples. The

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presence of large amounts of organic matter creates a problem of visibility in the MacMasters slide. The organic matter can mask eggs or hinder their flotation during the final stage of the technique, and would therefore lead to an under-estimation of the number of eggs present. This problem is magnified by the variation in the shape and size of eggs within species. Results obtained under such conditions suggest that the motivation and enthusiasm of the researcher will affect the results that are obtained.

Species	Cold samples Average(n=10)	.max	.min	Warm sample Average(n=1	es 0).max	.min
Ascaris lumbricoides live	5 75.9	121	22	111.1	198	33
Ascaris lumbricoides dead	12.1	22	0	13.2	55	0
Ancylostoma duodenale	47.3	88	11	48.4	77	22
Necator americanus	51.7	66	22	55	99	33
Trichuris trichiura	7.8	22	0	17.6	44	0
Enterobius vermicularis	33	55	0	31.9	55	11
Paragonimus	11	22	0	18.7	44	0
Unidentified	41.8	55	22	46.2	66	11
SUB TOTAL	281.6	363	242	342.1	462	275
Hymenolepis	2.2	22	0	1.1	 11	0
Diphyllobo-	1. 1	11	0	0	0	0
thrium latum Schistosoma	7.7	33	0	3.3	11	0
mansoni Schistosoma haematobium	2.2	11	0	0	0	0
TOTAL	294.8	363	242	346.5	462	286

Table 3.4: Average number of eggs recovered in Jordanian raw sewage (Jerash)

3.3.DISCUSSION

There are several conclusions that can be drawn from the results above. The first, and one that has already been stated, is that technique 1 has very poor recovery rates. The recovery rates were of such a low magnitude that it was not considered worthwhile continuing with the technique. The major reason for the poor showing of this technique probably lies in stage iv. On the addition of sodium nitrate the eggs need to be in a free state so centrifuging is able to liberate them from the surrounding organic matter. As will be discussed later this assumption seems unlikely and the eggs will not be liberated. Eggs are therefore missed during counting. Another possible reason for the lower recovery efficiency compared with technique 2 is the greater amount of decanting that occurs. For each stage of the technique and for each item of glassware used there is a proportional loss. Therefore technique 2 with fewer stages and less glassware is likely to have a greater percentage recovery.

Concentrating on technique 2 has led to findings of interest. First is the low recovery rates that are still occurring within this The WHO guidelines suggest that less than one viable egg technique. per litre should be present if reuse of sewage effluents are to be considered safe. With efficiencies in the region of 50% the analyst can not be completely confident that no eggs are present in samples where no eggs are detected. The results also show that there is an increase in efficiency with an increase in temperature and an increase The results in the U.K. show a 99% probability of there time. in being a significant difference between bench and oven samples (t=5) and between one night settling and three nights settling (t=6). The Jordanian samples show a 95% probability of there being a significant difference between warm and cold samples (t=2.69). It seems therefore safe to conclude that an increase in temperature will result in an increase in egg recovery. This could also be said of the length of time allowed for the settling of the sample. However, what mechanisms allow time and temperature to affect the percentage recovery?

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3.3.1.Sources of error

There were two sources of error evaluated. The first was the initial sedimentation of the organic matter and the second was the pellet removal. The error in sedimentation applies to both techniques while the error of pellet removal only applies to technique two.

1) Sedimentation

The possible error in sedimentation was evaluated in two ways, centrifuging the supernatant and random sampling of the supernatant.

a) Centrifuging

50ml of the supernatant was removed and centrifuged at 2500rpm for 10 minutes. The lowest 1ml of the resultant sample was enumerated for eggs under the microscope. An average of five samples gives six eggs per 50ml of supernatant. This implies a loss of 2.5% when the supernatant is discarded.

b)Random sampling

From anywhere within the supernatant a random 1ml sample was taken and enumerated. Ten samples were counted and an average of 1.2 eggs/ml was found to be present. Assuming that about 900ml of the sample is discarded as supernatant this would account for a loss of 26%.

Why are eggs found in the supernatant? Eggs of Ascaris suum have a density of 1.1 and an approximate diameter of 60 microns. Therefore the eggs would settle at a rate of 1.74mm/s according to Stokes Law. This rate is high enough to ensure that all the eggs reach the sediment in the time allowed for settling. It was assumed, therefore, that the eggs were being hindered in their settling. This was found to be true. Plates 3.1a and 3.1b show eggs closely associated with organic matter that has remained in the supernatant after settling. It was observed that organic matter remained in the supernatant under

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Plate 3.1a:Ascaris suum egg is within the circle.

Plate 3.1b:Ascaris suum egg is within circle





two conditions. The first is that some organic matter will have a slower sedimentation rate due to the resistance of the water and may not reach the sediment in the time allowed. The second and less common is that part of the organic matter remains attached due to the surface tension of the water and is therefore lost when the supernatant is discarded. The samples left at ambient temperature will also have impeded sedimentation rates due to convection creating water currents during changes of temperature. Any loss of organic matter, with eggs so closely associated, will result in a reduction of the efficiency of the technique.

With any increase in temperature and settling time more sediment is given the chance to reach the bottom of the beaker and the eggs associated will be counted. This is why time and temperature increase the efficiency of the technique.

ii) Pellet removal

In technique two, when the pellet of organic matter is removed in stage vi, the likelihood of the pellet retaining eggs was evaluated. Due to the amount of organic matter, counting was a problem, but eggs were found in this section of the sample. Six samples were taken and the average loss was calculated at 2%. Due to the counting difficulties this will probably be a gross under-estimation and the only conclusion to be drawn is that some eggs are lost at this stage.

The reason for the loss is again probably due to the close association of the eggs with the organic matter. This shows one of the fundamental problems with the technique, which is the difficulty of separating the organic matter from the eggs to allow the eggs to be enumerated.

There are other errors where egg loss will occur, such as with the removal of the supernatant, but these would seem to be minor to those evaluated above and can be minimised by good experimental technique. With 30% of the loss in the one night samples accounted for it is

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reasonable to see why the error is so large. It is also reasonable to assume that the results obtained in Jordan will be an under-estimation of the number of eggs present in the raw sewage. With numbers found being in the region of 300 eggs/l this fact must be of some concern. Similar experimentation was carried out at Southampton University by Da Lage (1990). The recovery rates from the techniques described here varied from 27% to 60% for samples seeded with 100 to 2000 A.suum eggs per litre. Technique 1 gave no results when 10 eggs per litre were added and technique 2 only gave 5% recovery for the same samples. In general Da Lage discovered that the more eggs present in the litre sample the greater the recovery rate. It was interesting to note that the reference technique using no reagents other than water gave significantly higher recovery rates than the other investigated techniques. No explanation was offered for this.

Da Lage concluded that 20 one litre samples free of eggs would be required to obtain 95% confidence of less than one egg per litre being present in effluents. Because of this, she states that none of the present techniques are satisfactory. Ayres (1989) went further and established the number of one litre samples required to be 80% confident that the sample mean (x) is under the underlying mean. She established that an increased organic load required an increase in the number of samples required. These results are shown in table 3.5.

The time required to analyse 32 to 50 samples is often beyond the means of a laboratory unless sampling is spaced out over several weeks. Ayres (1989) suggests that sampling may be spread over several months providing the samples are taken at the same time each day. This would mean that the total suspended solids should not vary too greatly and the mean suspended solids concentration over the period may be used for the calculations. If it is not possible to measure suspended solids then the worst case should be assumed and 50 samples would be required.

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total	probability of	probability of	Number of 1 litre
suspended	recovering 1 egg	detecting ≥ 1 egg	samples to be 80%
solids	per litre (p)	in four 1 litre	confident that
(mg/l)		samples if under-	<pre>sample x = under-</pre>
		ling mean x=1	lying mean (x)
***********		*********************	
0–30	0.8	0.96	32
0–30 30–60	0.8 0.7	0.96 0.94	32 36
0–30 30–60 70–115	0.8 0.7 0.6	0.96 0.94 0.91	32 36 42
0-30 30-60 70-115 >115	0.8 0.7 0.6 0.5	0.96 0.94 0.91 0.87	32 36 42 50

Table 3.5: Probability of detecting less than one egg per

litre in a sample

Taken from Ayres (1989).

3.4.2.Application of the techniques as routine procedures

The reason for evaluating the techniques was to assess their suitability as routine procedures to monitor effluents in reuse projects. Technique 1 with its low efficiencies would seem to be inappropriate although it is an easily repeatable technique. Technique 2 gives higher recovery efficiencies but doubt exists as to whether it is reliable enough for a routine procedure.

Despite the errors that may occur this technique can be standardised by stating the environmental conditions the sample needs to be kept under. Errors can be minimised by careful experimental procedure. The major difficulty with the technique is the counting of eggs in the presence of large quantities of organic matter. An example of a typical slide requiring counting is shown in plate 3.2. This not only masks some eggs so they are not counted but also makes identification difficult. Eggs rarely look like the "text book" egg and the presence of the organic matter complicates identification.

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Plate 3.2:Standard slide for counting helminth eggs in a <u>MacMasters slide after sediment removal by</u> <u>technique two(x40 magnification)</u>.

•



Although this is the best technique currently available for the quantification of helminth eggs in sewage, the results obtained should be treated with caution. A technique prone to the errors described and with the difficulties of enumeration must create questions over its reliability. The final conclusion this researcher can make is that the results obtained will be an indication rather than an exact quantification of the numbers of helminth eggs in a sewage sample. As such, any management policy should take this into account during a reuse project.
3.4.CONCLUSIONS

i)The technique, known as Bailenger's (technique 2, Bailenger 1979), is capable of detecting a wide range of helminth eggs in raw sewage.

ii)The technique known as Teichmann (Technique 1, WHO 1989) gives inadequate recovery for reliable detection.

iii)Factors improving the efficiency of sedimentation (increased time and temperature) will improve the overall efficiency of the technique.

iv)Even under the best conditions, the maximum recovery rate is only 60%. Under normal conditions (those used for the Jordanian results) 50% recovery was the best that could be expected.

v)The technique does not seem precise enough for reliable detection. The results will be an estimation rather than an exact quantification of the numbers of eggs in a sample. This will have a detrimental effect on the accuracy of any predictive model. It seems that the best estimation will be achieved from one litre samples left overnight at 37°C and the results doubled. Therefore if a standard of ten eggs per litre is required, this would be equivalent to a measured five eggs per litre. This would give a reference point for any model.

vi)Further work should be undertaken to determine how time and temperature could improve the efficiency of the technique.

vii)The technique does not allow for easy use for the sampling of a rapid sand filter. The procedure was, however, used as a basic starting point for the development of the methodologies discussed in Chapter 5.

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CHAPTER 4: THE THEORY OF FILTRATION

As filtration is the process of treatment considered within this thesis a knowledge of the mechanisms behind the removal of particles by sand grains is required. At this point it is appropriate to remark that there is not a comprehensive model of filtration that adequately resembles real life occurrences within a filter bed. To date all attempts have failed to develop an exact mathematical formulation for the filtration process to hold for any and all operational conditions. Current theories are either too simple to be widely applicable or too complex to be useful.

The mechanisms involved in filtration have, however, been extensively researched. It is generally agreed that the removal of suspended particles in a filter consist of at least two distinct steps. The first is the transport of particles to the solid liquid interface of the grain in the filter media or to another floc particle already retained by the bed. The particle then has to attach or be absorbed onto the surface of the grain. A third distinct step is that of detachment which may occur during filtration but is principally a process of cleaning. In the case of rapid sand filters the cleansing is usually achieved by backwashing the filter bed with a reverse flow of water and/or air. As the mechanisms fall into distinct areas each will be discussed separately.

4.1.TRANSPORT MECHANISMS

Transport mechanisms are the means by which the particles in suspension come into contact with the grains of the media. The mechanisms fall into groups, of which one or more may effect each suspended particle as it flows through a filter.

4.1.1.Straining and Interception

Straining takes place when a suspended particle is larger than the pore size of the media. Early descriptions of this filtration described it as 'complete blocking filtration'. No physical basis for this was suggested but a mathematically identical relationship was discovered for the straining of a suspension through a woven stainless steel mesh (pore size approx. 35μ m). In water treatment technology this was known as Boucher's Law which is:-

$$\Delta P = k_1 \exp(k_2 V) \tag{1}$$

where ΔP is the pressure drop. k_1 is the initial pressure drop through a strainer, k_2 is the Boucher filterability index and V is the volume strained per unit face area. Boucher's Law, however has not been analysed.

For this type of surface filtration to occur two mechanisms need to develop simultaneously. The first is the deposition of the suspended particles directly onto a pore thus blocking the pore and creating fewer channels. The second is the deposition of the particles directly on to the sand grain, which, if this happens several times around pores on the initial layer fewer, larger pores will be formed above the pores created by the sand grains. Consequently the surface filtration forms a discontinuous cake with holes in it corresponding with the pore sizes directly below. Equation (1), which was written for strainers, has however been found to describe the phenomenon of surface deposition on deep filters. This is an undesirable effect as it creates an exponential decrease in the pressure drop through the filter. Unfortunately the equation and the subsequent mathematical development (described by Ives 1985) are unable to predict whether surface deposition is likely, and what fraction of the inflow will be removed by it.

Interception can be considered as the final mechanism before contact in all cases. Transport mechanisms imply that, in the laminar flow found in filtration, the particles are conditions uniformly distributed through the liquid. The particles follow the streamlines of the flow and if the particles are in streamlines near to the pore wall, and the radii of the particle is large enough, the particle will touch the wall. This has been regarded as a filtration mechanism for characterised by the 30 years. Recently it has been over dimensionless parameter I given by (Ives 1975):-

$$I = \frac{d}{D}$$
(2)

where d and D are the particle and grain diameters respectively. The greater the value of I the more efficient the particle capture. As I approaches 1.0 from below straining becomes the dominant mechanism. In filtration of water and wastewater I lies between 2×10^{-4} and 1×10^{-1} , greater values leading to a straining effect. With reference to the helminth eggs discussed in section 2.3.5 this would aid the removal of the larger eggs and therefore <u>Ascaris suum</u> eggs will have a greater removal efficiency than the other major species.

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4.1.2.Inertia

The streamlines approaching a pore have to converge to be able to pass through the pore. If a particle in a streamline has sufficient density, its trajectory will be maintained so the sand grain will intercept it. Using the equations of motion of a liquid and a particle for flow past an isolated spherical grain, a dimensionless group can give an efficiency of collection of particles on the grain. This efficiency is the ratio of the number of particles sticking to the grain to the number of particles approaching the grain at an infinite distance up stream. This gives equation (3):-

$$E = \rho_{s} d^{2} U$$
(3)
18µD

where $\rho_{\rm S}$ is the density of the particle, U is the velocity of the fluid at infinite distance relative to the spherical grain and μ is the dynamic viscosity of the fluid.

It can be seen that E is independent of the density of the fluid and is inversely proportional to the grain size, D. The most significant parameters are the velocity U, and the viscosity μ . In water filtration, where U is approximately 2 mm/s and μ is 10^{-3} Kg/ms, the value of E will be between 2 x 10^{-9} and 1.5 x 10^{-5} . This gives a negligible collection efficiency. In air filtration where velocities are much higher and viscosity lower the values may be 10^3 or 10^4 times higher. It is however of little importance in water filtration. Therefore this process is unlikely to have any effect on the removal of helminth eggs.

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The idea that pores within slow sand filters act as miniature settling basins, and within them particles were effected by gravity, is a well established theory (Mints 1966). However, due to the speed of flow in rapid gravity filters this was thought unlikely to occur in these filter beds. This is substantiated by Stokes Law which implies the settling velocities of the particles within the pore spaces would be negligible compared with the filtration velocities. In a typical granular filter when the interstitial velocity v_i is 5 mm/s and the settling velocity of an average clay particle v_s is 0.1 mm/s the ratio of settling velocity to mean interstitial velocity is:-

$$v_{s} = 0.1 = 0.02 \text{ or } 2\%$$

 $v_{i} = 5$

This indicates a low gravity effect even for a dense particle like clay (density 2500 kg/m³). Many flocs have densities as low as 1005 kg/m³.

This theory was questioned after analysis of viscous flow equations round a sphere. These indicated that the tangential velocity rapidly diminishes to zero at the surface. The velocity parallel to the surface of a 0.5 mm diameter sphere at a distance of 20μ m from the sphere is 0.036U where U is the velocity of the liquid relative to the sphere. Therefore, if $U = v_i$, then for $v_i = 5$ mm/s the velocity parallel to the surface of the grain is 0.036 x 5 or 0.18mm/s. Using the same clay particle as before this makes the ratio of settling velocity to mean interstitial velocity as:-

> v_s = 0.1 = 0.55 or 55% v(tangential) 0.18

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Under these conditions sedimentation will have a greater effect on removal than the situation described above. Theoretical and experimental investigations by Ison (1969) have shown that particles are deposited on the upward surfaces of grains during downward flow and to a lesser extent in upward flow. This would suggest that gravity does play a part in the removal of particles in a rapid gravity filter. This process will aid the removal of the larger, more dense, eggs with higher settling velocity. Again his would promote the removal of Ascaris over the other common species of helminth egg.

4.1.4.Diffusion

Brownian motion, the random movement of particles in liquid due to the thermal energy of the molecules, has been known to assist filtration for some time.

Using the Stokes-Einstien diffusion coefficient the velocities imparted by Brownian motion over a distance of one grain diameter can be expressed as a ratio with the velocity of the particle due to liquid movement (advective velocity, which is proportional to v). This ratio is the reciprocal of the Peclet number P:-

$$\frac{1}{P} = \frac{kT}{3\pi\mu \, d \, v \, D}$$
(4)

where k is Boltzmann's constant (energy per degree) and T is absolute temperature. In water filtration 1/P ranges from 10^{-8} to 0.5 x 10^{-5} . Although experimentation is difficult with sub-micronic particles Yao (1971) has submitted some evidence that equation (4) does represent the removal characteristics for these particles. It has also been shown that the smaller the particle the more efficient the collection due to their greater relative Brownian motion. Particles larger than 1 µm have a viscous drag that prevents any movement but for particles below this size Brownian motion becomes increasingly significant. Helminth eggs however, will be too large for this process to have any effect on their removal.

4.1.5.Hydrodynamic Action

In a uniform liquid shear field which derived from a constant velocity gradient across streamlines, a spherical particle will experience a greater liquid velocity on one side than the other. This makes the particle rotate and in turn the relative differences in velocity between one side of the particle and the other produces a pressure difference laterally to the flow. Therefore the particle moves to the region of higher velocity.

In a non-uniform stream flow, such as a filter pore, the particles are still subjected to the same lateral forces but the forces are not constant with position. This, coupled with non-spherical particles rotating due to the centre of mass not coinciding with the hydrodynamic centre, creates a unpredictable movement of particles through the streamlines. This could cause some of the particles to collide with the sand grains. There is no direct evidence of this occurring but Ison (1969) demonstrated changes in filtering efficiency when only the Reynolds number was varied. This change in efficiency could only be attributed to the hydrodynamic effect. The Reynolds number could be calculated by:-

$$R = \underline{v \ D \ \rho}$$
(5)

where ρ is the density of the liquid and D is the grain sand. In water filtration R has a value of approximately 1.0. Although equation (5) is a convenient expression it ignores factors that could intrinsically effect hydrodynamic forces. These are the particle size d, shear gradient g, the velocity of the particle relative to the liquid v_p and the angular velocity of the rotating particle Ω . Incorporating these variables allows four more Reynolds numbers to be derived which equally express the hydrodynamic effect. Due to experimental difficulties there has been no assessment of the real meaning of these numbers. It is therefore difficult to say whether this process will effect the removal efficiency of helminth eggs.

Another hydrodynamic action occurs when a particle is very close to a grain, that is less than the diameter of the particle. For contact to occur the water between the particle and the grain must be displaced. This displacement is a form of radial viscous flow. This scarcely affects the transport mechanisms but comes into effect when surface forces are involved. Therefore it is more appropriate to discuss this in the section of attachment mechanisms.

4.1.6.Combined Mechanisms

It is likely that all the mechanisms above act in unison on particles to transport them to the filter grains. Straining, as it is not strictly a transport mechanism, and inertia, as it has little effect can be disregarded when one tries to evaluate the efficiency of particle retention. This may be expressed as Λ which is the fraction of suspension retained in a filter layer one grain diameter thick. Λ may be expressed:-

$$\Lambda = -\Delta C \frac{D}{\Delta L}$$
(6)

where $-\Delta C/\Delta L$ is the concentration change per unit filter thickness and C is the concentration flowing into the layer ΔL . Comparison of equation (6) with deep bed theory (discussed later) indicates that $\Lambda = \lambda D$ where λ is the filter coefficient. Therefore in terms of filter transport mechanisms, filter efficiency can be expressed as:-

$$\Lambda = \text{const.} \left(\frac{d}{D}\right)^{\alpha} \left(\frac{kT}{3\pi\mu dvD}\right)^{\beta} \left(g(\rho_{s}-\rho)d^{2}\right)^{\gamma} \left(\frac{\mu}{vD\rho}\right)^{\delta}$$
(7)

where α is the exponent of interception group

 $\boldsymbol{\beta}$ is the exponent of diffusion group

 $\boldsymbol{\gamma}$ is the exponent of gravity group

and $\boldsymbol{\delta}$ is the exponent of the hydrodynamic group

Collecting together the terms gives:-

$$\Lambda = \text{const.} \qquad d^{\alpha - \beta + 2\gamma} \qquad (kT)^{\beta} (\rho^{S} - \rho)^{\gamma} \qquad (8)$$
$$\mu^{\beta + \gamma + \delta} D^{\alpha + \beta + \delta} v_{\beta +} \gamma_{+ \delta} \qquad \rho^{\delta}$$

Equation (8) indicates that in general an increase in particle size d, will improve filtration. An exception is where β is large compared to $\alpha+2\gamma$, which occurs for very small particles effected by diffusion forces. When particle size d, is too large for effective diffusion (small β) but too small for interception (small α) or gravity (small γ) a minimum in filter efficiency will occur. This has been demonstrated experimentally by Yao (1971) who showed a minimum in filter efficiency occurs at the 1µm sized particles.

Equation (8) also indicates that a small grain size D, and lower velocities v, give improved efficiency of collection. This is a well known experimental fact. It also implies that increases in temperature will improve efficiency due to lower viscosity.

The form of equation (8) indicates why there is little agreement among published results of the dependence of filtration efficiency upon various operating variables. It is assumed that the transport mechanisms operate simultaneously and their relative importance depends on the nature of the suspension as well as the operation of the filter.

Flocculation is a phenomenon that occurs within filter pores but the theory (Ives 1975) has not yet been experimentally tested. It appears however that it is independent of flow rate and directly proportional to the internal pore surface per unit pore volume.

Despite the theory and careful experimental study it is still not possible to predict filter performance from prior knowledge of the physical constituents of the process. Therefore the knowledge of transport mechanisms can rarely be used to predict general filter efficiency but can still be used to describe processes involved.

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4.2.ATTACHMENT MECHANISMS

For a granular filter to be effective in removing small particles, the particles must adhere to the filter grains. The way in which particles adhere to grains is solely to do with the surface characteristics of the grain and the particle. It is quite separate from the way in which the particles are transported to the grains. As well as requiring the ability to adhere to the grains, particles should also adhere to existing deposits. Without this a filter would be exhausted as soon as each grain had a monolayer of particles adhered to it. In practice this means that suspensions to be filtered should be colloidally unstable. In other words the particles should not repel each other sufficiently to prevent contact. The stability of colloids arises from an electrical charge on a particle and the consequent electrical repulsion between particles. Opposing this repulsion are the universally attractive Van der vaals forces. If Van der vaals forces are dominant the particles colliding with each other will stick together. This is referred to as flocculation. The most direct method of reducing colloid stability is to lower the effective electrical repulsion between the particles thus improving the effect The mechanisms discussed will include of attachment mechanisms. electrical phenomenon at interfaces, electrokinetic effects and the electrical interaction between particles. Van der vaals forces will also be briefly described.

4.2.1.Electrical phenomenon at interfaces

Between two phases in contact there often exists an electrical potential between them. This was shown by Guggenheim (1929) to be unmeasurable and therefore strictly speaking has no physical meaning. The concept however is important in understanding surface interactions between particles.

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The overall potential difference between the phases, Φ , can be considered as two separate charges:-

$$\Phi = \Psi_{0} + \chi \tag{9}$$

where Ψ_0 is the surface potential and χ is the chi potential. Ψ_0 arises from the unequal division of charge between the two phases and χ results from the orientation of electric dipoles at the interface.

Interfacial potentials can be created in at least five different ways. These are:-

i) Unequal dissolution of constituents ionsii) Ionisation of surface groupsiii) Isomorphous substitutioniv) Specific absorption of ionsv) Dipole orientation

In practice two or more of these mechanisms occur at the same time. In general they are only important in creating colloid insability and therefore have little relevance to the attachment of helminth eggs to sand grains.

One electrical phenomenon at the interface which may determine helminth egg attachment is based on the electrical double layer. An interfacial potential between a particle and the surrounding liquid is accompanied by a characteristic distribution of charge between the phases. Such a system has an overall charge of zero. The charge on a particle, together with the equal and opposite charge in the solution make up the electrical double layer. A model of this layer has been developed from the Gouy - Chapman (Gouy 1910, Chapman 1913) theory and the Stern - Grahame theory (Stern 1924, Grahame 1947). The resulting model is one with a layer of tightly held counterions (the Stern layer) adjacent to the interface. The potential falls sharply across this layer from a surface value Ψ_0 to a value Ψ_{δ} . δ is the thickness of the Stern layer and is around 0.5µm. With increasing ionic strength more counterions are located in the Stern layer and Ψ_{δ} decreases. If the adsorption of counterions is very strong it is possible that Ψ_{δ} could be opposite to Ψ_{0} . This may occur at the surface of a helminth egg wall because of the protein constitution of the wall (Francis and Monroe 1984). If so, this would effect the eggs ability to remained attached to a sand grain.

Electrical phenomena at the interfaces of particles are created, and they are a part of the integral ability of a filter to keep suspended particles attached to filter grains. It is likely that the different mechanisms act in unison to create a potential. The combined charge on <u>A.suum</u> eggs will be investigated later to determine whether this is an important process for the attachment of helminth eggs.

4.2.2.Electrokinetic effects

Electrokinetic phenomena occur when there is a relative movement at the boundary between two phases of which one must be a liquid. This is due to some of the charge in the diffuse part of the double layer being mobile and therefore can move with the liquid. Electrokinetic data are usually interpreted at the point of contact of the two phases, known as the "slipping plane". The potential is known as the zeta potential. The slipping plane is often assumed to be just outside the Stern layer.

There are four distinct electrokinetic effects which theoretically all involve the zeta potential. Due to the complexities of the experimental techniques they will only be briefly described here. The different effects depend on whether relative motion between the phases is caused by electrical or mechanical means and whether discrete particles or porous media are concerned.

The first is a streaming potential which is established when a solution is forced through a porous plug and acquires a charge in contact with the solution. A sedimentation potential is set up when charged particles fall through a solution. The third effect is

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electro-osmosis in which the flow of liquid through a porous plug is under the influence of an applied electric field and the final effect is electrophoresis which is the migration of charged particles in an electric field. This last effect will be used to estimate the electrical charge at the surface of an A.suum egg later in the thesis.

4.2.3.Electrical interaction

When two charged particles in water approach each other the diffuse parts of their double layers overlap. If these charges are the same a repulsion will occur. As the diffuse layers overlap the resulting energies can only be theoretically evaluated and tabulated values are available. Initially the interactions between two parallel flat plates will be discussed.

The potential distribution in solution between two charged plates is shown systematically in figure 4.1, where Ψ_1 and Ψ_2 are the surface potentials of the plates at the Stern layer (ie Ψ_{δ}) and Ψ_{m} is the minimum potential.





The form of this distribution is, in principle, given by:-

$$\left(\frac{dy}{dx}\right)^2 = 2 \cosh y - C \tag{10}$$

where h is Planks constant and $y = ze\Psi/kT$. The difficulty of this equation is determining the value of C.

Derjaguin and Landan (1941) showed that:-

$$P_{2} = nkT(C-2)$$
(11)

where P_a is the force per unit area between the plates, n is the number of ions per unit volume (m⁻³), k is Boltzmann's constant and T is the absolute temperature.

When there is a minimum potential between the plates (ie $(dy/dx)^2=0$) then C = 2 cosh y_m. Therefore:-

$$P = 2nkT(\cosh y_m - 1)$$
(12)

Equation (12) has been derived in a number of ways (Verway and Overbeek 1948) but it is interesting to note that the force does not directly depend on the plate potentials but on the minimum potential between them.

When there is no minimum between the plates and C < 2 the force becomes an attraction. This obviously happens when particles have opposite charges but also happens even with potentials of the same sign but of a different magnitude. This occurs when the plates are closer than a certain distance (Derjaguin 1954). Most of this theoretical work assumes that the potentials of particles remain constant during approach. It now appears that colloidal dispersions could not maintain electrochemical equilibrium and so the potential need not remain constant (Ferns 1972). Indeed it would be better to assume that potentials increase on approach in the case of colloidal solutions. The theory behind this is extensive and complicated and would not apply to the removal of helminth eggs in filtration and therefore will not be discussed further here. For a more extensive argument read Gregory (1975).

The interaction between spherical charged particles can, in principle and numerically only, be obtained by considering the overlap of the spherical double layers. A simpler though approximate method is that

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of Derjagiun (1934). He considered the interaction of spheres as a summation of the interactions of concentric rings on the sphere surface. The method is only valid when the minimum separation is much smaller than the radii of the particles. This leads to the expression:-

$$U_{\rm R} = 64\pi a_1 nkT Y_1^2 \exp(-\kappa d)$$
 (13)
 κ^2

where a_1 is the sphere radius, κ is the Dedye-Hukel parameter and Y_1^2 = tanh (y o/4). In practice both equation (12) and (13) give results which are intermediate between those from exact calculations using a constant potential and constant charge cases. In a granular filter the interaction between a suspension particle and a filter grain can often be treated as that between an infinite flat surface and a sphere (Ives and Gregory 1966). This leads to an interaction energy twice that for two spheres of equal radii, a, at the same separation, d, and with equivalent potentials. This is verified by putting $a_2 = \infty$ into equations (12) and (13).

The electrical interaction caused by two charged particles approaching will have an effect on helminth eggs. Therefore equations 10 through to 13 may have some use in determining the attachment mechanisms for helminth eggs.

4.2.4.Van der Vaals Forces

Van der Vaal suggested attractive forces between molecules over a century ago. The forces were shown to be of three types (Chu 1966). The first is the orientation effect. If both molecules have a permanent dipole they will tend to orientate themselves to give the most favourable electrical attraction. To some extent the thermal energy of the molecules oppose this effect. An induction effect occurs when one molecule has a permanent dipole and this inducts a further dipole in a neighbouring molecule. The strength of this effect will depend on the polarizability of the non-permanent dipole molecule.

The third effect occurs in molecules which have no permanent dipoles but still attract each other. A fluctuating dipole occurs in these molecules arising from the random motion of electrons within the molecule. Although only transient, such dipoles can induce dipoles in nearby molecules to give an attraction. This is called the dispersion effect.

Between particles the dispersion contribution is the only approximately additive effect over a significant distance. For this reason Van der Vaals forces are often referred to as dispersion interactions.

The effects of Van der Vaals forces are reduced by having a third media between the two interacting molecules. In the case of filtration this is water. There are many theoretical uncertainties concerning these forces, and it seems that Van der Vaals forces have only a minor effect in interfacial phenomenon.

The interfacial phenomena discussed in this chapter generally refer to colloids and their stability. Although helminth eggs are an order of magnitude larger than colloid particles, the same general principles of attachment should apply. The experimental work on the electrical charge of an egg detailed later in the thesis helped in determining the attachment mechanisms involved for helminth eggs.

4.2.5.Detachment

There is experimental and practical evidence that increasing the flow in a deep bed filter, when deposited particles are present in the pores, leads to detachment of some of these particles. It has been suggested that this also occurs at initial flow.

The removal of particles already deposited occurs at the plane of weakness which is unable to adsorb the shear stress as water flows through the pores. With greater velocities the pressure drop increases so detachment is more likely. It is, however, difficult to achieve any guidance to the fundamental property which effects detachment; the shear strength of deposits. There is no adequate method of measuring shear strength, especially of deposits in situ.

4.3. THEORETICAL MODELS OF DEEP BED FILTRATION

Deep bed filters are normally operated to clarify dilute suspensions. The filter will exhibit a pressure drop and this will rise as the filter becomes clogged. Therefore any mathematical model must represent both processes of clarification and pressure drop. Both have initial conditions when the filter is clean and both change in a time dependent manner during the process of clogging.

4.3.1.Clarification Models

Mathematical models of the clarification process are created to produce better design and to aid optimisation studies. They are based on measurements of the changes in suspension concentration, C, depending on the quantity of deposit per unit filter volume σ (specific porosity) and the independent variables of the distance from the inlet face of the medium L, and the elapsed time of filtration t. Pressure drop ΔP variation will depend on the same variables. The models therefore produced are used to predict C and ΔP in terms of the physical parameters and the operating variables of filtration. These include inlet concentration C_0 , filtration approach velocity v, grain size D, initial porosity ϵ_0 , water temperature, suspension particle characteristics and the surface chemistry of the system.

Despite an extended knowledge of surface chemistry, transport mechanisms and attachment mechanisms, it is still impossible to predict removal efficiencies from the physical components of the system. Therefore an empirical measure of the interaction between a uniform suspension and a uniform filter medium has been used for the basis of filtration clarification theory. This is the filter coefficient λ .

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The basic theoretical hypothesis is that the removal of suspension with respect to distance (depth) in the filter is of the first order. That is:-

$$-\frac{\delta C}{\delta L} = \lambda C \tag{14}$$

Equation (14) assumes that initially, when the filter medium is clean, every layer of the filter is equally efficient at removing particles from suspension. A second assumption is that at every layer the suspension entering and leaving it is uniformly dispersed. The pattern of removal is illustrated in figure 4.2.

Figure 4.2:General curve for the removal of particles (C) with depth (L)



At the commencement of filtration, when the elapsed filtration time is zero, equation (14) becomes:-

$$C = C_0 \exp(-\lambda_0 L)$$
(15)

Where λ_0 is the initial filter coefficient at t=0 and C₀ is the inlet concentration at L=0. This initial exponential decline is illustrated in figure 4.2.

This is the basic pattern of removal in rapid sand filtration. Although the theory has been expanded greatly by many researchers (Source ref Ives 1975), the details of more complex models are not required here.

4.3.2.Pressure drop models

The second major parameter with which to define deep bed filtration is that of pressure drop. If a filter media is clarifying a suspension, it follows that the pores are accumulating deposits which will cause a loss of permeability and therefore increase flow resistance.

The filter media exhibits a resistance to flow, even to clear liquid, which can be calculated by the Carman-Kozeny equation:-

where $(\delta H/\delta L)_0$ is the hydraulic gradient at t=0, υ is the kinematic velocity of the liquid and S_0 is the initial specific surface of filter pores. Head loss, H, is more convenient to use than pressure drop. When deposits are retained in the filter, the pores begin to clog and the head loss increases. This increased head loss, caused by the deposition in the pores, can be shown to be linear with time for most practical cases as shown by equation(17):-

$$H = H_{0} + k \beta_{a} v C_{0} t$$
(17)

where β_a is the bulking factor. This is a general equation and is independent of the form of the functions relating σ to L and t. This all assumes the filtrate concentration is low compared with C_0 (ie $0.05C_0$). This means that all the suspension flowing in up to time t (vC_t) has been retained in the filter.

In addition to the pressure drop created by deposits in the pores there may be an extra contribution due to deposits on the surface forming a discontinuous cake. This possibility was discussed in section 4.1. When such a cake arises the increase in head loss is exponential with time and is a specific example of Bouchers Law given at the beginning of this chapter, namely:-

$$H_{s} = k_{s} \exp(k_{t}t)$$
(18)

where k_s is the initial head loss through the surface layer and k_t is a rate constant of the surface deposition process. In deep bed filtration it is desirable to keep H_s to a minimum as it drastically reduces the length of a filter run.

Therefore there are three components of head loss, H_0 the initial head loss, H_d the head loss due to deposition in the pores and H_s the surface deposit head loss.

A convenient way of conveying the problems of head loss is through a diagram such as figure 4.3. In a static state 1 metre of pressure head is gained for every metre increment of depth. A head loss is experienced through a media which increases linearly according to the Carman-Kozeny equation for a clean filter. However, as clarification proceeds, the quantity of deposit varies from layer to layer and the consequent hydraulic gradient increases to distort the pressure line. When the pressure line touches the atmospheric pressure value it is desirable to end filtration to avoid air being drawn out of solution by sub-atmospheric pressure conditions.



Depth of filter

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In conclusion it is possible to say that the length of a filter run will be dependent on two parameters. The first is that clarification continues at the desired rate and the second is the limitation placed by the pressure drop. As the predictive model to be described in chapter 8 is designed to determine the number of eggs per litre of effluent, the clarification models are of direct relevance to this work. Indeed the equation representing the pattern of removal of helminth eggs in a rapid sand filter described in chapter 5 derives from equation (14). It is, however, useful to emphasis that no mathematical model has yet been able to predict the efficiency of filtration from the physical components involved.

4.4.THE THEORY OF BACKWASHING

Backwashing is an area of filtration that has had limited research considering the major importance and the extensive use of rapid gravity filters in the water industry today. There are still many unanswered questions as to the optimisation of backwashing and the theory behind this. Here only the fundamentals will be discussed with reference to experimental work undertaken by Cleasby et al (1975).

4.4.1.Fluidisation Fundamentals

The principle of cleansing by backwash is to create agitation by fluidisation. The phenomenon of fluidisation can be best visualised by passing a fluid upward through a bed of solid particles. The fluid (liquid or gas) encounters a resistance to flow and therefore has a resultant pressure drop ΔP . As the flow rate V is increased there is a linear relationship between ΔP and V. As V is further increased a point is reached at which the pressure drop is sufficient to bear the weight of the solid particles. Any further increase in flow rate will cause the bed to expand to accommodate the extra flow while maintaining the pressure drop effectively the same. The fluidised bed now effectively acts as a liquid. The fluid velocity that creates the onset of fluidisation is referred to as the minimum fluidising velocity, V_{mf} (Coulson and Richardson 1968). In an idealised bed this can be calculated by:-

$$\Delta_{pw} = (\rho_s - \rho_f)g(1 - \epsilon)$$
(19)
$$I_e$$

where Δ_{pw} = pressure drop across the fluidised bed I_e = height of expanded bed ε = porosity of expanded bed g = acceleration due to gravity ρ_s = particle mass density and ρ_f = fluid mass density

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The simplest bed expansion can be calculated by considering a bed which is fluidised from initial porosity ε_0 at height I₀ to a porosity ε and a height I_e. Since the volume of solids within a bed remains constant, then:-

$$I_{\rho}(1-\varepsilon_{\rho}) = I_{\rho}(1-\varepsilon)$$
(20)

This can be related to bed expansion. If the depth and porosity of a fixed bed are measured, and then expansion of the bed is observed at various flow rates, the porosity at each flow rate can be calculated from equation (20). If the log of velocity is plotted against log of porosity, a straight line results. The line can be extrapolated to a porosity (ϵ) of 1.0 (log ϵ =0). The intercept velocity at this point is denoted v_i. In a bed of uniformly sized spherical particles v_i is equal to the unhindered settling velocity of the particles v_s. However, with non-spherical particles, v_i is appreciably higher than v_s. The equation for the straight line referred to above is:-

$$\frac{v}{v_{i}} = \epsilon^{n}$$
(21)

where v is the superficial velocity, v_i is the intercept velocity at ϵ = 1.0 and n is the slope of the straight line. This is the fundamental equation applicable to all fluidised beds and is useful in various mathematical models for bed expansion. For the special case of spherical particles:-

$$\frac{v}{v_{s}} = \epsilon^{n}$$
(22)

Where v_s is the unhindered settling velocity of the particles.

If fluidisation is essential for the cleansing of an expanded filter bed how can the optimum level of fluidisation be calculated from the measurable properties? A model for this was developed by Woods (1973). This model involved three steps. The first was to calculate v_i from measured properties by correlating the Reynolds number based on the intercept velocity, Re_i , and the Galileo number, Ga. The Galileo number can be calculated by:--

$$Ga = \frac{d^3}{\mu^2} \rho(\rho_s - p)g$$
(23)

where d is the mean diameter of bed particles and μ is the viscosity of the fluid. Re $_i$ can be calculated by :-

$$Re_{i} = dv_{i}\rho$$
(24)

An example of such a correlation, developed for garnet sand is:-

$$Re_i = 0.0702Ga^{0.823}$$

The second step calculates the slope of the straight line, n (as in equation (21)) using a correlation between n and Re_i. This correlation for garnet sand is:-

$$n = 5.768 \text{Re}_{1}^{-0.0541}$$

From this the bed porosity can be calculated at any desired superficial velocity using equation (21) and the values of v_i and n determined above.

This approach has been successfully applied to a graded and uniform garnet sand, and should be equally applicable to silica sand and crushed anthracite coal.

From the above model the optimum bed expansion could be calculated and discussed. The cleaning which results in a water fluidised bed is due to the hydro dynamic shear at the water/filter grain interface. A simple mathematical model was developed for the porosity of maximum hydraulic shear by Amirtharajah (1971). He used a classical equation

for the velocity gradients induced as a function of power input to a fluid body (Developed by Camp 1943 and Fair et al 1968), equations (19) and (21), to obtain:-

$$\varepsilon = \frac{n-1}{n}$$
(24)

From this equation it was shown that a maximum hydraulic shear occurs at a porosity of 0.68 to 0.71. Since to the curve of the shear forces against ε being flat around the optimum , the hydraulic shear is insensitive to changes in ε . For example a change in backwash porosity from the optimum of 68% to 60% results in a 2.5% decrease in the shear force.

The cleaning of granular filters by water backwashing alone is, however, an inherently weak cleaning method (Cleasby et al 1975). This is because particle collisions do not occur in a fluidised bed and thus abrasion between the filter grains is negligible. During backwash there is a slight saving in water by expanding the bed to the optimum porosity. Lower rates result in nearly the same efficiency cleaning but proportionally longer times will be required. Therefore there is no economy achieved in using lower backwash rates.

Due to the low efficiencies of backwashing with water alone Cleasby (1972) experimented on the benefit of air scour. This has the benefit of creating greater collisions between particles so abrasion as well as shear forces will remove particles. When flocs that have settled in pores rather than those crystallised on grains were used, there was no noticeable difference between water and air scour as opposed to water backwashing alone. A difference was only noted in flocs that crystallised on to the sand grains. Due to the fact that helminth eggs would be unable to crystallise on filter grains, and the increased technology required for air scour, it is doubtful whether this cleaning mechanism is applicable in developing countries.

During Cleasby's work (1972) there were two discoveries of note. The first is that their is no apparent difference in effluent quality even after the fifth run of a series and the other is that backwashing, even with air scour, does not reduce the quantity of media by any significant amount. It was calculated that a 3% loss of anthracite coal over five years' use was likely. The effective size of the media reduced slightly over this period but this was an insignificant amount compared to the 10% leeway suppliers request when supplying the media.

Backwashing with water only as a process for cleaning filters can be considered as adequate for removing helminth eggs. Despite research proclaiming it to be an inherently weak process, it would seem that a developing country interested in filtration as a method for removing impurities could rely on water backwash alone as a sufficient cleaning mechanism.

CHAPTER 5: MATERIALS AND METHODS

5.1.CHOICE OF INDICATOR ORGANISM

Having chosen to concentrate on the efficiency of rapid sand filtration in removing helminth eggs appropriate steps to ensure the safety of the researcher where required. It was decided that <u>Ascaris</u> <u>suum</u> eggs (pig Ascaris) would be used for the research. There were three major reasons for this: i) the organism is very similar to <u>Ascaris lumbricoides</u> (human Ascaris); ii) it has a low pathogenicy to humans; and iii) it can be readily obtained in large numbers from mature female worms obtained from a local abattoir.

i) Similarity to Ascaris lumbricoides

During the early years of parasitology there was no differentiation made between <u>A.suum</u> and <u>A.lumbricoides</u> (Roberts 1946, Pattersen and Axel 1941). The species were considered interchangeable between their hosts of pig and man. It was not until the early 1950's when <u>A.suum</u> was first considered as a variety and then as a distinct species (Jaskoski 1955). It is now widely accepted that <u>A.suum</u> is a separate species, with the similarities noted (Bradley and Hadidy 1981). The only described way of distinguishing between the species is by examining the morphology of the teeth of the adult worm. The eggs look identical, although <u>A.suum</u> eggs are slightly larger (Spencer and Monroe 1982).

ii) Pathogenicy of Ascaris suum

There are now only rare reports of children being infected with <u>A.suum</u> (eg Crewe and Smith 1971). It is generally accepted that <u>A.suum</u> is likely to infect only healthy individuals if they are already infested with <u>A.lumbricoides</u>.

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iii) Collection of Ascaris suum

The collection of A.suum eggs was a relatively simple procedure. Adult worms were collected from a local abattoir from slaughtered The worms were sexed and only the largest females (over about pigs. 150mm long) were chosen. The male of the species tended to be much smaller than the female and the male had a hooked posterior end. The female worms were then dissected under water and 5cm of uterus attached to the vagina was removed. This was an insurance that the eggs were fertilized. The uterus was then placed in a mortor and ground with a pestle in distilled water. The large pieces of uterus were removed by filtering through a wire mesh. The result was a milky solution which numbered about 200,000 eggs per female dissected. The eggs were then incubated at 37°C for four weeks so the majority of the eggs reached the blastulus stage of development. This was the most efficient way of determining viability. Egg development is shown on plate 5.1. One collection of worms would provide 30-40 million eggs. Availability was a great inducement for their use.

It can be deduced from the above that <u>A.suum</u> eggs are good indicators. The fact that the eggs are larger than most helminth species $(40-60\mu m \times 30-50\mu m$ Feachem et al 1983), have a sticky outer layer (Kennedy 1990 (personal communication)), and a relatively high density (1.1036 Magat et al 1972) means that these eggs are more likely to be removed by filtration. Therefore, if these eggs are capable of moving through a filter then it can be assumed that the eggs of other helminth species would also move through a filter. Conversely, any model developed through the use of <u>Ascaris suum</u> eggs would give a value that may not apply to smaller eggs. This was compensated for by a correction factor discussed in chapter 8.

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<u>Plate 5.1:Ascaris suum egg development during four weeks</u> <u>incubation at 37^oC</u>

plate 5.1a:Eggs at variousplate 5.1b:Egg at one cellstages of developmentstage of development.surounded by the uterus vall.

plate 5.1c:Egg at two cellplate 5.1d:Egg at four cellstage of development.stage of development.

<u>Plate 5.1e</u>:Egg at eight cell <u>plate 5.1f</u>:Egg at sixteen cell stage of development. stage of development.

plate 5.1g:Egg at the blastula plate 5.1h:Egg with worm larvae stage of development clearly visible









5.2.METHODS OF EGG DETECTION

The initial experiments were devised to determine the possibility of eggs passing through a sand filter and the techniques required to detect eggs within a column. For this a 50mm diameter column was used with a slow flow rate and a high concentration of eggs.

5.2.1. Apparatus criteria

A 1.5 metre column was filled with 75cm of coarse sand (effective size >1mm) and a constant flow of 22 ml/min was allowed to pass through the filter. This is equivalent to 0.65 m/h, a slow rate for an operational rapid sand filter. 2000 eggs per litre were added by means of a winchester drip feeding the top of the column. This high concentration was added to allow every chance for the eggs to be detected in the filter. The runs were for 20 hours. Water was allowed to flow over the top of the column so that a constant head of 70cm was maintained. After 20 hours the sand was sampled and treated for the detection of eggs.

Several methods for egg detection were evaluated. It has been reported that Ascaris eggs are difficult to detect due to a sticky outer coating. Therefore all the glassware was coated with a silicone non-stick lining called Repelcote (B.D.H). The sampling points referred to are shown in figure 5.1. The numbers refer to sampling points that were designed to allow only liquid out of the media. This was insured by a wire mesh placed on the inside of the sampling apparatus. The sampling was taken from 25mm inside the media to prevent the sample being influenced by the perspex wall. The letters refer to sampling points that consist of a drilled whole plugged with a rubber bung. At these locations the sampling procedure consisted of removing the bung and allowing the sample of sand and water (or if the column had been drained, sand only) to flow out of the hole.

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Method 1

A sample of water was removed from each of the sampling points one to nine at h end of the filter run. The sampling points had a wire mesh to prevent sand from entering the sample. 100ml of water was collected and placed in measuring cylinders to allow settling overnight. The supernatant was then removed using an upturned pastor pipette, and the resultant concentrate was enumerated for eggs in a MacMasters slide under a microscope at x40 magnification.

Method 2

A sample of sand and water was removed from each of the sampling points A to D at the end of the filter run. The sample was stirred for at least five minutes with a magnetic stirrer before the water was decanted off. The water was then enumerated as above. The volume of water was recorded and the sand was dried in an oven and weighed. To express the results as eggs/cm of column the total weight of the sand in the column was calculated and then divided by the depth of the sand. For the 50mm column each centimetre of sand was 54g. These units were used as the sampling points were 10mm in diameter and would take sand from a depth range rather than a specific point. These units also standardised the results for each column.

Method 3

The column was drained of water and samples of sand were taken from sampling points A to D at the end of the filter run. To the samples 20ml of surfactant was added (Deacon 90) and the mixture was stirred for at least five minutes. The surfactant was decanted and enumerated as for method 2. Again the results were expressed as eggs/cm column.

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Method 4

Samples of sand were collected as in method 3. The samples were placed in a glass tube and a known quanity of surfactant was passed through the sand and this 'filtrate' was enumerated for eggs. The results were expressed as with method 2.

Method 5

Samples were again collected as in method 3. To each sample 20ml of saline solution (NaCl) was added. The mixture was stirred for five minutes and the decanted samples enumerated. The saline solution was to encourage the eggs to float and therefore be less affected by the sand.

After draining, the column was backwashed for 20 minutes at half bed expansion and the above methods were repeated to determine the presence of eggs after cleaning.

5.2.2.Recovery rates from the proposed methods

The results for methods one to five are shown in table 5.1. The number in brackets denotes the number of eggs in 0.5ml which is the full quantity of liquid held in each half of a MacMasters slide. This was only counted if no eggs were found in the initial 0.15 ml, the volume of water held within the grid of a MacMasters slide.

Only Method 2 yielded any results for the backwashing and these results are shown in Table 5.1c.

The results from method 1 show that eggs have the ability to penetrate the sand bed. Method 1 is a grab sampling technique (ie the sample is taken at one particular point in time) and will measure the eggs not bound to the surface of a sand grain at the instance of sampling. The eggs detected by this method still have the ability to pass further into the filter bed.

Method 3 measures eggs that are bound to sand grains surfaces only to be removed by agitation and the action of the surfactant. There is a strong possibility that as the filter is drained eggs will be deposited onto the sand grains, which in the course of normal filtration, they would pass. It is, however, methodologically a better technique than method 2 which does not maintain a constant Method 2 will not detect eggs that remain in the volume of water. water within the pores of the sand after sampling. Tests on the efficiency of this technique (appendix B) show that anything between 2 and 10ml of liquid can be lost within the pores of the sand during This is an error that method 3 overcomes. sampling. Method 2 however, gives the higher results. This was thought to be due to the procedure of detecting eggs from the sand surface and within the It is not possible to compare methods 1 with methods 2 and 3 water. as methods 2 and 3 are accumulative procedures detecting eggs from the complete run whereas method 1 measures eggs present at the instant of sampling.

Method 4 gave no results. The reason for this is not known but may be because the surfactant did not get sufficient time or agitation to wash the sand surface and remove the eggs. This method was not considered further.

Method 5 gave few results. There are several possible explanations for this. The first is that the saline solution was unable to remove the eggs once they were attached to the surface of the egg. The second is that the eggs floated to the surface of the solution and were not taken into the pipette before the sample was placed in a MacMasters slide. The technique was not successful and therefore was no longer considered.

After backwashing only method 2 detected eggs. This was thought to be because the other techniques were not sensitive enough to detect eggs.

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TABLE	5.1a:REC	OVERY	RATES	0F	METHODS	1	ΤO	3

sampling	; c	oncen-	coun	ts pe	r	eg	gs/ml	eggs/100m	
=									
	1	10	9	11	 14	9	71.67	717	
	2	10	0.00	1	1	1	3.16	32	
	3	10	0(0)	0,00	0,00	0,00	0.50	5	
	4	10	0(1)	0(0)	0(0)	0(0)	0	0	
	5	10		0(0)	0(0)	0(0)	1.00	10	
	6	10	0(0)	0(1)	0(1)	0(0)	1.50	15	
	7	25		0(2)	1	0(1)	2.42	10	
	8	25	0(0)	0(1)	$0_{(0)}$	0(2)	0.50	2	
	9	25	$^{0(1)}_{(0)}$	$^{0}(0)$	$^{0(0)}_{(1)}$	$^{0(0)}_{(1)}$	1.00	4	
Results	from Met	thod two							
sampling	depth	weight	ml of	coui	nts p	er	Total	eggs/g	eggs
point	sample	of sand	water	0	.15ml		eggs	dry	per cm
	(cm)	(g)						sand	column
======================================	======== 2	21.2	28	====== 81 72	2 66	===== 72	13580	640.6	15810
B	5	36.7	26	33 42	2 27	23	5416.7	147.6	3642
c	34	30.4	21	0,,,3	3 0	0	76.4	2.5	62
D	59	29.1	21	$0^{(1)}_{(0)}$	$10^{(1)}_{(1)}$	$)_{0}^{(1)}$	25.5	0.87	21
effluent	74	_				 -			0
Results :	from Met	hod three	e						
sampling	depth	weight	ml of	cour	nts pe	er	Total	eggs/g	eggs
point	sample	of sand	water	0.	15ml		eggs	dry	per cm
	(cm)	(g)						sand	column
A	2	3.1	20		8	8	966.7	311.8	12246
В	5	4.9	20	33	2	3	366.7	74.8	1892
С	34	6.4	20	$0_{(1)}$	000°	ω^1	12.1	1.89	88
D	59	7.1	20	$^{0(0)1}_{(0)}$	· · · · · · (0) ⁰ (0)) $^{12.1}$	1.71	42
effluent	74								0
		-	_			_	_		

Results from Method 1

TABLE 5.1b:RECOVERY RATES OF METHODS 4 AND 5

Results from technique four

sampling point	depth sample (cm)	weight of sand (g)	ml of water	counts per 0.15ml	Total eggs	eggs/g dry sand	eggs per cm column
=============		==========		*================	*******	=======================================	
A B C D	2 5 34 59	37.4 45.3 42.1 47.3	30 30 30 30	${}^{0}_{0}(0){}^$	0 0 0 0	0 0 0 0	0 0 0 0
effluent	74		_		_		0

Results from Method five

sampling point	depth sample (cm)	weight of sand (g)	ml of water	counts 0.15m	per nl	Total eggs	eggs/g dry sand	eggs per cm column
						100	E 10	
A	2	19.3	20	$0_{(0)}$	1 1	100	5.18	280
В	5	13.2	20	0(0)(1)	0,00,0	33.33	2.525	136
С	34	17.1	20	0,00,0,0		<u> </u>	0	0
D	59	15.3	20	0(0) 0(0) 0(0)	0(0) 0(0) 0(0)	<u> </u>	0	0
effluent	74	_	_			_		0

TABLE 5.1c:RECOVERY RATES OF METHOD 2 AFTER BACKWASHING

sampling point	depth sample (cm)	weight of sand (g)	ml of water	counts per 0.15ml	Total eggs	eggs/g dry sand	eggs per cm column
A B C D	2 5 34 59	9.8 24.4 31.2 39.9	43 40 25 27	${\overset{0}{_{2}(1)}}^{0}_{1}(0)^{0}_{1}(2)^{1}_{2}$ ${\overset{1}{_{0}(1)}}^{0}_{0}(0)^{1}_{0}$ ${\overset{0}{_{(1)}}}^{1}_{0}(2)^{1}_{2}$	104.2 400 57.7 103.8	10.6 16.4 1.85 2.6	666 450 109 64
effluent	74	_			_		0

From the results it was decided to concentrate on methods 1,2 and 3. These were the techniques that seemed most likely to give consistent and reliable results. Efficiency experiments were conducted on these techniques involving known volumes of sand and known volumes of water being seeded with known quantities of eggs. The mixtures were then stirred for at least five minutes and the eggs were counted in a MacMasters slide as before. The results from these efficiency tests (see appendix B) gave recovery rates of approximately 40%. Although this is a low efficiency the methods were easy to conduct and relatively reliable. The results from each sample had variation of $\pm 28\%$. Although this variation is larger than is desirable it was considered that these techniques would give the most consistent results.

After a repeat run on the 50mm column a pilot study was conducted with a pair of identical 125mm columns.

5.3.PILOT STUDY

The major reasons for a pilot study with the 125mm diameter columns was to assess the similarity of two identical columns and to help confirm the reliability and consistency of the detection techniques. The size of the column was increased to reduce the effect of the walls the filtration. It has been established that if the diameter of on the column is 50 times greater than the largest sand grain the wall effect will be negligible (Ives 1975). The 125mm column allowed this ratio with the largest sand grain only 1.1mm. As the column diameters increase there is an exponential increase in the volume of water and the number of eggs required to maintain equivalent flow rates and The 125mm column satisfied the wall influent egg concentrations. effects and did not place unmanageable demands on the pumps or the number of eggs required. Two identical rigs were used to provide an element of control. To be statistically correct six columns would be required but this was impractical in both material and time.

The column was made out of perspex with a conical iron detachable base. A layer of glass balls, designed to produce an even flow of water during the backwash procedure, was placed inside the base. The efficiency of this will be discussed later. Fine gravel was placed above the glass balls before the 77cm of sand media was added. A one metre head was produced by bolting a perspex tube above the column.

The rigs used for the pilot study had nine water sampling points, but the sand sampling points were increased to six as shown in figure 5.2. These were placed at 1,16,31,46,61 and 76cm of sand depth. The methods used were 1,2 and 3. It was considered unnecessary to persevere with methods 4 and 5. The concentration of eggs in the influent was 250 eggs/litre. This value was obtained from field work in Jordan where concentrations of over 400 helminth eggs per litre were not uncommon (Dunn 1988). This also allowed enough eggs to be present so detection would be possible. The flow rate was one litre a minute (equivalent to 4m/h) and the length of run was 22 hours. Five duplicate runs were carried out.

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5.3.1.Results of pilot study

Table 5.2 gives the results from both rigs using methods 2 and 3 and after the backwash when eggs were detected with method 2.

Run	depth of	Meth	od 2	Meth	od 3	Back	wash
	<pre>sample(cm)</pre>	Rig A	Rig B	Rig A	Rig B	Rig A	Rig B
ONE	1	13278	9949	11308	5299	2049	0
	16	739	236	335	99	79	0
	31	177	138	158	335	138	0
	46	315	276	414	118	118	0
	61	79	177	138	99	59	0
	76	0	39	217	197	0	0
TWO	1	21394	14066	6757	9515	1592	1221
	16	264	193	384	843	83	106
	31	278	207	437	333	41	37
	46	207	130	325	193	158	97
	61	181	53	201	230	75	0
	76	93	106	230	106	156	95
THREE	1	17986	16587	19267	11383	611	861
	16	384	370	985	690	276	209
	31	236	116	376	286	0	71
	46	126	41	234	534	118	35
	61	65	87	242	189	191	0
	76	110	65	158	242	59	51
FOUR	1	20094	13140	11682	12372	524	532
	16	453	164	345	307	49	39
	31	175	179	215	116	53	41
	46	0	104	508	296	83	0
	61	83	63	272	112	100	106
	76	73	41	197	223	116	57
FIVE	1	18991	24763	10441	15760	205	709
	16	191	158	648	374	47	49
	31	122	97	234	331	73	97
	46	67	112	357	242	47	47
	61	55	63	89	0	0	126
	76	61	61	45	171	39	45
Average	e 1	18349	15701	11891	10866	996	665
	16	406	240	539	463	107	81
	31	198	147	284	280	61	49
	46	143	133	368	277	105	36
	61	93	89	188	126	85	46
	76	67	63	169	188	74	50

Table 5.2:Results of five runs using 125mm columns (eggs/cm column)

Method 1 gave no results. This was considered to be due to the greater volumes of water involved and the lower concentration of eggs in the influent. As this method is based on a grab sample the sensitivity of the technique is reduced by the lower concentration of eggs in the filter.

5.3.2.Statistics on pilot study

Two major statistical tests were carried at on the data. The first was the comparison of two independent variables to see if there was any significant difference between the results given at each sampling point from both rigs. The second was a matched pairs analysis to establish if there was any significant difference between the rigs as a whole. The calculations of these statistical tests can be seen in appendix C.

The F-values indicate the difference between corresponding sampling points of the rigs. These results show that there is no significant difference between the sampling points of each rig (p>0.05). This implies that the sampling points on each of the rigs are sampling from the same population. In other words, it is valid for the results from the sampling points of each rig to be statistically combined to give an overall mean.

The t-values for the comparison between the two rigs as a whole also indicates that there is no significant difference between the two rigs (p>0.05). This implies that one rig is equivalent to the other rig. As there is no significant difference between the two rigs it is valid to combine the results of both as a whole. This also suggests that the results are an accurate representation of the removal efficiency of these model rapid sand filters with respect to Ascaris suum eggs.

Therefore, the combined averages of these results can be used to establish a mathematical equation that will represent the pattern of removal of <u>A.suum</u> eggs during rapid sand filtration using the apparatus discussed in this chapter.

5.3.3.Mathematical representation of the removal pattern

The results show a trend of a decreasing pattern in the numbers of eggs as deeper parts of the filter bed are reached. The decrease seems similar to an exponential decrease. Tests using the χ^2 significance procedure showed that an exponential decrease significantly varied from the decrease illustrated by the combined averages from the pilot study using method 2 (shown in table 5.3). Therefore the rate of decrease was not purely exponential. This can be illustrated by graph 5.1.

Table 5.3:Combine averages Results of method 2 during the pilot study

Depth of sample(cm)	Eggs/cm Column
===============	
1	17025
16	315
31	173
46	138
61	91
76	65

The rate of decrease can be represented by the differential equation:-

$$\frac{\mathrm{d}N}{\mathrm{d}x} = -\mathrm{k}N \tag{1}$$

where x is the sand depth, N is the number of eggs per centimetre of column and k is a constant. k is negative as the number of eggs decreases with depth. Dividing equation (1) by N and integrating with respect to x gives:-

$$\int \frac{1}{N} dN = \int -k dx$$
 (2)

If k is constant then:-

$$\ln N = -kx + D \tag{3}$$

where D = the number of eggs in the first centimetre of sand (N_0) .

$$\Rightarrow \text{ when } x=0 \quad N = N_{o} \tag{4}$$

Therefore equation (2) can be expressed:-

$$\ln \left(\frac{N}{N_{o}}\right) = -kx$$
 (5)

$$N = N_{o} e^{-kx}$$
(6)

Equation (6) is an exponential curve which was shown graphically (Graph 5.1) to be unlike the real curve. The equation of the line depicted by the results can be more closely adhered to by assuming k is related to x by:-

$$k = cx^{p}$$
(7)

where c and p are constants. This would change equation (2) to become:-

$$\int \frac{1}{N} dN = \int cx^{p} dx$$
 (8)

$$\equiv \ln \left(\frac{N}{N_{o}}\right) = \frac{-cx^{p+1}}{p+1}$$
(9)

$$N = N_{o} e^{-(c/p+1)x^{p+1}}$$
(10)

or

or

This is equivalent to the general equation:-

$$N = N_0 e^{-ax}$$
(10a)



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where a = c/p+1 and b = p+1. From the equation above, it can be seen that the term p has the most influential effect on the shape of the line of removal. Limits on p can be obtained.

If p > 0 then as x increases the e term decreases. This creates a line below that of an exponential curve. As has already been shown (Graph 5.1) the true line is above an exponential curve. Therefore p must be less than zero.

If p < -1 and c remains positive the e term becomes positive which would show an increase in the number of eggs caught with an increase in depth. This too is not the case.

If p = -1 then when x = 0, N = 0, which is not the case, and if p = 0 the line would be an exponential curve. Therefore the limits on p can be expressed as:-

$0 > p \ge -1$

As will be shown later all p values calculated for the data collected comply with these limits. This is a good indication that the assumed relationship:-

$$k = cx^p$$

is an accurate representation of the processes of removal as shown by the data collected in the pilot study.

Equation (10) can be simplified into a straight line by the following procedure:-

In equation (9) let $y = \ln(N/N_0)$ and a = c/p+1. Equation (9) then becomes:-

$$-y = ax^{p+1} \tag{11}$$

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By taking natural logs equation (11) becomes:-

$$\ln (-y) = \ln (ax^{p+1})$$
 (12)

$$\equiv \ln (-y) = \ln a + (p+1)\ln x$$
(13)

Then let $\ln (-y) = F$, $\ln x = G$ and $\ln a = A$ and the equation becomes:-

$$F = A + (p+1)G$$
 (14)

which is a straight line.

Equation (14) can be used to assess the validity of equation (10) as the equation of the line. By using the values in table 5.3 the equation of the line for equation (14) can be calculated and a correlation of F agaisnt G performed. This in turn can be used to calculate values for the constants in equation (10). The calculations for obtaining the line are shown in appendix D where a = c/p+1 and b =p+1. These calculations show that the equation for the line of decrease in the pilot study can be expressed as:-

$$N = N_0 e^{-2.2403202x^{0.2066313}}$$
(15)

The correlation coefficient, r, is an indication of how close the line of best fit correlates with the actual data. The r value for the pilot study data using method 2 is 0.98 and this indicates an excellent correlation.

5.4.DATA COLLECTION

The apparatus was altered with the addition of an extension tube to allow for a one meter head. The sampling points designed for method 1 were used to create a manometer so the head loss could be measured. A photograph of the apparatus used during the data collection is shown in plate 5.2. A flow rate of one litre per minute was applied (equivalent to 4 m/h) and the influent had a concentration of 250 eggs/l. Five duplicate runs were carried out on each of five various sand sizes. These were 0.3,0.4,0.5,0.6 and 0.7 mm effective sizes. The uniformity coefficient was 0.5,0.57,0.62,0.7 and 0.8 respectively. measurements were obtained using British standard sieves. A11 Porosity experiments showed that minimum porosity was reached within each rig for each sand size. As the sand maintained the same sphericity (0.7) it was calculated that porosity remained the same for each sand size. The typical sphericity of the sand grains from the various sampling point are shown in plate 5.3. The calculations on porosity were confirmed by data from Kolbuszewski and Alyarak (1964). Therefore, the standard weight for each centimetre of column was calculated at 197g regardless of the effective size of the sand.

The experiments were then extended to include varying flow rates. Using the 0.5 effective sand sizes flow rate was increased to 1.5 litre/min (equivalent to 6m/h) and decreased to 0.5 litre/min (equivalent to 2m/h).

Experiments were then conducted to observe the effect of dual medias. Both sand/anthracite and sand/garnet dual medias were used. The anthracite had a porosity of 0.749 (42% was void) so each centimetre was equivalent to 98g. The garnet had a weight of 352g/cm of column.

A final set of experiments were conducted using sewage effluents seeded with eggs. Due to the difficulty of obtaining large quantities of sewage effluent it was impractical to use effluents all the time. These runs were to validate the results of the water only experiments.

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Plate 5.2:Colour photograph of the apparatus used during the data collection



<u>Plate 5.3:Average sand grain size and sphericity in a 0.5mm</u> effective sand size filter bed

<u>plate 5.3a</u>:Sand grains from sampling point A (mean diameter 442 µm) plate 5.3b:Sand grains from sampling point B (mean diameter 656µm)

plate 5.3c:Sand grains from sampling point C (mean diameter 819µm) plate 5.3d:Sand grains from sampling point D (mean diameter 882µm)

plate 5.3e:Sand grain from sampling point E (mean diameter 959µm) plate 5.3f:Sand grain from sampling point F (mean diameter 1036µm







Eggs were tested in the effluent of the filter from the 0.7mm runs The late start of this detection method was due to onward. the difficulties in finding a reliable method to detect low numbers of eggs in large volumes of water. The method eventually used was a large bucket with a removable glass bottom. The effluent was fed to the bottom of the bucket and allowed to dribble over the top at a velocity lower than the settling velocity of Ascaris suum eggs. Therefore, the eggs were allowed to settle on to the glass. The glass was washed and the eggs present were enumerated in the MacMasters slide. This method is very crude but it was the only method which gave results for eggs in the effluent of the 125mm diameter rapid sand filter.

Other techniques tried included filtering the effluent on glass fibre filters and staining eggs with iodine. Filtration failed due to the slow rate of flow through the glass fibre filters which created an unmanageable back log of water. The staining of the eggs with iodine was reasonably successful when observed using reflective microscopy. Fluorescent staining of the eggs was less successful but neither method gave any results for the eggs in the effluent. Therefore the method using the bucket was continued.

CHAPTER 6:RESULTS

The complete set of results is shown in appendix E. In this section the mean results will be discussed with respect to making them comparable and determining their reliability. The main trends will be highlighted for discussion later.

6.1.RESULTS FROM EFFECTIVE SIZE EXPERIMENTS

The results for the data collection from the variation of the effective size are shown in tables 6.1-6.3.

TABLE 6.1:Results from method two

	Effec	ctive	Sand S	Size	(mm)					
depth of	mean	sd	mean	sd	mean	sd	mean	sd	mean	sd
sampie(cm)			0.4				0.0			
1	23108	7255	39256	1174	38303	4740	35559	8918	40590	5131
16	209	59	441	231	639	248	1034	626	1569	603
31	73	40	170	60	302	133	454	335	840	490
46	66	18	141	48	253	111	251	137	619	277
61	35	16	87	35	184	127	202	79	438	227
76	31	22	58	13	148	45	137	54	219	65

TABLE 6.2:Results from method three

depth of sample(cm)	Effec mean 0.3	ctive sd	sand s mean 0.4	size sd	(mm) mean 0.5	sd	mean 0.6	sd	mean 0.7	sd
1	29664	3589	34851	5494	35300	5897	37887	6831	39959	7226
16	688	319	897	362	918	493	2056	895	2330	663
31	238	84	332	46	513	160	775	299	1251	490
46	170	97	226	56	365	129	466	112	786	326
61	84	53	121	44	297	171	308	103	508	168
76	63	46	99	25	229	140	238	70	366	111

	Effec	tive	sand s	size	(mm)					
depth of	∎ean	sd	mean	sd	mean	sd	mean	sd	mean	sd
sample(cm)	0.3		0.4		0.5		0.6		0.7	
1	1156	598	1677	1301	 789	1288	1314	724	3173	2474
16	96	97	106	51	164	241	101	58	209	137
31	12	26	18	12	77	117	64	92	55	74
4 6	25	23	21	20	153	200	50	69	0	0
61	20	27	26	44	96	79	25	55	43	78
76	18	23	30	25	64	98	21	46	17	38

TABLE 6.3:Results for backwash using method two

All the mean results show the same pattern of removal from 1cm depth to 76cm depth. Between effective sand sizes there is an increase in the number of eggs captured at one centimetre depth as the effective size increases. There is also an increase in the number of eggs captured at 76cm sand depth with increasing sand sizes. The results from each effective sand size are not however comparable in the present form. They first need to be corrected by relating the number of eggs detected to the number of eggs added. This can be done by first calculating the constants of each line of removal using the procedure described in section 5.4. Due to the randomness of the backwash results, illustrated by the large standard deviation, it was not considered reliable to apply the same procedure to these results. It is however, interesting to note that eggs were detected at all the levels of the filter bed after the backwash procedure. This implies that the backwash was not a totally efficient method of cleaning the filter. This point will be discussed later. The constants for the pattern of decrease as detected by methods 2 and 3 are shown in Table 6.4. 'a' refers to the term c/p+1 and 'b' refers to the term p+1 in equation (10) of section 5.3.3. The actual numbers in table 6.4 describe the equations of the straight lines, and, as such, the number of decimal places shown is chosen to indicate differences between the lines.

It is interesting to note that the 'a' values generally seem to decrease with increasing effective sand size, and the 'b' values generally increase. The notable exception to these trends is that the results from Method 3 during the 0.5 effective sand size. However,

Table 6.4:Calculated constants for the effective size

experimental data

EFFECTI	VE METHO ZE	D 2		METHO	D 3	
(mm)	a	b	a x b	а	Ъ	a x b
0.3	2.634	0.216	0.568	1.597		0,500
0.4	2.470	0.228	0.563	1.594	0.304	0.485
0.5	2.449	0.190	0.465	2.086	0.204	0.425
0.6	1.611	0.287	0.463	1.109	0.357	0.395
0.7	1.472	0.281	0.414	0.999	0.369	0.368

the values for the product 'ab' show a general decrease with increasing sand sizes. The straight lines represented by the constants indicated in table 6.4 are illustrated in graph 6.1. From these it is possible to see that, in general, the lower effective sand sizes have the lower gradients and start higher up the y-axis. This means that the lines cross and for seven of the ten lines the cross occurs at around the value of 7.5.

With the equation of the line for each effective sand size the total number of eggs detectable by the detection methods in the filter bed can be calculated. As the equations are designed to find N, which is the number of eggs in a centimetre of column, No is equivalent to the number of eggs detected at sample point A. Using this value the number of eggs that could be detected in each of the 77cm of sand can be calculated. An illustration of this from the data of the pilot study is shown in graph 6.2. The summation of the eggs detected in each centimetre of sand is equivalent to the number of eggs detectable by the methods employed. This can be compared to the actual number of eggs added (330000) to give a percentage recovery rate of the detection procedures during each run. The number of detectable eggs and the percentage recovery are shown in tables 6.5 and 6.6.



Graph 6.2:Bar chart of the number of eggs in each centimetre

of filter bed (data from pilot study using method two)



depth of sand (cm)

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effective sand size(mm)	0.3	0.4	0.5	0.6	0.7
Total eggs detectable	35372	65897	74545	95449	135703
% recovery range of	10.7	20.0	22.6	29.0	41.4
recovery(%)	7.6→17.6	19.2→20.9	18.5→26.6	22.6→42.0	31.8→53.5
Table 6.6:Total	l Numbers	of eggs de	etected usi	ing method	3

Table	6.5:1	Cotal	Number	of	eggs	detected	l usin	g Metl	hod 2	2	
	((calcu	lated	usi	ng the	e calcula	ated v	alues	of a	and	b)

effective sand size(mm)	0.3	0.4	0.5	0.6	0.7
Total eggs detectable	71484	87800	88530	151419	180827
% Recovery range of	21.7	26.6	27.0	46.2	55.2
recovery(%)	18.1→25.3	18.2→29.8	21.1→34.5	34.1→56.6	43.6→74.5

The two noticeable characteristics of these results are the generally low recovery rates and the increasing efficiency of detection with an increase in effective sand size. These general statements are subject to the interpretation of the range of recoveries obtained. Although the ranges overlap, sometimes considerably, the general trend is still well illustrated. Therefore it seems valid to use the mean recovery rate as the basis for any discussion. The low recovery rates were discussed in the chapter 5 with the efficiency results for the procedures shown in appendix B. The reason for the increase efficiency with increase effective sand size is not as clear. To test this phenomenon, known numbers of eggs were placed in samples of various sand sizes and stirred at various rates for various lengths of These results are shown in table 6.7. The experiments were time. done in the same glassware as that used for the data collection. The speeds are arbitrary values that increase with the higher number. These are used to illustrate the difference caused by speed of stirring. The results confirm that the efficiency of detection is improved with increased sand size. Efficiency is also increased with increased time and stirring speed.

parameter	variation				Control	
<pre>sand(mm) size</pre>	0.3-0.42	0.42-0.6	0.6-1.18		======================================	
mean (n=8) %recovery	43	49	66		minutes	
speed	5	6	7	8	sand size +0.6mm	
mean (n=8) %recovery	30	45	67	88	time 5 minutes	
time(min)	3	5	7	9	sand size	
mean (n=8) %recovery	48	53	68	83	speed 7	

TABLE 6.7:Tests for procedure efficiencies.

The mean percentage recovery rates shown in tables 6.5 and 6.6 can be used as correction factors for the results shown in tables 6.1 and 6.2. The corrected results shown in tables 6.8 and 6.9, are now directly comparable. The corrected values reverse the trend of the number of eggs detected at the first sampling point. This is a significant change in the results and due to the large range of recovery percentages possible for each effective sand size, some caution should be attached to these data. However, without the correction factor the results are not directly comparable and therefore it was considered that this analysis was valid.

Table 6.8:Corrected number of eggs at eachsampling point using method 2

depth of	Effe	ective sa	and size	(mm)	
Sample(cm	ı) 0.3	0.4	0.5	0.6	0.7
*******	=======	=======	======		======
1	215317	196281	168920	122482	98025
16	1791	2110	2668	3434	3955
31	859	1009	1531	1625	2052
46	526	615	1061	967	1303
61	361	419	803	642	912
76	264	305	639	456	677

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depth of Effective sand size (mm)							
sample(cm	ı) 0.3	0.4	0.5	0.6	0.7		
2222222	=======	=======	=====	======			
1	136664	130649	130729	82094	72398		
16	3040	3208	3336	4263	4503		
31	1266	1404	1966	1942	2091		
46	684	787	1384	1103	1200		
61	419	498	1058	699	766		
76	278	339	848	474	521		

Table 6.9:Corrected numbers of eggs at each sampling point using method 3

These values are shown graphically in graph 6.3. These show that more eggs are present at the first sampling point for the smaller effective sand size. However by the second sampling point the trend is reversed with the smaller effective sand sizes indicating lower numbers of eggs sampling point the present. After the second decrease was approximately parallel for all lines until the bottom of the media. The notable exception for both methods is the results for the 0.5mm effective sand size. The reason for this is not known but is The relative differences between attributed to experimental error. the results for each sand size at the lower sampling points are smaller than the relative differences at the top. To explain this a profile of the grain size of each sample point for each effective sand size was recorded. These results are shown in table 6.10.

Table 6.10:Mean diameter of sand grains at each sampling point for each effective sand size (µm,n=20)

depth of	Effe	ctive	sand s	ize (mm)			
sample(mm)	0.3	0.4	0.5	0.6	0.7		
			======	======	=====		
1	266	345	442	521	622		
16	542	620	656	768	797		
31	692	667	819	821	857		
46	706	718	882	879	901		
61	735	778	959	997	1015		
76	772	818	1036	1052	1079		

Graph 6.3:Corrected pattern of removal of helminth eggs with depth for various effective sand sizes



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There is an increase in sand size with depth because backwashing creates hydraulic regrading of the sand grains during fluidisation. It is interesting to note that the differences between the sand sizes at the lower sampling points for each effective sand size are smaller than the differences at the higher sampling points. This is a similar trend to the differences in the corrected results for each effective sand size. This apparent connection will be discussed.

The two other observations made on the effective sand size experiments were head loss and effluent egg detection. As was stated early the methods tried for egg detection in the effluent failed to give results until the method of the glass bottomed bucket was used. This was only applied to the 0.7mm effective sand size on rig B. The results for the effluent detection are shown in table 6.11.

Table 6.11:Eggs in the effluent of the 0.7mm sand size.

Run	1	2	3	4	5	average
number of eggs in effluent (13201)	2540	1210	680	380	412	1044

As can be seen, large numbers of eggs were detected in the effluent of the 0.7mm effective sand size. Although the method is a crude estimation of the number of eggs passing the filter, this shows that 0.3% of eggs can pass through a filter. This is equivalent to approximately 1 egg per litre of effluent which is the WHO guideline value. The procedure is crude and it is likely that it does not detect all the eggs passing through the filter. If this is the case the value shown in table 6.11 would imply that a 0.7mm effective sand size would not treat the effluent to the required standard if 250 or more eggs per litre are in the influent.

The head loss experiments showed no difference in head loss between the start and the finish of the run. As only water was used, the volume of eggs captured by the filter was not enough to create any detectable difference in the head loss.

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6.2.RESULTS FROM FLOW RATE EXPERIMENTS

The flow rate was varied to $2m^3/m^2/h$ and $6m^3/m^2/h$ while the effective sand size remained at 0.5mm. The results are shown in table 6.12.

Table 6.12:Results from varied flow rate.

depth of	Method 2		Method 3		Backwash		
<pre>sample(cm)</pre>	2m³/m²/h	6m³/m²/h	2m ³ /m ² /h	6m³/m²/h	2m³/m²/h	6m³/m²/h	
1	======================================	28623	= ==== ===============================	23890	======================================	======================================	
16	811	1067	860	1928	132	115	
31	187	462	333	680	66	54	
46	111	283	246	429	35	85	
61	74	202	134	319	59	60	
76	52	141	73	263	26	28	

The same general trend of egg removal is again obvious. For these results to be comparable to each other, and to the effective sand size results, the same procedure used to correct the results in the previous section will be applied here. The constant of 'a' and 'b' for the flow rate experiments are shown in table 6.13.

Table 6.13: Values of a and b for flow rates

Flow	w Method 2				Method 3		
rate	(m/h)	а	b	a x b	а	Ъ	a x b
=====	======	======		=======================================			
	2	1.484	0.348	0.516	1.398	0.328	0.459
	4	2.449	0.190	0.465	2.086	0.204	0.425
	6	1.429	0.304	0.435	0.931	0.374	0.348

The bold values are the values from the initial data collection using effective sand size 0.5 and an equivalent flow rate of 4m/h. Although the 'a' and 'b' values do not follow an easy pattern, which is illustrated by the straight lines in graph 6.4, the values for the product of 'ab' decrease with increasing flow rate. The trend shown by the 'ab' values suggests that increasing the flow rate to 6m/h is equivalent to increasing the effective sand size to 0.65mm and decreasing the flow rate to 2m/h is equivalent to decreasing the effective sand size to 0.45mm.





There is more evidence for the fact that an increased flow decreases the removal efficiency from the corrected results shown in table 6.15. These were obtained by using the same method as for the effective sand sizes. The correction factors are shown in table 6.14. Again the range of recovery percentages are noted but the means are discussed.

Table	6.14:1	「otal	numbe	ers o	of	egg	s de	etec	ted
	ເ	using	both	meth	lod	2	and	3	

	Method	12	Method 3		
	6m/h	2m/h	6m/h	2m/h	
Total eggs detectable	89507	80867	120325	80166	
% Recovery	27.2	24.5	36.7	24.3	
recovery(%)	17.0→34.5	19.9→31.7	30.8→56.0	15.3→35.6	

Table 6.15:Corrected numbers of eggs per centimetre columnat the sampling points using methods 2 and 3

Depth of	Method	2	Method	3
<pre>sample(cm)</pre>	6m/h	2m/h	6m/h	2m/h
==============		********	========	=======================================
1	105022	138650	64929	113764
16	3786	2837	4691	3529
31	1805	1038	2242	1520
46	1075	505	1312	837
61	713	283	849	520
76	504	173	585	347

A graphical illustration of these data is shown in graph 6.5. Graph 6.5a includes the results from the 0.5mm, 4m/h flow rate experiments. Although this graph approximately indicates that decrease flow increases removal, this trend is clearer when the 4m/h flow rate runs are removed as in graph 6.5b. Therefore, the corrected values for the flow rate imply that a lower flow rate will improve the efficiency removal of helminth eggs. As with the values at 1cm depth for the effective sand size the lower flow rate has a greater number of eggs detected at the 1cm level than the higher flow rate. The proportional removal in the lower regions of the bed are much closer for the different flow rates which reinforces the suggestion that the general removal efficiency is dependent on the removal in the first few centimetres.

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The head loss observations again showed no significance difference before and after the run. The effluent results, shown in table 6.16, reinforce the fact that the removal efficiency is improved with a decreased flow rate.

Flow rate	Run 1	2	3	4	5	average
====== 6m/h 2m/h	============ 104 0	====== 84 12	128 0	======= 12 0	======= 112 16	====== 88 6

Table 6.16:Eggs in effluent for flow rate experiments

6.3.RESULTS FROM THE MIXED MEDIA EXPERIMENTS

Experiments were carried out with mixed media to investigate the removal efficiencies of more advanced filters. The mixed media investigated were anthracite with sand (effective sand 0.5mm) and garnet with sand. Due to a failure with one of the pieces of apparatus the garnet with sand experiments were conducted with only one column. As previous duplicate runs have shown that there is no significant difference between the rigs it was considered valid to the accept the results from the single rig. The results from the mixed media experiments are shown in table 6.17.

depth of	Method	2	Metho	d 3	Backwash	
<pre>sample(cm)</pre>) A/S	S/G	A/S	S/G	A/S	S/G
================		=2=2=2=3	==========	=========		*******
1	31136	34692	24914	28120	1630	1415
16	5504	883	3057	1938	194	64
31	727	266	580	478	64	48
46	729	112	1426	244	103	26
61	171	213	347	352	54	40
76	86	0	156	0	35	0

Table 6.17: Results from the mixed media experiments

A/S = anthracite/sand S/G = garnet/sand

The boundary between the anthracite and sand medias was between the sampling points at a depth of 31 and 46cm. The boundary between the sand and garnet medias was between the sampling points at 46 and 61cm. The effect of the boundary is illustrated in graph 6.6. Graph 6.6b clearly shows an increase in the number of eggs present just after the boundary between medias for the two sets of experiments. This is consistent with established filtration theory which suggests that removal is more efficient at the boundary as the pore sizes are smaller. This is because the smaller particles of the lower media mix with the larger particles of the upper media. Although this occurs throughout the bed, the boundary is the main area where this occurs. This creates a level of substantially smaller pores.






This type of removal is not consistent with the equation developed for the removal from a sand only media. Therefore it is not valid to apply the equation as with the other sections. A correction factor can not be established for the same reason. The results from the mixed media will be discussed as they are presented in table 6.17. It is interesting to note that the sand/garnet mixed media had no detectable eggs in the effluent. This was the only regime at which total removal of Ascaris suum eggs was achieved.

The effluent results for the mixed media, shown in table 6.18, indicate as above that the sand/gannet dual media will remove all helminth eggs in 76cm of sand. These results also suggest that the anthracite sand dual media is an efficient remover of eggs.

mixed	Run					
media	1	2	3	4	5	average
A/S	28	32	40	0	0	20
S/G	0	0	0	0	0	0

Table 6.18:Eggs in effluent from the mixed media experiments

6.4. RESULTS FROM THE CONTROL EXPERIMENTS USING WASTEWATER

Control experiments were conducted for 12 hours with an effective sand size of 0.5mm and a flow rate of $4m^3/m^2/h$ using water for one set of experiments and effluents for the other. The effluent was a good final effluent from a conventional activated sludge treatment plant with a suspended solids concentration of 8mg/l. The reason for the control experiments was to establish if there was any difference between water and effluents in the removal of helminth eggs. These results are shown in table 6.19.

depth of	Method	2	Metl	nod 3	Bad	ckwash
<pre>sample(cm) </pre>	Water I	Effluent	Water	Effluent	Water	Effluent
1	33691	33742	30137	32797	2356	2155
16	634	790	1294	1484	85	69
31	279	286	454	449	26	55
46	172	193	334	298	8	46
61	148	134	254	167	31	28
76	95	95	162	138	18	16

Table 6.19:Results from the control experiments

These results can be made comparable using the method of calculating the constants of 'a' and 'b'. This is possible as the removal has the same general pattern as that which was established for the equation of the line in sections 6.1 and 6.2. The values for 'a' and 'b' are shown in table 6.20.

Table 6.20:Values for a and b for control experiments

Filtrate	Method 2 a	2 b	axb	Method 3 a	З Ъ	axb
water	2.054	0.242	0.497	1.374	0.309	0.425
effluent	1.788	0.281	0.493	1.159	0.366	0.424

Although the values for 'a' and 'b' are different for the water and effluent experiments, when the product of 'ab' is calculated the values are all but identical. The differences in the 'a' and 'b' values are illustrated by the straight lines shown in graph 6.7.

Graph 6.7:Straight lines described by the constants 'a' and 'b' for the control experiments



The calculated correction factors are shown in table 6.21.

Table 6.21:Total number of eggs in column using methods 2 and 3

	Metho	od 2	Method 3		
	water	effluent	water	effluent	
#22223222 4 2;					
Total eggs	70580	78918	99375	113247	
%Recovery	21.4	23.9	30.2	34.4	
range of	16 7-226 /	10 / 20 5	22 0.220 6	22 5.3/2 0	
recovery(%)	10./720.4	17.4720.J	22.0730.0	22•J742•0	

From these correction factors, the corrected values can be calculated. These values are directly comparable and are shown in table 6.22.

Table 6.22:Corrected number of eggs at sampling points using methods 2 and 3

depth of sample(cm)	Method water	2 effluent	Method 3 water	effluent
1	157074	140718	99549	95234
16	3005	3277	4160	4200
31	1459	1462	1947	1706
46	900	843	1151	896
61	620	548	760	534
76	448	375	525	334

The results are very similar and suggest that there is no difference in the removal efficiency between the water and effluent. This is emphasised by the similarity of the corrected lines of removal shown in graph 6.8. These results will be discussed later.

There was a large difference in the head loss of the water and effluent experiments. These results are shown in table 6.23.

depth in filter(cm)	water Initial	Final	effluent Initial	Final
0	0	0	0	0
7.5	6	6	6	59
22.5	13	13	13	78
37.5	23	22	21	83
52.5	32	31	29	85
67.5	42	41	37	87

Table 6.23:Mean head loss of control experiments (cm, n=5)



Graph 6.8:Corrected pattern of removal of helminth eggs

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The head loss for the water experiments again show no difference between the initial and the final readings. The effluent head loss shows that the vast majority of head loss occurs in the first few centimetres. This is consistent with the fact that the majority removal of suspended solids will occur in the first few centimetres. The head loss in the lower regions of the bed is proportionally lower than the upper regions of the bed. This implies that the filter was not at the end of its useful run due to clogging. Whilst suspended solids were being trapped by the filter efficiently there was no difference in the corrected number of eggs between the two rigs. It is suggested that, while the filter is still operating efficiently, the presence of organic matter will not affect the removal of helminth eggs under the conditions of the experiments conducted in this research. This is also implied by the effluent results shown in table 6.24. These results show that there is no difference in the number of eggs detected in the filtrate of the water and effluent runs.

	Table	6.24:Eggs	in	effluent	of	control	ex	periments
--	-------	-----------	----	----------	----	---------	----	-----------

control	Run 1	2	3	4	5	average
water	24	0	80	36	24	33
effluent	16	12	72	40	8	30

Ultimately as a filter becomes clogged, the passage of eggs will be affected by the presence of organic matter. This stage of filtration was not reached with the experiments undertaken in this research.

6.5.RELIABILITY OF THE RESULTS

To establish the reliability of the results the theory of linear regression needs to be applied to the straight lines developed in section 5.3. Although the lines referred to in section 5.3. are expressed as F = Ax + b, the theory of linear regression is based on the line $y = \alpha + \beta x$. Although these lines are effectively the same, the theory here will be discussed referring to the line $y = \alpha + \beta x$.

The difference between the actual data points and the points on the line of best fit is referred to as the errors, ε_i , which is an indication of the 'goodness of fit' of the line. If the object is to predict y from x it is reasonable to model the behaviour of y in the form:-

$$y_i = \alpha + \beta x_i + \varepsilon_i$$
 (1)

where y_i and x_i are observed x and y, α and β are parameters and ε_i is the random error. The values for α and β have been established using the methodology described in section 5.3.3.

To calculate the random error the value y_i^{1} , calculated from the line of best fit, needs to be compared with the corresponding data point, y_i , in the form:-

$$e_i = y_i - y_i^1$$
 for $i = 1, 2, 3...n.$ (2)

The magnitude of Σe_i^2 can be used as the goodness of fit. This is called the sum of the squared residual and will be small for a good fitting line and large for a poorly fitting line. The value of Σe_i^2 is related to the total sum of squares of the y values, ie $\Sigma(y_i - y)^2$. Appendix F shows the proof that the total sum of squares may be partitioned into two components. These are the the sum of the squared residuals (Σe_i^2) and the regression sum of squares $b^2 \Sigma(x_i - x)^2$. The relationship can be expressed by:-

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$$\Sigma(y_{i}-y)^{2} = \Sigma e_{i}^{2} + b^{2} \Sigma(x_{i}-x)^{2}$$
(3)

The value of b is the calculated value of the gradient. It is only an estimate of β , but as β can not be calculated exactly, the term b is used to show it is an estimation of the true value. The residual sum of squares and the regression sum of squares measure different sources of variation. The first measures the variation about the regression line where as the second measures the variation due to the regression. The regression about the line is the distance from the actual data points to the line of best fit and the variation due to the regression is the distance between the points on the line and imaginary points created by drawing a perpendicular line from the data points to the line of best fit.

A comparison of the relative sizes of these components provides a test of the significance of the regression. If Σe_i^2 is small compared to a large $b^2 \Sigma (x_i - x)^2$ then it is possible to state that the line of regression is significant. The values for Σe_i^2 and $b^2 \Sigma (x_i - x)^2$ are given in tables 6.25 and 6.26.

sand	parame	ters (α and β)	variations		
(mm)	b	А	Σe _i ²	$b^{2}\Sigma(x_{i}-x)^{2}$	
pilot	0.207	0.807	0.0009922	0.0646398	
0.3	0.212 0.228	0.968 0.880	0.0031421 0.0018053	0.0704233 0.0786229	
0.5	0.190	0.896	0.0011446	0.0546735	
0.6	0.287	0.477	0.0009785	0.1249961	
0.7	0.281	0.386	0.0046627	0.1197698	

Table 6.25:Reliability of the regressions from method 2.

sand	parame	eters (α and β)	variation	S
(mm)	b	А	Σe _i ²	$b^{2}\Sigma(x_{i}-x)^{2}$
pilot	0.189	0.622	0.0103281	0.0539410
0.3 0.4 0.5 0.6 0.7	0.313 0.304 0.204 0.357 0.369	0.468 0.466 0.735 0.103 0.001	0.0019507 0.0015750 0.0001865 0.0021842 0.0038728	0.1484190 0.1401936 0.0627270 0.1924421 0.2057822

Table 6.26:Reliability of the regressions from method 3.

Tables 6.25 and 6.26 show that Σe_i^2 is small compared with $b^2 \Sigma (x_i^-x)^2$ and this is an indication that the regressions are significantly reliable. There are, however, some assumptions to this theory. These are:-

1) $\epsilon_i \sim N(0,\sigma^2)$, which means the random error is normally distributed about the point $(0,\sigma^2)$.

2) σ^2 is constant, which means the the standard deviation of the points is the same through out the regression.

3) ε_i and ε_j are independent for $i \neq j$, which implies that there is no pattern in the distribution of the random error.

The theory of linear regression allows for two theoretical results. The first is an estimate for the standard deviation σ^2 with (n-2) degrees of freedom. This is:-

$$\sum_{i=1}^{2} (n-2)$$
 (4)

The n-2 degrees of freedom is because there are two constraints on y i due to α and $\beta.$

The second theoretical result is the estimation of σ^2 with one degree of freedom. This is:-

$$\frac{b^{2}\Sigma(x_{i}-x)^{2}}{\Sigma e_{i}^{2}/(n-2)}$$
(5)

This is equivalent to an F value for the significance of the fit with $F_{1,(n-2)}$ degrees of freedom. The F values for the various lines are shown in table 6.27.

The 95% significance level of $F_{1,4}$ degrees of freedom is 7.71 (White et al 1979). All the values are much greater than this value. This means that the null hypothesis that there is no significance to the regressions can be rejected. Therefore there is a significant relationship in the regressions of the lines.

Table 6.27:F-values for the regression analysis from methods 2 and 3.

F-value method 2	F-value method 3
260.6	20.8
89.7 174 2	304.3
191.1	1345.2
510.9 104 1	352.4 212 5
	F-value method 2 260.6

From these results it is possible to obtain confidence limits for the parameters α and β . The variability of α and β can be estimated by V(a) and V(b) respectively where V(a) and V(b) are given by:-

1)
$$V(b) = \frac{\sigma^2}{\Sigma(x_i - x)^2}$$
 (5)

for $100(1-\alpha)$ % confidence interval for β is given by:-

$$b \pm t_{n-2, \alpha/2} \sqrt{\left(\frac{s^2}{\Sigma(x_i - x)^2}\right)}$$
 (6)

where $s^2 = \Sigma e_i^2/(n-2)$, α is the level of chosen significance and t is the value from t tables with n-2 degrees of freedom at the significance level of $\alpha/2$.

2)
$$V(a) = V(y) + x^2 V(b)$$

$$\Rightarrow \qquad = \frac{\sigma^2}{n} + \frac{x^2 \sigma^2}{\Sigma(x_1 - x)^2}$$

 $\Rightarrow \qquad = \sigma \left(\frac{1}{n} + \frac{x^2}{\Sigma(x_i - x)^2} \right)$

Thus $100(1-\alpha)$ % confidence limits for α is:-

$$a \pm t_{n-2, \alpha/2} \sqrt{\left(\frac{s^2}{n} \left(\frac{1}{n} + \frac{x^2}{\Sigma(x_1 - x)^2} \right) \right)}$$
(7)

The confidence intervals for A, b and the resulting a and ab values are shown in tables 6.28 and 6.29. These values are calculated with the $t_{3,(95/2)}$ value of 3.18.

Table 6.28:Variences for the constant of the regression lines and the equation of removal constants using method 2.

sand size	variation	of const	tant:-	
(mm)	V(b)	V(A)	V(a)	V(ab)
pilot	0.047	0.176	1.192	0.056
0.3	0.084	0.312	1.367	0.114
0.4	0.063	0.237	1.267	0.080
0.5	0.050	0.189	1.208	0.061
0.6	0.047	0.174	1.191	0.056
0.7	0.101	0.378	1.460	0.148

sand size	variation	of const	tant:-	
(mm)	V(b)	V(A)	V(a)	V(ab)
pilot	0.152	0.567	1.762	0.267
0.3	0.066	0.246	1.279	0.084
0.4	0.059	0.176	1.192	0.071
0.5	0.020	0.076	1.079	0.022
0.6	0.070	0.261	1.298	0.091
0.7	0.093	0.347	1.415	0.131

Table 6.29:Variences for the constant of the regression lines and the equation of removal constants using method 3.

The values, when added or subtracted to their means given in tables 6.25 and 6.26, give a variation that will include all the other values of the other lines. The significance between the lines can be calculated. This can be done by comparing the gradients of the lines with the formula (Bailey 1984):-

$$d = b_{2} - b_{1}$$

$$\sqrt[4]{(x_{i1} - x_{1})^{2} + (x_{i2} - x_{2})^{2}} \\ \sqrt[4]{(z_{e_{11}}^{2}/n - 2} \sum_{i_{2}} \sum_{j_{2}} \sum_{j_{1}} \sum_{j_{2}} \sum_{j_{2}} \sum_{j_{1}} \sum_{j_{2}} \sum_{j_{1}} \sum_{j_{2}} \sum_{j_{1}} \sum_{j_{2}} \sum_{$$

where d is equivalent to a z score which can be interpretated into a percentage significance for the differences between the lines. The d scores and the percentage significant difference between the lines from the data collection are shown in table 6.30.

Table 6.30:The significance difference between the regression lines for methods 2 and 3

difference	Method 2		Method 3				
between lines	d-value	percentage significance	d-value	percentage significance			
0.3-0.4	0.402	66.3	0.108	57.2			
0.4-0.5	0.316	62.6	8.339	99.9			
0.5-0.6	1.000	84.1	8.882	99.9			
0.6-0.7	0.500	69.2	0.121	54.8			
0.3-0.7	1.768	96.1	4.761	99.9			

The significance level for the results to be determined as reliable is 95%. Between the curves only 0.4-0.5 and 0.5-0.6 differences using method three could be described as reliable. Both methods show that there is a significant difference between the 0.3 and 0.7 curves. This implies that the results indicating that sand size has a profound effect can be taken as reliable. The differences that are inconclusive, because of the lower percentage significance levels, still indicate that there is an effect due to sand size. This can be tested for by using a χ^2 test on the means and the individual results from the runs. The χ^2 values for the means are shown in table 6.31.

Table 6.31:Chi squared values for the differences between the results of different effective sand size runs

difference between	X² value method 2	X² value method 3
===============	=========================	===========
0.3-0.4	17.71	14.58
0.4-0.5	189.14	184.05
0.5-0.6	165.14	381.20
0.6-0.7	282.99	203.05

The 95% significance level with 5 degrees of freedom is 11.07. All the χ^2 value are higher than this which means that the null hypothesis, that the distribution of eggs in the sand filters between the different runs is the same, can be rejected. These show that the from the different sand sizes derive from different results populations and are therefore significantly different. Using a χ^2 test on the mean is open to error as only six points are available for The test needs at least five points but the greater the analysis. number of points the more reliable the significance. If the χ^2 test is carried out with the raw data 60 points are available. These results also show that the results from the different effective sand sizes derive from different populations.

As the χ^2 test shows that the results are significantly different it suggests that the significant differences between the lines would be reliable if more results were available. Time did not permit more experimental runs and the equipment available prevented more than two rigs being used.

A similar procedure can be undertaken for the results from the change in flow rate. These show that the regressions from the data are significant with F-values with $F_{1,4}$ degrees of freedom of 1171 and 104 for the 6m/h and 2m/h runs respectively. Caution is required when evaluating these results as significance is difficult to justify when only two data points are available. The d scores were 0.378 and 0.346 for the two methods which indicates a 65% reliability, but the argument that these results would be significant if more duplicates were taken still holds.

The control experiments can be shown to have no significant difference through the method of matched pairs. With t-values of 1.319 and 0.925 for the two methods, no significant difference is present. The easiest way of establishing that there is no difference between the control experiment results is by looking at the corrected values in table 6.22. These results, and the raw results in table 6.19, are very similar. From this it can be said that the presence of suspended solids does not significantly affect the removal of Ascaris eggs while the filter is running efficiently.

CHAPTER 7: DISCUSSION

This section will attempt to explain the observations highlighted in chapter six. The aim is to illustrate that the calculated lines of removal closely resemble the actual process of removal in the filter. To achieve this the mode of egg removal will be discussed with reference to the mechanical removal mechanisms of filtration and the physical properties of the <u>A.suum</u> egg wall. To validate the work the control experiments will be discussed in order to indicate that applying eggs in water will not adversely affect the results.

7.1.CONTROL EXPERIMENTS

As the data were obtained with the use of tap water as the suspending medium, it was necessary to establish whether the use of effluents would affect the removal of Ascaris eggs in the filter. The logistical problems of supplying effluents to the laboratory made their consistent use impracticable. It was also impractical to maintain the effluent experiments for a period of 24 hours. Therefore a 12 hour run was used. Twelve hour experiments using tap water were required to directly compare the results. The total number of eggs added was maintained at 330000, giving a concentration of eggs added during the control experiments of 500 eggs/l. A reduction in this number would make detection difficult.

The results shown in section 6.4 indicated that the removal efficiencies for the control experiments are very similar. It is interesting to observe that the constants of the equations when multiplied together are all but identical. Although the values of 'a' and 'b' vary individually by up to 15%, the compound value would suggest that there is no significant difference between the two sets of results. This is confirmed by the work shown in section 6.5 on the reliability of the control experiments.

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The compound 'ab' value for the control experiments is similar to the 24 hour 0.5mm effective size value, especially for method 3. This would suggest that length of filter run has little effect on egg removal which in turn implies that eggs penetrate to the depth of the filter from the start of a run. This is consistent with Boller (1984) who showed that the efficiency of a filter is lower in the initial parts of the run.

From the results of the control experiments it is possible to state that the removal efficiency of Ascaris eggs in a rapid sand filter is not affected by the presence of suspended solids while the filter is clarifying effluent to an extent acceptable by an operational filter. Suspended solids will clog a sand filter but do not otherwise interfere with the removal of Ascaris eggs. If a clogged filter is backwashed, the majority of particles, including helminth eggs will be removed. Thus the experimental results obtained using just water can be accepted as realistic for a filter which does not need backwashing.

7.2.MODE OF EGG REMOVAL

From the results in section 6 it is clear that Ascaris eggs do pass through filters even at the smallest sand sizes. Therefore the processes by which they are removed, or conversely allowed to continue, are important if the movement of eggs within a filter is to be understood. The determining transport mechanisms during filtration were discussed in section 4.1. These processes will now be directly related to Ascaris eggs.

7.2.1.Interception

Interception is the process by which an egg collides with, and sticks to, a sand grain as the flow line the egg is in is closer to the grain than the diameter of the egg. The effect of interception is determined by the equation:-

$$I = \frac{d}{D}$$
(1)

where d is the particle size and D is the grain size. For example, when d is 40µm and the grain size is 0.2mm, I is equal to 0.25. When the grain size is 1 mm I is equal to 0.05. These values are dimensionless but if I is greater than 0.1 it is suggested that interception is a determining removal process (Ives 1975). An Ascaris egg, with a diameter of 40µm, achieves this value when the sand grain is 0.8mm or smaller. From the distribution of sand grain size shown in table 6.10, this grain size is reached at varying points in the filter beds. For 0.3mm effective sand size all the sand is smaller than 0.8mm. As the other effective sand sizes increase the depth of bed with grain size smaller than 0.8mm decreases , until the 0.7mm effective sand size where the grains are smaller than 0.8mm for only the first 16cm. Therefore, interception is a major removal mechanism for at least part of the filter for all the effective sand sizes but for varying depths. The length of bed during which this process is a determining mechanism appears to be influential in fulfilling the observed trend of a lower efficiency for larger effective sand sizes.

7.2.2.Inertia

The Inertia of an egg in a filter (E) can be calculated by the equation:-

$$E = \rho_{S} d^{2}U$$
(2)
18µD

where $\rho_{\rm S}$ is the density of the egg, U is the velocity of the water and μ is the viscosity of the liquid. When calculated for the criteria of the experiments for a sand size of 0.5mm, E = 3.6 x 10⁻³. This value is very small which indicates that inertia is not a determining mechanism.

7.2.3.Sedimentation

Sedimentation is the process where the eggs settle on the sand grains within the pores. The effect of sedimentation can be determined by calculating the ratio between the settling velocity of the egg in solution and the velocity of the flow, ie v_s/v_i . v_s can be estimated by calculated by Stokes Law:-

$$v_{s} = g(\rho_{s} - \rho)d^{2}$$
(3)
18u

where g is gravity and ρ is the density of the liquid. This is only an estimation of the settling velocity as Stokes Law is only truly valid for a sphere in a column of liquid of a diameter 100 times the diameter of the sphere. For Ascaris eggs this is not fulfilled. It is however the best estimation available and for an Ascaris egg v_s is equal to 0.14mm/s. v_i is equal to 1.1 mm/s in the experiments so the ratio v_s/v_i is 0.09. This value suggests that sedimentation is not a significant process in the removal of Ascaris eggs. However, as explained in section 4.1, the true v_i decreases as the particle approaches the sand grain. For example, the velocity of the liquid parallel to the grain surface at a distance of 20µm from the grain is

equal to $0.036v_i$. In other orientations of flow, v_i is drastically reduced close to the sand grain. In case of parallel flow, v_s is greater than v_i when the egg is close to the sand grain. This makes sedimentation an important mechanism for the removal of Ascaris eggs in a filter when eggs are close to the grain. As the average pore size for 0.5mm diameter sand grains is 100µm, then an Ascaris egg will never be more than 30µm from a sand grain. Therefore sedimentation is likely to be another determining mechanism of removal.

7.2.4.Diffusion

Diffusion is the random movement of a particle in a filter. The larger the diffusion rate the greater the effect on removal. Diffusion (B) can be calculated by:-

$$B = \underline{kT}$$
(4)
3 $\pi\mu dUD$

where k is Boltzmann's constant (1.38×10^{-23}) and T is absolute temperature. For Ascaris eggs diffusion has a value of 6.36×10^{-2} . This is a value that suggests diffusion has a negligible effect on the removal of Ascaris eggs. This is of no surprise as an egg is a large particle in terms of filtration and diffusion is only important for sub-micronic particles.

From the above calculations it is possible to state that the determining mechanisms involved with the removal of Ascaris eggs are interception and sedimentation.

7.3. THE EFFECT OF THE EGG WALL

The physical properties of the egg wall may have some effect on whether removal is likely to be enhanced or reduced. The following properties of the wall were investigated.

7.3.1.Stickiness of an Ascaris egg

It had been reported (Kennedy personnal communication) that an <u>Ascaris</u> <u>suum</u> egg wall has a sticky albuminoid layer. The extent of the stickiness is important to establish if an egg is able to resist the shear pressures present in a filter. Only in this case will a sticky egg significantly aid removal.

the egg The easiest way to investigate wall was through electronmicroscopy. Whole eggs were filtered out of a solution using a glass fibre filter for photographs to be taken of the eggs on the filter. From the photographs (plates 7.1a and 7.1b), there are two reasons for assuming that the eggs are not sticky. The first is that the eggs seem to be evenly distributed on the filter. If they were sticky the chances are that they would bunch together and appear on the filter in this way. The second reason is that the close up of the egg wall shows minimum detritus. If the surface was sticky it would be expected that the surface would be covered with greater amounts of microscopic detritus. This does not seem to be the case, so again the eggs seem to be non-sticky.

These results need to be treated with caution. During the preparation of the eggs for electronmicroscopy the eggs were coated to improve vision. This coating may remove any sticky layer and therefore any impurities would be removed with it. The eggs are also present in a vacuum and are therefore likely to be turgid which may well effect the property of the wall. Caution too is required when considering egg extraction from the female worm. This procedure may also remove any sticky layer. Plate 7.1:Ascaris suum eggs shown by electron-microscopy

plate 7.1a:Collection of eggs on a glass fibre filter
 (Bar = 25µm)

plate 7.1b:Two Ascaris suum eggs revealing matter attached to the surface of the wall (Bar = 10µm)

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The evidence indicates that the egg wall is smooth and has no property of stickiness. Therefore, the egg wall will have no effect on the removal mechanisms due to stickiness.

7.3.2.Electrical charge

It has been well established that all organic particles have an electrical surface charge. This is created by the diffuse double layer in the ionic atmosphere surrounding each ion. Clean sand grains have a small negative charge of 5-10 mv (Huisman and Wood 1974). If an egg has a charge of the same sign, some repulsion will occur which will hinder the efficiency of removal. With this in mind the surface charge of an Ascaris egg was investigated. The technique used to discover the charge on the surface of a particle is called electrophoresis.

The technique of electrophoresis is a very complex procedure if all the errors are to be removed. There are complex, expensive items of glassware that will reduce errors to a minimum in the experimental chamber. However, as only an indication of charge was required in this study, a much simpler technique was used which would give an adequate estimation of the charge.

Using a MacMasters slide, an electrical potential was applied across one of the chambers by sliding steel electrodes into the chamber. A suspension of eggs in chlorobenzene was placed in the chamber. A known electrical current was applied and the speed of movement of an egg was timed by observing the egg with a graticule. Chlorobenzene was used as the solute as it has a known dielectric constant (5.8) and has a relative density of 1.106. This is the same density as an egg so the eggs would not move out of the field of view while being timed. The known dielectric constant is important for the calculation of the charge. It would be impractical both through time and equipment to try and establish this in the laboratory.

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The applied current was one volt and the mean movement of 10 eggs was 125 μ m in 25 seconds. This movement needs to be converted into units of (μ /s)/(abvolt/cm) which equates to 300 (μ /s)/(v/cm). The one volt was applied over a distance of 2.5 cm so 125 μ m in 25 seconds is equivalent to 2 (μ /s)/(v/cm) or 1/150 (μ /s)/(abvolt/cm). This can then be placed in the Helmholtz-Smolucchowski equation:-

$$\mu = \underline{\zeta d}$$
(5)
4 \pi \mu

where μ = electrophorectic mobility (motion of a particle in solution caused by an externally applied electrical field)

ζ = potential difference between the surface and solution (surface charge)

d = dielectric constant

and η = the viscosity of the liquid

There are several assumptions of the equation:-

1) the product of the Debye-Huckel constant, k, and the radius of curvature of the particle surface is large (>10 μ m)

2) particle surface is non-conducting

3) conductivity, dielectric constant and viscosity have the same values within the double layer as within the bulk of the liquid.
 4) the applied electrical field, although distorted by the presence of the particle can be simply added to the field of the double layer
 5) the liquid at the particle surface has the same velocity of the particle and the velocity gradient begins at the surface.

By putting the known values into equation (5) achieves a value for ζ of 14.4. This is equivalent to a surface charge of approximately 14 mV (Bier 1959). As the eggs headed towards the positive electrode the surface charge on the eggs is -14mV. Due to errors in experimentation described above, this can only be an approximation of the surface charge. This is within the range expected for such a particle.

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Although this is a negative charge and is the same sign as the charge on a clean sand grain, the small value of the charge indicates that it will have little effect on either transport or attachment mechanisms. Any repulsion that may occur between an egg and a sand grain will be negligible compared to the other forces on an egg in a filter. As the electrical charge seems to have a negligible effect, it is likely that the main mechanism for the attachment of helminth eggs to the sand grains will be settling on the upper surfaces.

7.3.4.Malleability of an egg

As an egg seems to have the potential to move through a filter it was suggested that the egg may be malleable to enable it to move through the pores. This was tested in two ways. The first was by using sintered bronze filters and the second was a direct attempt to squeeze an egg using micromanipulaters.

Sintered bronze filters are made by fusing together bronze balls of a known size. The idea is that this will give an accurate pore size. The size of the three filters tried were $40-80\mu m$, $30-40\mu m$ and $15-20\mu m$. The filters were sealed into porcelain buchner funnel and eggs in a solution of a known concentration were added with the pressure of a one meter head. The eggs were then allowed to flow through the filter under the influence of gravity. The number of eggs in the filtrate was then calculated and expressed as a percentage of the number of eggs in the influent.

For the 40-80 μ m filter 50% of eggs passed into the filtrate. For the 30-40 μ m filter 5% of eggs passed. No eggs passed through the 15-20 μ m filters. It is not surprising that no eggs pass the very small filter. It is a surprise that eggs of an average size of 40 μ m were able to pass through the 30-40 μ m filter. This would suggest that some eggs are flexible enough to pass through pores smaller than themselves. It is impossible to state this categorically. Although it was not noticed, there is a good chance that the eggs that passed were the smaller eggs passing the through larger pores.

The work carried out with the micromanipulaters was fraught with experimental problems. First there was the problem of obtaining an instrument small enough and blunt enough to push an egg against a wall. Secondly there was the problem of obtaining a flat wall to push the egg against and which did not deflect the light of the microscope in such a way that observation was difficult. Several combinations were tried but due to the lack of time it was impossible to persevere with the technique. From the observations of these tests it is believed that the eggs are not malleable. They broke when too much pressure was applied. This is a purely qualitative observation as no photographs were possible due to the diffraction of the light and there was no method for the quantification of the force required to break the eggs.

The fact that the eggs did not seem to be malleable is consistent with the literature on the constituents of an egg wall. It has been well established that the wall of Ascaris eggs are proteinacous and can protect the egg against great environmental harshness (Magat et al 1972, Spencer and Monroe 1982, Feachem et al 1983). Wall protein, such as keratin, has a rigid structure, which in turn makes the egg wall rigid. It is the presence of lipids that enable a wall such as an Ascaris egg wall to be flexible. There is no account of lipids being present in the Ascaris egg wall. This would indicate that Ascaris eggs are unlikely to change shape in order to pass through a pore of smaller size.

The smallest sand size present in the filters was 0.2mm which corresponds to a pore size of approximately 40μ m. It was illustrated by the reuslts from the mid sized sintered bronze filter that some eggs pass this size of pore. Therefore, the presence of eggs in the lower regions of the filter bed is possible without the eggs being malleable.

This piece of work is only applicable to <u>Ascaris suum</u> eggs. Other helminth species may well have the ability to pass pore sizes smaller than themselves although this is thought to be unlikely

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The equations illustrated in section 6 were developed in order to resemble the pattern of removal of helminth eggs during filtration. The removal described by the equations is consistent with established filtration theory. If a filter is considered in sections, the finer particles will rise to the upper surface during backwashing. As explained above, this means there is a greater percentage removal in the upper than the lower sections. This is illustrated by the theoretical numbers shown in Table 7.1.

section	%efficiency of removal	concentration in flow	retained deposit
======	=======================================		===========
1	65	1000	650
2	60	350	210
3	55	140	77
4	50	63	32
5	45	31	14
6	40	17	7
7	35	10	4
8	30	6	2
· 9	25	4	1
10	20	3	1

Table 7.1: Theoretical removal of solids in a rapid sand filter.

Source: Ives (1975)

As the efficiency decreases moving down the filter the retained deposit in the lower regions of the bed is much lower than that in the upper regions. The retained deposit column is directly comparable to the data collected from the experiments on egg removal. The equivalent data to table 7.1 for the removal of Ascaris eggs in a rapid sand filter is illustrated in table 7.2.

Table 7.2a:Removal of Ascaris eggs in a rapid sand filter (Method 2)

Effective sand size (mm)

	0.1	3	0.4	0.4		0.5		5	0.7	
depth	egg	eggs in	egg	eggs in	egg	eggs in	egg	eggs in	egg	eggs in
(cm)	conc	section	conc	section	conc	section	conc	section	conc	section
	-=2==:						=====		=====	
0-15	250	288891	250	281817	250	253554	250	253857	250	230404
16-30	31	18914	37	22255	58	30531	58	36137	75	43355
31–45	17	10229	20	11984	35	19275	31	19117	43	24852
46-60	9	6611	11	7703	20	13944	17	11987	24	16537
61-76	4	4944	5	5727	10	11448	8	8673	11	12575
eggs i	.n									
efflue	ent O.	31	0.	39	0.	95	0.	93	1.	74
%Remov	al 99	.88	99.	84	99.	62	99.	63	99.	30

egg conc = concentration of eggs per litre entering the section eggs in section = number of eggs caught during the 22 hour run

Table 7.2b:Removal of Ascaris eggs in a rapid sand filter (Metod 3)

Effective sand size (mm)

	0.1	3	0.	4	0.5	5	0.0	5	0.	7
depth	egg	eggs in	egg	eggs in	egg	eggs in	egg	eggs in	egg	eggs in
(cm)	conc	section	conc	section	conc	section	conc	section	conc	section
======	=====		=====		=====		=====	.======:	=====;	-======
0–15	250	271089	250	264271	250	230835	250	238825	250	231459
16-30	45	36297	50	32649	75	38668	69	44245	75	47154
31-45	22	14282	25	16092	46	24936	36	22401	39	24246
46-60	11	8185	13	9548	27	18275	19	13393	21	14624
61–76	5	5477	6	6584	13	15145	8	9231	9	10139
eggs i	n									
efflue	ent O.	51	0.	. 65	1.	62	1.	44	1.	80
%Remov	al 99	.80	99.	.74	99.	35	99.	42	99.	28

egg conc = concentration of eggs per litre entering the section eggs in section = number of eggs caught during the 22 hour run The lowest removal efficiency indicated here is 99.28%. This figure shows better removal than has been suggested by Schwartzbrod et al (1989) which was 98.8%. Although no operational criteria are given with this figure, the values are close enough to indicate good agreement.

The removal pattern shown in table 7.2 is very similar to the theoretical pattern in table 7.1. The pattern of removal is illustrated in graph 7.1 where the number of eggs/l entering each section is plotted against the depth of the top of each section. Although the values for the number of eggs leaving the filter are difficult to distinguish, the trend that fewer eggs enter the corresponding depths of the smaller effective sand sizes is well illustrated. The decrease for all the curves is exponential which implies that 100% removal of eggs is theoretically impossible. However if the sand bed was deep enough, a practical removal of 100% of the eggs is possible. The equations calculated in chapter 6 take this into account and show a close resemblance to the actual removal.

The accuracy of the line of removal is difficult to assess. One way of achieving this is by calculating the number of eggs deposited at each sampling point using the equation and comparing these results to the actual number detected. Using this method the difference between calculated and observed values can be assessed by applying the matched pairs method to see if their was any significant difference (table 7.3).

The t-value for the 95% significance level is less than 2.72. These results show there is no significant difference between the calculated and observed values. Therefore, it is possible to say that the lines of removal illustrate an accurate representation of the overall process of helminth egg reduction.

<u>Graph 7.1:Number of eggs/l entering each section of a</u> filter bed for various effective sand sizes.



Table	7.3:Matched	pairs	analysis	for	calculated	and	observed	data
		-	-					

sand size	0.3	0.4	0.5	0.6	0.7	0.5	0.5	0.5	0.5
flow rate	4	4	4	4	4	6	2	4	4
time (h)	22	22	22	22	22	22	22	12	12
suspension	water	water	water	water	water	water	water	water	effluent
	======						=====	======	
Method 2									
t-value	0.52	0.84	0.78	0.75	0.74	1.85	0.35	1.29	0.97
Method 3									
t-value	1.18	1.33	1.85	1.11	0.21	0.51	1.62	0.62	0.71

Having established that the equations of the lines are acceptable as an accurate representation of the process of removal in a rapid sand filter, the pattern of removal can be discussed. All the results indicate that the vast majority of removal occurs in the first centimetre of the sand bed. This suggests that an important removal mechanism is the eggs settling on to the first layer of the sand grains. Assuming that when an egg touches a grain it will be intercepted and that the eggs are evenly distributed within the influent, settling on the initial surface layer will account for approximately 75% of the total number of eggs present in the influent. Therefore only 25% of the eggs will pass the first layer of sand grains. Although not measured directly, this is assumed to be the major reason why the first centimetre of column gives results of large numbers of eggs. As there is less surface area available on the surface of the beds with larger effective sand sizes, the number of eggs trapped in this region decreases as the effective sand size increases. Of the eggs that do pass the first layer of sand grains the percentage removal reduces as the eggs progress. This is due to the changes in efficiency of the determining removal mechanisms as discussed in section 7.2.

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Once below the first sampling point, the results show that the number of eggs present increase as effective sand size increases at corresponding sampling depths. This may be emphasised by the fact that more eggs are available for detection as fewer eggs are removed in the initial stages of the larger effective sand sizes. This does however, imply that more eggs will pass through a filter with a greater effective sand size. This trend is highlighted by the calculated results for the number of eggs in the effluent shown in table 7.2. Although the results for the 0.5mm effective sand size are high the is still acceptable. This is illustrated by the graphical trend representation of the proportion of eggs caught in each section of filter bed (Graph 7.2, data from table 7.2). Graph 7.2 not only illustrates the greater number of eggs caught in the higher regions of bed but also shows the effect of effective sand size on each section. As stated before, the highest region has fewer eggs captured as the effective sand size increases. The other four regions illustrated show an increase in the number of eggs captured as effective sand size increases. It also interesting to note that the gradient of increase of eggs captured for the four lower regions decreases as one gets lower in the bed. This trend is consistent with the profile of the grain size throughout the bed as illustrated in table 6.10.

The two methods give different results because they sample different variables. Method 2 measures the total number of eggs at a sampling point which includes the eggs that are not attached and have the ability to move further in the column. Method 3 only measures the eggs that have been caught at the sampling point. This difference in detection is why method 3 indicates slightly lower removal efficiencies than method 2. The efficiencies of Method 3 are more likely to be an accurate representation of the removal as only the eggs which can no longer penetrate further into the bed are measured. The methodology of method 3 was also theoretically more accurate than method two as a standard amount of liquid was used. Method 2 allowed more eggs to be detected in the sample although the calculated eggs per centimetre column were lower.

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Both methods gave higher recovery rates with larger sand grains, increased stirring speed and increased stirring time as shown by table 6.7. The results from the increased stirring speeds suggest that pore size is an important factor. With increased agitation the pore sizes increase which may well give an egg a greater opportunity to enter the solution and therefore be counted when the solution is decanted. The increase time results suggest that once an egg is in the solution it is more likely to stay there than move back into the sand. This would imply a good circulation of sand during stirring.

To suggest an effective sand size required to obtain the WHO guidelines of less than one egg per litre of effluent, the results from method 3 should be considered. From table 7.2b an effective sand size of 0.4mm or less and 77 cm of sand would be required to obtain the guidelines on the addition of 250 eggs/l in the influent.

7.5.DESIGN PARAMETERS AND EGG REMOVAL

There is an obvious correlation between the effect of the various operational changes and egg removal. This section will discuss why a change in design parameters will affect the removal efficiencies of the filter.

7.5.1.Effective sand size

The first and most investigated operational variable was the change in sand size. It has been shown that a finer sand will improve the removal efficiency but should this be so? From section 7.2 it was established that interception and sedimentation were the dominant processes involved in the removal of Ascaris eggs. The effect of interception is determined by the equation I = d/D, where d is the diameter of the particle and D is the diameter of the grain. As D gets smaller I gets larger implying that a decrease in sand grain diameter improves the effect of interception. This occurs because there is less space between grains and therefore more laminar flow lines will be close enough to the sand grain to allow a particle present to touch the sand grain and be intercepted.

Sedimentation is affected by a smaller grain size as there is a greater surface area available for Ascaris eggs to be attached. The flow lines will also be closer to the sand grains and, as the velocity of flow decreases as the flow lines approach the sand grains, the settling velocity of the egg will have a greater effect on the egg than in larger pores.

The effect of these two processes are improved by the presence of a smaller sand grain, and these processes have the dominant effect on egg removal. Therefore this is the reason why a reduced sand size will improve the removal efficiency.

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7.5.2.Flow rate

The results from the changes in flow rate indicate that an increase decreases the efficiency of removal and vice versa. The velocity of the water flowing past the grain is therefore an important factor in removal. As velocity has no direct effect on interception the changes in the efficiency must be related to sedimentation. When water is flowing faster through a filter the effect of the settling velocity of the particle is proportionally lower. Therefore a particle is less likely to settle onto a grain.

Interception is indirectly affected by velocity. If a particle has a density greater than water it will leave a flow line when that flow line changes direction to move past a sand grain. At higher velocities this process may well be enhanced. It's new position will be closer to the sand grain than the corresponding flow line and it is therefore more likely to come into contact with the sand grain. When a particle is travelling faster it has a greater momentum and is therefore more likely to leave the flow line. Therefore an increased flow rate should aid the removal of dense particles. However, as Ascaris eggs are only just denser than water with a relative density of 1.1 it is unlikely that this phenomenon will have a great effect in changing the removal efficiency.

7.5.3.Mixed media

The third variable in the operational parameters was that of the dual media experiments. Dual media using anthracite and sand is a well established practice in Europe. The basic idea is to try and maintain a gradient of coarse to fine particles from the top to the bottom of the filter. This is maintained because the coarse anthracite is less dense than the sand and will therefore remain above it during backwashing. One of the phenomenon of a dual media is that the region of the interface between the medias tends to be a layer of efficient removal. This is because the fine sand and the coarse anthracite from the respective layers meet and the fine sand fills the gaps of the

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coarse anthracite. This gives a region of small pore size and, as stated before, this aids removal. This is the observation gained from the results shown in table 6.13. The Anthracite sand boundary occurs between 31 and 46 cm depth and both methods 2 and 3 show an increase in the number of egg at 46cm depth as opposed to the 31 cm sampling point. This is contrary to the previous results using a single media. Although the method two results for the anthracite sand experiments do not significantly show this fact, the observation that there is no decrease between the 31 and 46 cm results indicate a variance from the single media results.

The boundary between the sand and garnet dual media was between 46 and 61 cm. Again, there is a noticeable increase in the number of eggs detected at the 61cm sampling point as opposed to the point at 46cm depth. It is worth noting that the sand garnet mixed media runs were the only experiments that gave results of no eggs at the lowest sampling point. This indicates that garnet is very efficient at removing Ascaris suum eggs. This was surprising because the grading of garnet is similar to the the sand grading (0.4-0.7 mm). Although there are more fine particles in the garnet this would imply that either garnet packs closer together (sphericity 0.85) or there is some property of the garnet surface that attracts the eggs. This is unlikely because previous sections have indicated that the egg wall is inert to the processes responsible for egg attachment. Therefore the sand and garnet boundary probably has very fine pores due to tight packing which makes the passage of eggs very difficult.

It can be stated from the above discussion that the operational criteria have a profound effect on the removal of <u>Ascaris suum</u> eggs. If necessary these criteria can be used to adjust an operational filter so the WHO guidelines are obtained. This would also depend on the other operational restraints and the priority which is placed on the removal of helminth eggs.

7.6.BACKWASHING

The results from the backwashing of the filter with water only indicate that eggs are present at all levels of the filter after a 20 minute cleaning period. The quantity of eggs present indicates a greater than 95% removal efficiency. This is the efficiency expected by water only backwashing (Cleasby et al 1975). A backwash with air scour would improve efficiency but the technology required for this was not practical for the laboratory and it is unlikely that air scour would be used in an operational filter in countries where helminth eggs are an inherent problem.

Although the efficiency of backwashing is as good as expected, the fact that eggs remain at all depths of the filter after cleaning is of concern. Boller (1984) showed that filter efficiency is lower in the initial periods of a run and increases as some clogging occurs. This would imply that the eggs present after backwashing, especially in the lower regions of the filter, will be more likely to flow through the filter at the beginning of the next run. Although the sand was not replaced between runs, no build up of eggs was detected. This was due to a further 30 minutes backwashing after sampling was completed. Ιt was noticed however that a large circulation of sand occurred during the backwashing procedure. This phenomenon implies that the glass balls at the base of the filter were not working efficiently. It was expected that the backwash efficiency would improve with a more even fluidisation of the bed.

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7.7. SUMMARY OF DISCUSSION

One of the major findings of this work is the difficulty of obtaining concrete conclusions using present methods of detection of helminth eggs when conducting filtration studies. The difficulty in converting real number data into comparible information to establish the effects of an operational variable is compounded by the low efficiency and high variability of the detection procedures. Taking this into account, however, the data collected in this research provides useful information about the efficiency of rapid sand filtration for removing helminth eggs.

Rapid sand filtration does allow helminth eggs to pass into the effluent. However, operational criteria can be set such that an effluent containing less than one egg per litre can be obtained and thus comply with the WHO guidelines for reuse. Efficiency of removal will be improved by:-

i)Decreasing the effective sand size. This will change the efficiency from 99.3% removal for 0.7mm effective sand size to 99.8% removal for the 0.3mm effective sand size when 77cm depth of sand is used. This is a difference of between 1.8 eggs/l and 0.5 eggs/l in the effluent on the addition of 250 eggs/l. An effective sand size of 0.4mm was required to ensure an effluent quality of less than one egg per litre. These results were not proved conclusively but the evidence indicating that effective sand size has a major influence on filtration efficiency is very strong.

ii)Decreasing the flow rate. Decreasing the flow rate from 4m/h to 2m/h was equivalent to reducing the effective sand size to 0.45mm if 4m/h had been maintained. Increasing the flow rate from 4m/h to 6m/h was equivalent to increasing the effective sand size to 0.65mm.

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iii)By using a sand/garnet dual media with 50cm depth of 0.5mm effective sand size over 27cm depth of 0.4mm effective garnet size. This appears to remove all <u>Ascaris suum</u> eggs. It was established that this is probably due to the small pores present within the boundary of sand and garnet medias. Sand/anthracite dual media filters give the same general pattern of removal but <u>Ascaris suum</u> eggs will still pass.

The use of a sand only media can never theoretically achieve 100% removal of <u>Ascaris suum</u> eggs. This is because egg removal in a sand bed is based on an exponential curve which will never touch the x-axis. Practically, if the sand bed was deep enough, 100% removal would be possible. It is important to note that the majority of <u>A.suum</u> egg removal probably occurs on the surface of the filter bed. Once eggs prenetrate the surface layer the removal efficiency is dramatically reduced and continues to reduce as eggs progress through a sand filter.

Interception and sedimentation are the determining processes in Ascaris removal. Inertia, diffusion and hydrodynamic processes are less important as the egg particle is too large for these to have a profound effect.

The <u>Ascaris suum</u> egg wall is relatively inert and will not aid or hinder the processes of filtration removal. The wall possesses a small negative electrical charge of approximately 14mV but this will have a negligible effect compared to other dictating forces in a rapid sand filter.

As <u>Ascaris suum</u> eggs are in general larger than most other helminth eggs, it can be suggested that the eggs of other species will also pass a rapid sand filter. The process of removal is likely to follow a similar pattern, ie exponential, and as they are smaller a greater proportion are likely to pass. These differences are discussed in the next chapter.

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CHAPTER 8: THE PREDICTIVE MODEL

The experimental data indicate that eggs pass through a rapid sand filter. Under certain conditions the number of eggs passing are below the WHO guideline levels for effluent reuse. The data collection for the filtration indicated that by varying the effective sand size, flow rate, and media, the efficiency of removal varies. From these facts it is possible to achieve a general equation for the removal of helminth eggs in terms of these operational criteria. The object of this chapter is to obtain a model that will predict the number of helminth eggs in the effluent of a filter. The determining factors will be discussed with reference to the experiments conducted in previous chapters.

8.1. DETERMINING FACTORS

The determining factors for the model can only involve those which have been investigated. These include load of eggs, effective sand size, flow rate and mixed medias.

8.1.1.Load of helminth eggs

The determination of the load of helminth eggs in the influent of a rapid sand filter involves the use of the detection procedures described in Chapter 3. A model of the removal of helminth eggs in rapid sand filtration requires a starting point to assess the efficiency of the technique. For the predictive model discussed in section 5.2. the reference point is the number of eggs entering the filter. As such, the reliability of the model is dependent on the efficiency of the detection procedures that are used. This makes the detection procedure an important determining factor on the reliability of the model.

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8.1.2.Effective sand size

The effective sand size was the most investigated operational variable. From the results it was shown that the sand size was an important determining factor in the efficiency of a sand filter. As the effect of sand size had conclusive results the model is based on this variable.

8.1.3.Flow rate

The results for the variation in flow rate indicate that flow rate altered the efficiency of removal. However, time did not permit flow rate to be investigated enough to involve it in the model. The effect of flow rate, Q, can be illustrated by the equation:-

$$ab = -0.020Q + 0.553$$
 (1)

As this equation is based on only three points, it was considered that it was not reliable enough to incorporate it fully into the model. This is an obvious failing of the model, but if investigated further, the effect of flow rate could easily be incorporated or be used to create design tables. The model is only valid for a flow rate of 4m/h. From the flow rate observations an estimation of the efficiency can be made. Increasing the flow rate to 6m/h had the effect equivalent to reducing the sand size to 0.65mm at a flow of 4m/h. Decreasing the flow to 2m/h had the effect of using a sand size of 0.45mm at a flow of 4m/h. At present, however, this is the best estimation available.

8.1.4.Mixed media

It was observed from the experiments with dual media that the distribution of removal within the filter followed a different pattern to that of a single media filter. As the predictive model is based on the equation of removal from a single media it is invalid to suggest that it will predict the removal efficiency from a dual media. Therefore dual media are not included in the model. Any model trying to predict the removal efficiency of a dual media bed would need to have a much more complex mathematical basis to involve the region of removal occurring around the boundary between the media. It is however, useful to note that the only 100% removal of eggs occurred when a sand garnet media was used.

Other factors that may effect the removal efficiency of helminth eggs were not tested and therefore cannot be incorporated.

8.2. DEVELOPMENT OF THE MODEL

The basic equation for the removal of helminth eggs in a sand filter has been established as:-

$$N = N_0 e^{-aX^b}$$
(2)

To create a predictive model from the above equation the constants of N_0 , 'a' and 'b' need to be expressed in terms of the parameters of the rapid sand filter. These include effective sand size (E), uniformity coefficient (u) and number of eggs added to a filter.

 N_0 is related to the effective size by a straight line, correlation of 0.99. Therefore $N_0 = -cE + d$ where d is the total number of eggs added. This is calculated by the number of eggs per litre in the influent (C) multiplied by the flow rate (Q) in meters per hour and the time of the run (t) in hours. A correction factor of 15 is required to combine eggs/l and m/h. -c is the gradient which was 176000 or 0.53CQt. Therefore $N_0 = 15(CQt - 0.53CQtE)$ or 15CQt(1-0.53E).

Correlations were also done from the above data relating E, u and the product Eu against the values of 'a', 'b' and the product 'ab' obtained through experimentation. The closest correlations occurred for the results from method 2. 'a' was most closely correlated to the product Eu (r=-0.94) giving the equation of:-

$$a = -3Eu + 3.14$$
 (3)

The correlations of 'b' to E, u and the product Eu gave low correlations in the region of 0.74. The product ab was, however, closely correlated to all three being most closely correlated to E (r=0.96) giving the equation of:-

$$ab = -0.39E + 0.69$$
 (4)

As the a value was related to E (r=0.92) with the equation:-

$$a = -3E + 3.66$$
 (5)

equation (5) can be substituted into equation (4) to give:-

⇒

$$(-3E + 3.66)b = -0.39E + 0.69$$
 (6)

$$b = \frac{(-0.39E + 0.69)}{(-3E + 3.66)}$$
(7)

These separate equations can be placed into equation (1) to give an overall general equation of:-

$$N = 15CQt(1 - 0.53E)e^{-(-3Eu+3.14)X(-0.39E+0.69)/(-3E+3.66)}$$
(8)

where, N = Number of eggs at depth (X) X = depth of sand (cm) C = number of eggs in the influent (eggs/l) Q = flow rate (m/h) t = time of run (hours) E = effective sand size (mm) u = uniformity coefficient and e = exponential.

The equation is only valid when 3Eu < 3.14. This is effectively equivalent to E < 1. As the majority of sand filters are run with an effective sand size of 0.4-0.7mm (Barnes et al 1984) this should not be a problem. Some filters in Europe do contain sand up to 1.0 mm effective size and this would be on the limit for the suggested model. To assess accuracy of the calculated values 'a' and 'b', their values were compared with the experimental data. First the calculated values of 'a', 'b' and 'ab' were compared with the experimental values of a, b and ab respectively. If the results are similar to a straight line, a gradient of one and an intersection of zero would be shown. The results from this calculation are shown in table 8.1.

Table 8.1:Comparison by correlation of calculatedand experimental constants

corr	ela	ation	r-value	gradient	intersection		
==:							
'a'	v	а	0.702	1.008	-0.01		
'b'	v	ь	0.747	1.338	-0.07		
'ab'	v	ab	0.92	0.853	0.08		

As the correlations are reasonably good and the gradients and intersections do not vary far from one and zero respectively, the calculated lines can be said to represent the pattern of removal.

A second method for assessing the difference between the two sets of values is by using the matched pairs method. The t values from the method were -0.033, -0.548 and -0.721 for the differences between calculated and experimental a, b and ab respectively. All these values are below the t value for significant differences (t=2.776, d.f.=4) at the 95% confidence limits. This also implies that there is no significant difference between the calculated constants and the values obtained experimentally.

8.3.LIMITATIONS OF THE MODEL

The model has limitations which arise out of experimental and mathematical assumptions. These errors need to be considered when applying the model.

8.3.1.Experimental errors and assumptions

Whenever a generalised model is produced from experimental data some assumptions are required to enable the data to be manageable. The first experimental problems come from the detection of the helminth eggs. One of the terms in the model is the number of eggs in the influent and requires a reliable technique for the detection of helminth eggs. Chapter three has shown that the present techniques are not reliable enough to obtain accurate data on the number of helminth eggs in a sample. The unreliability of detection procedures will have a detrimental effect on the accuracy of the model.

There is also a problem with the detection procedures of the eggs in the filter. The sampling techniques were simple, repeatable and reasonably reliable in detecting eggs. There was, however, a variance in the numbers of eggs found from one run to the next. This was calculated as 80% for method 2 and 71% for method 3. Although this error is large and should be acknowledged during the use of the model, the methods showed obvious differences in the results for the various alterations of operational parameters. This was particularly clear in the results after the correction factor had been applied. Therefore the general trends indicated in the results are distinct enough for the development of the model. The first and determining mathematical assumption is that the equation

$$N = N_o e^{-aX^b}$$

is an accurate representation of the removal pattern of helminth eggs This assumption is acceptable due to the lack of in a filter. significant difference between the calculated and observed values. From this equation there were several assumptions in interpreting the constants into the physical parameters of filtration. The first was the creation of the term 15CQt(1 - 0.53E) as a representation of the term N. The correlation between effective size and the number of eggs present in the first centimetre of sand was very close (r=0.99), with a gradient of 176000. The assumption from here is that the figure of 176000 is related to the number of eggs added in a manner that is directly proportional. The factor 0.53 is a direct proportion of the number of eggs added. Although there is no direct evidence to support this assumption, it seems the most likely relationship. When incorporated into the model the results are reasonably accurate compared to the experimental data collected.

The next assumption made is perhaps more important to the accuracy of the model. To obtain the values of a and b of 3Eu + 3.14 and (0.39E + 3.14)(-3E+3.66) respectively, correlations were taken of the relationships between 'a', 'b' and the product 'ab' against E, u and the product Eu as explained in section 8.2. The terms were based on the correlations that gave r values closest to 1. That was the relationship between 'a' and Eu and between 'ab' and E. The other relationships had r values relatively close to 1 and could be the determining correlations. Again there is no direct evidence to suggest that the correlations chosen to obtain the values of a and b are the most accurate relationships. However, these values give results that are a fair representation of the removal pattern. At it is not possible to assess the reliability of the present assumptions as the model has not been tested in the field.

The model is designed to help a wastewater engineer establish whether a rapid sand filter will achieve the WHO guidelines of less than one egg per litre of effluent. Effluent that meets this guideline can then be used in irrigation safely with regard to preventing the transmission of excess helminthic diseases. This aim of a general application is not directly achieved by the model as it stands. The model is specific to a flow rate of 4m/h and to the use of <u>Ascaris</u> <u>suum</u> eggs only.

The effect of flow rate has been discussed in previous sections and it seems that a decrease in flow rate will increase the efficiency of removal and vice versa. The relationship between the changes in flow rate and removal efficiency was not investigated enough to enable reliable incorporation into the predictive model. This obviously limits the application of the model and further work is required before this variable can be included in the model.

The reasons for using <u>Ascaris suum</u> eggs were explained in section 2.4. They are an excellent indicator organism and they are available in large quantities. When the model is applied in design situations there will be many more helminth species present than <u>Ascaris suum</u>. A species list of the Jordanian raw sewage is given in section 3.3. which indicates five species regularly present and another four species occasionally present in the raw sewage. The number of eggs found was up to 350 eggs per litre using a technique that was established at around 50% efficient. The difference in the other species would suggest that they will be removed with different efficiencies. Therefore, is the use of <u>Ascaris suum</u> reliable in creating a generalised predictive model?

The other major helminth eggs, with their size, relative density and settling velocity are shown in table 8.2. This shows that apart from Schistosoma mansoni the other eggs are slightly smaller than Ascaris

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Species	Dimensions µm	density	settling m/h
=======================================		===============	
Ascaris	65 x 45	1.13	0.95
suum			
Ascaris	55 X 40	1.11	0.43
lumbricoides			
Schistosoma	50 x 150	1.18	5.23
mansoni			
Trichuris	22 x 50	1.15	0.48
trichuria		1112	00.0
bookworms	60 v 40	1 055	0.26
	00 X 40	1.000	0.20
(Anclystoma,			
Necator)			

Table 8.2:Size, density and settling velocity of major helminth species

<u>suum</u> eggs and their relative densities are similar. The smaller size effects the settling velocity and therefore will reduce the effect of sedimentation, one of the determining removal mechanisms of filtration. The small size also reduces the effects of interception. The removal efficiency of these species will therefore be lower than Ascaris suum.

The physical properties of the walls of the eggs in these other species have not been researched and therefore cannot be compared to the work discussed in chapter 7 on the <u>Ascaris suum</u> egg wall. As the literature suggests the eggs of the other species are less robust in the environment it can be assumed that their egg walls will give less protection than that of the Ascaris wall. How this effects the removal efficiency of the other species cannot, however, be determined.

To incorporate the other species into the model at this stage a margin of error, equivalent to the percentage decrease in the effects of the determining mechanisms, should be applied to the number of eggs leaving the filter. This is a maximum of $\pm 50\%$ and is calculated by determining the lower efficiency of the determining mechnisms due to the smaller sizes and lower settling velocities. These values for the effects of sedimentation and interception on the eggs of other species are related to the values for Ascaris. The margin of error will not

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alter the fact that rapid sand filtration can achieve the WHO guidelines. To assess the actual removal efficiencies of the other species field experiments would have to be under taken. It would be impractical to obtain the numbers of eggs required for extensive laboratory testing in this country. Laboratory testing has the added disadvantage of bringing highly pathogenic organisms into the laboratory.

The size of <u>Schistosoma mansoni</u> eggs will prevent them from passing through a filter. The presence of a prominent lateral spine will also aid their removal. This genus however, has the ability to hatch while in the filter and the resulting miricidium can then swim through the filter. The model presented here has no means to incorporate such a scenario and therefore can not predict the presence of schistosome larvae in the effluent of a filter.

The model is a simple representation of the removal of <u>Ascaris suum</u> eggs during rapid sand filtration. As such, it does not try to predict the overall efficiency of a filter run. As the model is simple it avoids the generalisations and complications of other filter equations. The sole aim of the model is to predict the number of helminth eggs emerging from a filter so the effluent can be reused safely in irrigation. Although untested and incomplete in the respect of flow rates, it begins to represent the removal pattern of helminth eggs and is therefore a useful starting point for further work in filtration prediction.

Figure 8.1 gives three examples of where the model may be useful in predicting the sand size, depth of filter and the time of filter run to maintain an effluent that will achieve the WHO guidelines for helminth eggs. The examples show that the criteria needed to obtain efficient removal can be achieved by simple, practical adaptations to the operating filter.

Figure 8.1:Examples of applications for the model

Example 1

What effective sand size is required to achieve the WHO guidelines of less than one egg per litre if:influent egg concentration is 200eggs/1 i) ii) flow rate is $4m^3/m^2/h$ iii) length of run is 24 hours and iv) the filter can hold 1 metre of sand. The solution involves calculating the number of eggs at the lowest sampling point for various sand sizes. Suggested E as 0.6mm with u as 0.7mm. (-0.6x0.39+0.69) $N = 15x200x4x24(1 - 0.53x0.6)e^{-(-3x0.6x0.7+3.14)100(-3x0.6+3.66)}$ $\Rightarrow N = 19641e^{-1.88 \times 100}^{0.456/1.86}$ $\Rightarrow N = 586.35$ \Rightarrow 99.8% removal \Rightarrow 0.41eggs/l in the effluent. This value needs to be multiplied by 1.5. This is equivalent to the correction factor required to included other species of helminth egg. Therefore the number of eggs in the effluent is 0.615 eggs/l. This complies with the guidelines. Suggested E as 0.7mm with u as 0.8mm (-0.7x0.39+0.69) $N = 15x200x4x24(1 - 0.53x0.7)e^{-(-3x0.7x0.8+3.14)100(-3x0.7+3.66)}$ $\Rightarrow N = 181152e^{-1.46 \times 100^{0.417/1.56}}$ ⇒ N = 1220.6 \Rightarrow 99.58% removal \Rightarrow 0.85 egg/l in the effluent. Applying the correction factor of 1.5 this value becomes 1.275 eggs/l. This does not comply with the WHO guidelines. Therefore the maximum effective sand size required to achieve the WHO guidelines under the above criteria is 0.6mm with an uniformity coefficient of 0.7mm.

Example 2

What length of sand bed is required to achieve the WHO guidelines if
i) influent egg concentration is 300eggs/l
ii) flow rate is 4m³/m²/h
iii) length of run is 24 hours
iv) sand available has an effective size of 0.7mm with an uniformity
coefficient of 0.8mm

Solution

To achieve less than one egg per litre 99.6% removal is required. Therefore the number of eggs at the lowest N must be less than 1728. Applying the correction factor of 1.5 this becomes 1152. To achieve the guidelines for all eggs the value of 1728 needs to be divided by the correction factor. The value of 1728 is calculated by multiplying the total number of eggs that will enter the filter (15CFt) by 0.004.

$$_{-1.46x}^{0.417/1.56}$$

⇒ 1152 = 271728e^{-1.46X}

⇒ $4.239 \times 10^{-3} = e^{-1.46 \times 0.417/1.56}$

 \Rightarrow -5.463 = -1.46X^{0.417/1.56}

⇒ X = 139

To achieve the WHO guidelines using the criteria above the sand bed would need to be 139cm or more deep.

Example 3

How long could a run last before eggs in the effluent would exceed the
WHO guidelines if:i) influent egg concentration is 300eggs/l
ii) flow rate is 4m³/m²/h
iii) sand available has an effective sand size of 0.7mm with a
uniformity coefficient was 0.8mm
iv) the bed depth was 75cm.

Solution

Again N < 1152.

⇒ 1152 = 15 x 300 x 4 x t(1-0.53 x 0.7) $e^{-1.46}$ x 75^{0.417/1.56} ⇒ 1152 = 15 x 503 x t x 9.75576x10⁻³

⇒ t = 15.65

Therefore the maximum amount of time available before the effluent exceeds the WHO guidelines is 15 hours.

CHAPTER 9:GENERAL CONCLUSIONS

The research undertaken during this study illustrates well the problems involved in obtaining reliable, conclusive data concerning the efficiency of filtration. In addition, detection procedures for helminth eggs have low recovery rates and high variability and extensive statistical correction is required to enable information from different operational criteria to made comparable. This inevitably leaves conclusions obtained in this type of research open to statistical debate. Nevertheless, the evidence presented in the thesis clearly identifies the mechanisms that effect helminth egg removal during rapid sand filtration.

The concluding statement from the work presented here is that helminth eggs will pass through operational rapid sand filters. It is possible, however, to reduce the number of eggs in the effluent to a level below the WHO guideline value for the safe use of wastewater in irrigation. A model was developed for the prediction of the removal efficiency of a rapid sand filter and from this several, more specific, conclusions can be made.

9.1. THE EFFICIENCY OF RAPID SAND FILTRATION

i)The postulated removal efficiency of <u>A.suum</u> eggs at 4m/h and 77cm depth of sand varies from 99.3% to 99.8% depending on the effective sand size. This is equivalent to 1.8 eggs/l to 0.5 eggs/l on the addition of 250 eggs/l in the influent. An effective sand size of 0.4mm or less is required to achieve the WHO guidelines for effluent reuse of less than one helminth egg per litre.

ii)The wall of the <u>Ascaris</u> <u>suum</u> eggs neither hinders nor helps removal by filtration.

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iii)It is postulated that the determining helminth egg removal processes are sedimentation and interception. Both these processes have greater theoretical removal efficiencies when sand grains are smaller. The changes in efficiency of these removal mechanisms is probably the major reason for observing higher overall efficiencies in helminth egg removal when smaller effective sand sizes were used.

iv)Other non-hatching species of helminth eggs will be removed by the same determining processes as <u>A.suum</u> although probably at a lower efficiency. The number of eggs of these species passing a rapid sand filter can be estimated by adding 50% to the number of <u>Ascaris suum</u> eggs passing the filter. This equates to the decrease in efficiency of the determining processes due to the smaller size and the lower settling velocities of these species. The removal of hatchable eggs cannot be assessed.

9.2. THE EFFICIENCY OF PRESENT HELMINTH EGG DETECTION PROCEDURES

i)Present techniques under best conditions are only up to 50% efficient.

ii)20 one litre samples free of eggs are required to be confident of a value of less than one egg per litre of effluent being detected reliably (Da Lage 1990).

iii)Sources of error include hindered settling and egg attachment to organic matter.

iv)Even with present techniques 350 eggs per litre were detected in a raw sewage during field studies.

9.3. THE PREDICTIVE MODEL

i)The reliability of the model is prone to the variances of the detection procedures for the number of eggs entering the filter, and the processes for identifying eggs once they are trapped in the filter.

ii)The model does not include flow rate, due to the effects not being extensively investigated. Flow rate does however, effect the efficiency of removal. This is an obvious failing of the model that can be rectified by further work.

iii)The model is only valid for values of Eu < 3.14. This is equivalent to effective sand sizes of less than 1 mm. The majority of operational rapid sand filters operate under this constraint.

iv)The model is a simple representation of the removal of helminth eggs during rapid sand filtration. It is not trying to generalise the overall removal of all particles during filtration but is aimed at allowing the operator to assess whether the conditions present in the filter will reduce helminth egg loads to levels that will make the subsequent effluent safe for reuse in irrigation.

9.4. FURTHER WORK

The practical extension to the work discussed in this thesis can be separated into two distinct areas. The first is the detection of helminth eggs in wastewaters and the second is the determination and reliability of the predictive model.

At present the detection of helminth eggs in wastewater is not reliable for detecting one egg in one litre of effluent. Work is currently being undertaken to assess the reliability of present techniques and to improve on their recovery rates by Aryes at Leeds University. Work carried out by Da Lage (1990) indicated that the best of these techniques gives 50% recovery of eggs under most favourable conditions and large numbers of eggs added. This recovery rate was greatly reduced when only 10 eggs per litre were added. It was interesting to note that Da Lage found the best recovery rates from a technique that used only water as the reagent. The reason for this and the continuation of this theme would be extremely useful work. Any increase in the reliability of the detection procedures will increase the reliability of the suggested model.

The predictive model represents the general removal pattern of helminth eggs through a rapid sand filter. It can not predict general filtration removal and it does not incorporate flow rate. Further work should include a continuation of the investigation on the effect of flow rate followed by work on real life situations to enable the model's reliability to be assessed with other species under operational conditions.

Procedures for the detection of eggs passing a filter is another area which warrants further investigation. The large variance within the techniques suggests that improvements should be investigated. Detection in a filter has always been a problem and the simple, repeatable techniques used here were adequate for the work undertaken. To improve the reliability of the model however, other more accurate methods of sampling sand in a filter at various depths need to be Others techniques that may be worth investigating include developed. the use of endoscopes and the use of dyes or other tracers to enable easier identification of the eggs during sampling.

The continuation of this research is worthwhile to develop further the model for the prediction of the removal of helminth eggs. The model could be developed into a useful representation of the pattern of removal in a rapid sand filter, particularly if this work were undertaken on a pilot filter under field conditions.

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APPENDIX A:Detection results of samples taken in Jordan, summer 1988

These results from sewage in Jordan were used to establish the number of eggs per litre that represented a realistic value to add to the filters during the data collection.

Table A.1:Helminth eggs/l in raw sewage from Jerash, Jordan samples kept in fridge overnight

Species	eggs/l	in s	amples	s take	en on	(1988	3)				
	19.7	19.7	30.7	31.7	31.7	3.8	3.8	7.8	7.8	7.8	Ave
Ascaris lumbricoides live	88	55	121	77	121	88	77	22	22	88	75.9
Ascaris lumbricoides dead	0	11	11	11	22	11	22	11	0	22	12.1
Anclostoma duodenale	22	66	11	44	44	22	44	55	88	77	47.3
Necator americanus	55	66	55	22	44	44	44	55	66	66	51.7
Trchuris trichiura	11	11	0	0	0	11	11	11	11	22	7.8
Enterobius vermicularis	55	33	33	55	11	44	55	33	0	11	33
Paragonimus westermani	0	11	11	11	11	0	11	22	11	22	11
Unidentified	55	55	44	44	22	22	33	44	44	55	41.8
SUB TOTAL	286	308	286	264	275	242	297	253	242	363	281.6
Hymenolepis	0	22	0	0	0	0	0	0	0	0	2.2
Diphyobo-	0	0	0	11	0	0	0	0	0	0	1.1
Schistosoma	0	22	0	33	0	0	0	11	11		7.7
Schistosoma haematobium	0	0	11	11	0	0	0	0	0	0	2.2
TOTAL	====== 286	== = == 352	===== 297	=== = 319	===== 275	===== 242	===== 297	===== 264	===== 253	===== 363	294.8

Species	eggs/l in samples taken on (1988)										
	19.7	19.7	30.7	31.7	31.7	31.7	3.8	3.8	7.8	7.8	Ave
Ascaris lumbricoides live	===== 143	132	121	77	121	198	110	121	33	55	111.1
Ascaris lumbricoides dead	33	0	22	11	0	55	0	11	0	0	13.2
duodenale	55	22	55	55	66	55	33	22	44	77	48.4
Necator americanus	44	44	99	44	44	33	44	55	66	77	55
Trchuris trichiura	11	0	11	22	0	11	44	22	22	33	17.6
Enterobius vermicularis	44	11	11	55	44	44	22	55	22	11	31.9
Paragonimus westermani	22	44	22	22	0	11	0	22	22	22	18.7
Unidentified	33	55	11	66	22	55	44	44	66	6	46.2
SUB TOTAL	385	308	352	352	297	462	297	352	275	341	342.1
Hymenolepis	0	0	0	0	0	0	11	0	0	0	1.1
Diphyobo-	0	0	0	0	0	0	0	0	0	0	0
Schistosoma	0	0	0	11	11	0	0	0	1	0	3.3
Schistosoma haematobium	0	0	0	0	0	0	0	0	0	0	0
TOTAL	385	308	==== 352	363	= = ==== 308	462	===== 308	352	===== 286	31	346.5

Table	A.2:Helminth	eggs/l	in r	raw se	wage	from	Jerash,	Jordan
	Samples	left on	the	bench	over	night		

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Species	Salt, 8 WARM W	.8.88 ARM C	Ai DLD Co	nman, 1 DLD W	31.8.88 ARM WA	B ARM C	OLD C	OLD	
Ascaris lumbricoides live	105	84	66	 63	70	 77	===== 55	===== 143	
Ascaris lumbricoides dead	0	0	0	0	0	11	15	11	
Anclostoma duodenale	63	42	66	63	10	33	25	33	
Necator americanus	84	63	66	63	10	11	15	11	
Trchuris trichiura	0	21	22	0	5	11	5	0	
Enterobius vermicularis	0	0	0	0	20	11	10	11	
Paragonimus westermani	21	0	0	0	0	11	0	0	
Hymenolepis	21	0	0	0	0	0	0	0	
Unidentified	63	63	44	42	0	55	10	22	
TOTAL		====== 273	===== 264	===== 231	115	====== 220	====== 135	231	

Table A.3:Helminth eggs/l in raw sewage from Salt and Amman, Jordan

•

•	00	-		` '			
	19.7	19.7 30.7	31.7 31.7	3.8 3.8	7.8 7.8	7.8	Ave
Ascaris lumbricoides live	60	30		60			===== 50
Ascaris lumbricoides dead	20	60		20		2	33.3
Anclostoma duodenale	20	0		20		-	13.3
Necator americanus	40	30		0			0
Trchuris trichiura	0	0		0			0
Enterobius vermicularis	0	0		20			6.7
Paragonimus westermani	0	30		0			10
Unidentified	60	60		0			40
SUB TOTAL	200	210		160			190
Hymenolepis	0	0		0			0
Diphyobo-	0	0		0			0
Schistosoma mansoni	0	0		0			0
Schistosoma haematobium	0	0		0			0
==== = === = ===== TOTAL	===== 200	============= 210	============	======================================		228253	190

Table A.4:Helminth eggs/l in oxidation ditch from Jerash JordanSamples left in the fridge overnight

Species eggs/l in samples taken on (1988)

The gaps in the data represent days when the quanity of organic matter present made any detedction impossible.
Species	eggs/l	in sample:	s taken on	
	19.7	19.7 30.7	31.7 31.7 31.7 3.8	3.8 7.8 7.8 Ave
Ascaris lumbricoides live	s 80	90	80	83.3
Ascaris lumbricoides dead	s 20	60	40	40
Anclostoma duodenale	20	0	20	13.3
Necator americanus	80	90	40	70
Trchuris trichiura	0	· 0	20	6.7
Enterobius vermicularis	0	30	40	23.3
Paragonimus westermani	20	0	0	6.7
Unidentified	100	120	0	0
SUB TOTAL	320	390	240	316.7
Hymenolepis	0	30	0	10
Diphyobo-	0	0	0	0
Schistosoma mansoni	0	0	0	0
Schistosoma haematobium	0	0	0	0
TOTAL	== = ==== 320	== = =================================	240	326.7

	Samples	left on	the	e bench d	overnigh	ıt		· · · ·
Table	A.5:Helminth	eggs/l	in	oxdation	n ditch	from	Jerash	Jordan

Gaps in the data represents dates when the quantity of organic matter present made detection impossible.

				19.7	19.7	30.7	31.7	31.7	3.8	3.8	7.8	7.8
NO.	OF	EGGS/L	W	0	0	0	0	0	0	0	0	0
			С	0	0	0	0	0	0	0	0	4
NO.	OF	SPORES/L	W	0	0	0	62	44	42		310	600
			С	0	0	0	0	0	10		500	400

Table A.6:Helminth eggs/l in Sedimentation tank from Jerash Jordan

Table A.7:Helminth eggs/l in sedimentationtank from Salt Jordan

Conditio Warm	n of Warm	sample Cold
10	5	4
10	0	6
L		
5	0	0
	-	Ū
5	0	0
2	Ŭ	Ū
5	Ο	4
		4
35	5	14
	Conditio Warm 10 10 5 5 5 	Condition of Warm Warm 10 5 10 0 5 0 5 0 5 0 5 0 5 0

APPENDIX B:Efficiencies of filter detection procedures

Table B.1:Method two when 300 eggs/g dry sand is added

	weig	ht of 10	sano	d (g)	20			30			40			50	
ml of water added	ml left	mean eggs n=6	% rec												
10	6	89	30	5	98	33	2	14	4.7						
15	10	52	17	8	41	14	5	59	20	3	19	6.4			
20	15	105	35	13	64	21	9	97	32	8	51	17	8	2.1	0.7
25	21	124	41	17	88	29	15	52	17	16	56	19	14	136	45
30	25	264	88	23	77	26	20	64	21	19	113	38	17	91	30

Table B.2:Method two when 200 eggs/g dry sand is added

ml of water added	weig ml left	ht of 10 mean eggs n=6	sano % rec	d (g) ml left	20 mean eggs n=6	% rec	ml left	30 mean eggs n=6	% rec	ml left	40 mean eggs n=6	% rec	ml left	50 mean eggs n=6	% rec
10	7	124	62	4	56	28	2	8.7	4.3						
15	11	123	62	8	74	37	5	27	14	4	17	8.3			
20	17	196	98	12	89	45	11	59	30	9	78	39	8	17	8
25	22	19 8	99	18	157	79	16	107	53	14	35	17	10	62	31
30	26	162	81	23	156	78	20	142	71	19	82	41	17	39	19

Table B.3:Method two when 100 eggs/g dry sand is sand

ml of water water	weig ml left	nt of 10 mear eggs n=6	f san n % s rec	d (g) ml left	20 mean eggs n=6	% rec	ml left	30 mean eggs n=6	% rec	ml left	40 mean eggs n=6	% rec	ml left	50 mean eggs n=6	% rec
10	8	107	107	5	24	24					=====			=====	
15	11	44	44	10	26	26	9	37	37	7	22	22			
20	16	48	48	17	31	31	14	17	17	10	26	26	5	1.9	1.9
25	22	56	56	21	30	30	18	29	29	16	30	30	11	32	32
30	27	81	81	25	94	94	25	48	48	20	43	43	18	46	46

Table B.4:Method two when 50 eggs/g dry sand is added

	weig	ht of 10	sand	d (g)	20			30			40			50	
ml of water added	ml left	mean eggs n=6	% rec												
10	8	42	84	5	20	40									
15	11	34	68	9	16	32	7	12	24	5	5	10			
20	17	43	86	15	19	38	12	19	38	9	9	18	8	5	10
25	23	51	102	19	19	38	18	15	30	16	19	38	12	21	42
30	28	62	124	26	38	76	23	28	56	20	23	46	18	26	52

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Table B.5:Method two when 25 eggs/g dry sand is added

ml of water added	weig ml left	ht of 10 mean eggs n=6	sano % rec	d (g) ml left	20 mean eggs n=6	% rec	ml left	30 mean eggs n=6	% rec	ml left	40 mean eggs n=6	% rec	ml left	50 mean eggs n=6	% rec
10	8	17	 68	6	9	36		=====						=====	
15	11	12	48	10	13	52	7	7	29	4	2	8			
20	17	21	84	14	19	76	12	12	48	11	6	24	6	1	4
25	22	29 1	118	20	18	72	18	8	32	15	15	60	11	6	24
30	26	24	96	25	31 1	26	21	20	80	20	11	44	18	10	40

Table B.6:Method two when 10 eggs/g dry sand is added

	weig	ht of	sand	d (g)	00			20			10			50	
ml of water added	ml left	nean eggs n=6	% rec	ml left	20 mean eggs n=6	% rec	ml left	30 mean eggs n=6	% rec	ml left	40 mean eggs n=6	% rec	ml left	mean eggs n=6	% rec
10	8	4.9	49	5	2.5	25									
15	11	4.3	43	10	3.1	31	6	1.8	18	4	0.5	5			
20	17	8.1	81	15	3.6	36	11	2.6	26	8	0.9	9	7	0.5	5
25	22	7.3	73	19	7.4	74	18	3.5	35	15	2.3	23	11	2.1	21
30	26	12	12 0	25	5.4	54	22	4.8	48	19	2.7	27	18	2.9	29

Table B.7:Method three when 20ml of sufactant is added

eggs/g dry sand	weigh 15 mean eggs n=6	nt of % rec	sand 20 mean eggs n=6	(g) % rec	25 mean eggs n=6	% rec
200	111	55	111	-==== 55	===== 91	 46
100	54	54	51	51	44	44
50	38	76	21	58	25	50
25	10	40	10	40	8	32
10	5	50	4.6	46	3.9	39

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Comparisor	n of tw	vo independer	nt variable	s		
standard	3106	5592	4562	3850	784	450
dev	212	. 87	280	300	96	81
	60	45	118	94	51	37
	123	87	102	158	42	40
	51	51	75	89	70	64
	42	27	75	53	62	34
Variance 9	644735	31307026	20810666	14824521	615102	202462
	44944	7650	78548	90197	9195	6570
	3634	2050	13815	8816	2556	1333
	15061	7511	10394	24948	1736	1596
	2577	2608	5632	7931	4873	4101
	1772	728	5567	2792	3835	1140
F-value 3.	246022		1.403800	3	.038115	
5.	874949		1.148298	1	.399409	
1.	772624		1.566976	1	.917031	
2.	005271		2.400142	1	.087549	
1.	012048		1.408048	1	.188210	
2.	433138		1.994300	3	.364998	

APPENDIX C:Statistical analysis of the pilot study results (Section 5.3.2)

The F-value is calculated by dividing the larger variance value for the same sampling point of the different rigs by the other value from the same sampling point. This results in a number greater than one and can be related to the confidence limit.

The 95% confidence limit for f4,4 degrees of freedom is 6.39. As can be seen from the F-values above, the comparison values of all the sampling points of the rigs are below this value. Therefore it can be said that there is no significant difference been the sampling points of the two rigs (p<0.05).

Macthed pairs

Technique	Two	Technique	Three	Backwash

Differences

А	MEAN	FOT MEAN	А	MEAN	TOT MEAN	А	MEAN	TOT MEAN
16.9	13.440	2.453	30.5	5.204	1.050	10.4	1.682	0.424
37.2	SD	TOT SD	-14	SD	TOT SD	1.88	SD	TOT SD
7.1	27.024	12.057	40.02	28.869	11.758	-1.27	5.141	2.155
35.3	t-VALUE	t-VALUE	-3.5	t-VALUE	t-VALUE	-0.04	t-VALUE	t-VALUE
-29.3	1.1121	0.4549	-27	0.4031	0.1996	-2.56	0.7316	0.4399

-1**9**8-

В	MEAN	В	MEAN	В	MEAN
2.55	0.924	1.2	0.390	0.4	-0.012
0.36	SD	-2.33	SD	-0.12	SD
0.07	1.067	1.5	1.607	0.34	0.150
1.47	t-VALUE	0.19	t-VALUE	0.05	t-VALUE
0.17	1.936	1.39	0.543	-0.01	-0 .179
С	MEAN	С	MEAN	С	MEAN
0.2	0.256	-0.9	0.020	0.7	-0.182
0.36	SD	0.53	SD	0.02	SD
0.61	0.240	0.46	0.669	-0.36	0.122
-0.02	t-VALUE	0.5	t-VALUE	0.06	t-VALUE
0.13	2.380	-0.49	0.067	-0.12	-3.332
D	MEAN	D	MEAN	D	MEAN
0.2	0.052	1.5	0.462	0.6	-0.148
0.39	SD	0.67	SD	0.31	SD
0.43	0.418	-1.52	1.167	0.42	0.093
-0.53	t-VALUE	1.08	t-VALUE	0.42	t-VALUE
-0.23	0.278	0.58	0.885	0	-3.575
Е	MEAN	E	MEAN	E	MEAN
-0.5	0.020	0.2	0.316	0.3	-0.104
0.65	SD	-0.15	SD	0.38	SD
-0.11	0.417	0.27	0.352	0.97	0.391
0.1	t-VALUE	0.81	t-VALUE	-0.03	t-VALUE
-0.04	0.107	0.45	2.009	-0.64	-0.594
F	MEAN	F	MEAN	F	MEAN
-0.2	0.024	0.1	-0.094	0	-0.120
-0.07	SD	0.63	SD	0.31	SD
0.23	0.174	-0.43	0.493	0.04	0.230
0.16	t-VALUE	-0.13	t-VALUE	0.3	t-VALUE
0	0.309	-0.64	-0.426	-0.03	-1.166

ø

X G- (depth)	VALUE (lnX)	× _i -	-X [–] (X _i -X) ²		
16 2 31 3 46 3 61 4 76 4	.7725887 .4339872 .8286414 .1108739 .3307333	-0.9227 -0.2013 0.1332 0.4155 0.6353	7762 0. 3777 0. 2765 0. 5090 0. 3684 0.	======= 8515159 0683183 0177626 1726477 4036930		
Σ Σ Σ ²	3.695364 18.476824 69.792546	49 45 50	Σ	1.5139376		
N-Valu	es F-VAL	LUES	$Y_{i} - \overline{Y}$	$(Y_{i}-\overline{Y})^{2}$	X _i xY _i	$(X_i - \overline{X})(Y_i - \overline{Y})$
A 17025 B 315 C 173 D 138 E 91 F 65	1.383 1.523 1.571 1.654 1.717	7575 -0 6941 -0 7744 0 7131 0 0451 0	0.1864393 0.0465027 0.0015775 0.0845163 0.1468482	0.0347596 0.0021625 0.0000025 0.0071430 0.0215644	3.8365905 5.2323459 6.0177603 6.8023167 7.4360643	0.172041744 0.009364616 0.000210249 0.035117266 0.093312734
	Υ 1.5701 Σ 7.8509	96813 84067	Σ	0.0656320	29.3250778	0.310036609
To find	the poin	$r = \frac{\Sigma(1)}{\sqrt{[X_i]}}$ ts on th	<u>X;-X)(Y</u> -X)'(Y <u>i</u> e line:-	$\frac{-Y_{0}}{(2)^{2}} = 0.98$	35606	
		$b = \frac{\Sigma(X)}{\Sigma X}$	$\frac{i \frac{XY}{2}}{i} \frac{j - (\Sigma)}{[(\Sigma)]}$	$\frac{(\Sigma_{i})^{2}}{(1-1)^{2}} = \frac{(1-1)^{2}}{(1-1)^{2}}$	0.2066313	
		A= Σ¥ī	- (b x Y	() = 0.80661	88	
		a=	e	$e^{A} = 2.24032$	02	
These va	alues can	be plac	ed in equ	ation (10a)	to give:-	

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APPEDIX D:Calculations to obtian the equation of the line for method two of the pilot study

 $N = N_0 e^{-2.2403202x} 0.8066188$ This procedure was followed for the creation of all the lines from the data collection except for the variation in media.

Ta	Table E.1:Raw data from method two at various effective sand sizes for 4m/b											
		errec	LIVE SA		<u>s 101 4</u>	<u>III / 11</u>						
de	pth of											
sa (c	umpie m) r	un 1	r	un 2	r	un 3	rı	ın 4	ru	n 5		
==		=======	=	=======	=======			========	=======	=====		
	Effec	tive sa	nd size	0.3mm,	Flow ra	ate 4m/h 	1 					
1	23482	29826	37982	24073	24448	11071	25374	16371	18065	20390		
16	209	181	148	181	278	120	386	193	225	173		
31	162	59	5/	28	6/	108	130	32	5/	35		
40	57	51 47	53 53	51 51	67 47	110	67	40	79	69		
76	45	47	41	53	47	39	39	49	0	41		
==	Effec	======= tive sau	nd size	 0.4mm.	 Flow ra	======= ate 4m/h	======= 1			=====		
				,								
1	40287	37765	38139	39282	40070	39636	39794	38888	37726	40976		
10 21	300	315	229	250	400	567	66U 270	938	481	120		
46	130	120	97	209	290 294	99 95	240 197	142	114	114		
61	49	120	37	134	100	83	118	104	85	39		
76	65	71	41	41	49	87	55	53	59	57		
==:	Effect	tive sar	nd size	0.5mm,	Flow ra	te 4m/h	=#2====			=====		
	 38257	43773	38060	31185	44227	38179	44975	35322	36031	33017		
16	861	1030	461	437	887	313	875	437	552	536		
31	528	349	185	102	378	165	347	225	449	294		
46	378	274	229	106	191	108	433	321	319	171		
61	518	234	130	57	197	126	165	142	171	99		
/6	1/5 ======	136	116	/5	244	114 =======	142 =======	140 ======	1/3 =======	160		
	Effect	ive san	d size	0.6, Fl	ow rate	4m/h						
1	29905	29077	27738	29806	45527	31244	379 4 2	28762	44128	51456		
16	453	329	1123	294	2266	1503	1745	676	812	1143		
31	384	268	453	229	1442	581	611	313	51	209		
40 61	93	47	230 107	110 144	430	398	246	319 225	299	223		
76	89	67	175	69	193	164	89	142	213	165		
===	====== Effect	====== ive san	d size	====== 0.7mm,	====== Flow ra	====== te 4m/h	======		======			
	26762									01105		
⊥ 16	36/6U 1320	40385	39006	3/332	46453	39085	52205	41626 1915	41863	31185 1757		
10 31	1000 556	777 7700	202 266	1112	1942 000	50 707	5034 730	1215 797	214/ 1772	1688		
46	362	292	422	554	794	202 394	1095	794	628	942		
61	307	487	266	201	516	280	810	491	262	764		
76	284	142	140	165	325	199	282	193	181	282		

APPENDIX E:Raw data of eggs at each sampling point for each run

de sa (c	pth of mple m) ru	1 n	rı	un 2 run 3			rı	un 4	run	5
==	Effect	tive sar	nd size	0.3mm,	Flow ra	ate 4m/ł	======== 1		:======	=====
1 16 31 46 61 76	26831 439 366 370 116 0	31599 1584 217 223 162 112	27304 615 106 91 0 0	31382 810 136 0 195 0	34554 855 225 167 0 97	34022 441 286 181 81 83	31678 493 384 203 93 61	26871 575 205 91 102 100	27698 619 225 288 0 99	24704 445 229 89 93 81
==:	Effect	ive san	d size	0.4mm,	Flow ra	ate 4m/h	:==232222 	:======		=====
1 16 31 46 61 76	28723 1182 234 110 108 100	38198 847 353 203 136 93	33372 678 282 203 97 104	38454 1753 459 333 97 102	35519 721 272 244 93 79	23207 841 471 225 193 91	38908 656 276 256 201 175	38021 778 422 266 110 93	39814 1048 256 217 77 83	34298 463 297 201 97 71
	Effect	ive san	d size	0.5mm,	Flow ra	ite 4m/h				
1 16 31 46 61 76	44857 782 735 426 554 359	31461 753 327 469 623 538	37824 341 758 331 120 120	38868 747 502 199 97 85	30102 418 299 236 227 164	42119 1190 402 264 189 89	35657 1799 656 522 321 232	36524 802 457 573 250 307	28053 1692 550 378 272 191	27541 654 445 248 323 205
	Effect	ive san	d size	0.6mm,	Flow ra	te 4m/h				
1 16 31 46 61 76	27954 926 550 325 229 272	32604 634 398 394 122 160	30712 3388 477 345 299 116	38573 1227 583 406 240 303	46000 2679 1537 567 339 205	41370 1842 577 364 361 215	44089 3605 1537 556 343 258	47142 2344 583 496 333 221	35440 1885 916 668 536 313	34987 2031 597 540 274 317
===	Effect:	ive san	d size	======= 0.7mm,	===== Flow ra	te 4m/h				
1 16 31 46 61 76	37292 1728 508 526 420 183	31579 1501 796 632 437 313	53958 3310 1048 601 554 435	36997 2356 1024 849 388 305	35421 2311 1499 1501 648 408	49250 3231 1706 626 402 250	37923 2581 1541 644 705 388	41961 2660 2167 1192 810 536	34987 1850 869 693 345 479	40227 1775 1355 591 368 361

Table E.2:Raw data from method three at variouseffective sand sizes for 4m/h

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de sa (c	epth of ample am)									
==	Effect	ive sand	size	0.3mm,	Flow ra	te 4m/h	=====	======	=======	
 1 16 31 46 61 76	2506 105 0 53 63 0	1082 90 94 56 0 33	927 181 39 34 0 48	658 150 0 61 0 60	1724 0 0 0 0 0	2236 362 0 0 63 62	1105 128 0 0 52 0	1003 0 0 44 0 0	1264 59 0 34 0 0	408 0 0 0 48 0
	Effect	ive sand	size	0.4mm,	Flow ra	te 4m/h				
1 16 31 46 61 76	3191 0 0 0 0 0 0	1058 51 0 41 149 67	2073 117 0 33 0 0	1485 108 39 68 0 40	3072 149 0 0 46 38	822 143 64 44 0 56	638 100 0 0 49	225 78 32 0 0 0	374 145 0 0 32 0	3415 142 40 22 29 43
	Effecti	ive sand	size	0.5mm,	Flow ra	te 4m/h				
1 16 31 46 61 76	404 369 224 115 153 121	392 164 106 282 235 0	425 234 173 126 96 78	339 0 49 107 52 0	1599 36 82 51 59 0	94 84 0 0 0 68	2047 83 43 119 43 83	653 43 116 68 0 121	361 211 118 139 89 59	82 198 0 46 56 38
	Effecti	ve sand	size	0.6mm,	Flow ra	te 4m/h				
1 16 31 46 61 76	787 165 39 0 0 0	590 68 0 100 0 0	1235 67 108 81 0 0	716 34 0 0 0 41	738 91 0 107 0 0	1165 0 233 106 0 0	1630 89 49 0 33 0	112 0 0 0 0 0 0	1105 197 67 0 0 121	2133 63 0 0 159 0
	Effecti	ve sand	size	0.7mm,	Flow rat	te 4m/h				
1 16 31 46	390 67 0 0	482 0 40 0	2709 122 0 0	884 360 11 0	3217 117 0 0	3032 167 248 0	172 0 0 0	2100 355 47 0	330 109 28 0	5368 231 32 0

Table E.3:Raw data from Backwsh using method 2 for variouseffective sand sizes, Flow rate 4m/h

de sa	pth of mple									_
(ci	m) ru ======	n 1 ======	r =====	un 2 =======	ru =======	ın 3 =======	r: ======:	un 4 =======	run ======	5 ======
	Method	2:Effe	ctive	sand si	ze 0.5mm	, Flow	rate 6	n/h		
1	31776	26403	2446/	241/2	34928	17750	36209	30547	28067	3221(
16	1294	920	146/	/23	1158	1214	1003	920	660	134
15	266	270	851	د ۱/ د ع د	276	294	512	349	403	3/6
40	150	183	20/	202	284	250	248	2/0	223 171	29,
76	120	170	529 199	57	402	240 211	132	116	162	116
-==		=======		-======		=======		========	======	
	Effect	ive san	d size	0.5mm,	Flow ra	te 2m/ł	ı			
1	30535	23325	39262	39556	27639	30653	26885	39006	43931	39400
16	1954	890	520	772	794	402	1371	467	579	362
31	463	171	93	110	209	227	102	167	248	77
46	110	132	93	100	124	130	93	138	104	81
61	138	118	99	49	53	0	45	57	106	73
76	55	93	138	0	45	45	0	39	45	61
===	Method	3:Effe	ctive :	sand si	======= ze 0.5mm	======= . Flow	rate 61	:====== 1/h		=====
1	29471	36347	20370	21512	19976	18597	31500	20685	21631	18814
16	2593	1942	1702	1186	1745	1942	2933	1653	1302	2271
31	772	717	762	869	410	587	469	603	778	835
46	528	366	431	361	431	615	345	422	351	473
61	364	335	303	357	305	345	317	288	242	299
76	266	238	286	189	230	286	175	315	384	260
===	Fffecti		======================================	-======= 0 5mm	Flow ra	====== to 2m/h	======	======	======	=====
1	22606	17454	32998	40444	26195	28240	30334	31323	23246	24369
16	589	786	721	1367	715	782	591	1326	638	1089
31	305	266	282	246	463	388	343	477	250	307
46	380	252	116	240	305	193	339	195	240	201
61	118	126	252	118	193	102	97	95	128	108
76	85	97	95	87	95	97	95	0	0	85
===	====== Backwar	=======	====== d 2					-=====	=========	
	Effecti	ve sand	l size	0.5mm,	Flow rat	te 6m/h				
1	2358	1298	2851	3593	3723	1673	1814	3000	1489	2640
16	116	343	0	43	116	41	0	100	187	203
31	167	0	0	240	0	95	0	37	0	0
46	91	71	136	128	0	0	244	0	0	179
51	37	109	53	51	122	0	120	0	43	0
76	63	0	0	57	33	0	83	0	0	47
===:	====== Effecti	======= ve sand	====== size	·======== Ω 5mm	Flow rat	:====== • 2m/h			======	=====
1	10140	999	666	1533	7427	4289	3727	2384	662	4289
16	201	93	211	91	75	97	223	0	112	07
31	201	0	160	0	0	69	130	104	0	69
i6	181	71	0	Ō	Ō	49	0	45	0	49
51	57	203	69	83	45	43	47	0	0	43
'6	55	71	0	0	49	43	0	39	0	43
					-204-					

Table E.4: Raw data for the altered flow rate for methods two and three

de sa	pth of mple									
(c ==	m) ru: =======	n 1 ======	ru: ========	n 2 ======	ru ======	n 3 ======	rı =======	ın 4 =======	ru: =======	n 5 =====
1 16 31 46 61 76	Method 34996 4264 719 524 128 0	2:Antl 36756 7164 1284 398 124 65	ricite/: 35025 5116 838 1517 264 128	sand me 36015 7007 1823 715 138 122	edia, F1 25323 2239 185 611 154 49	ow rate 28312 5223 340 1009 83 118	4 m/h 22246 7095 507 623 221 63	23892 3674 357 477 116 79	35329 5485 657 918 341 126	33467 5831 561 504 144 106
1 16 31 46 61 76	Sand/ga 37982 1391 380 112 268 0	arnite	media, B 40582 1174 234 110 306 0	low ra?	ite 4m/h 25748 512 203 148 155 0		34672 388 203 97 208 0		34475 950 311 93 123 0	
1 16 31 46 61 75	Method 32663 3187 617 376 100 102	3: An th 28763 2540 305 2149 597 116	ricite/s 24294 5596 387 1056 301 390	sand me 27028 3479 415 1450 847 102	dia, Flo 24823 1666 245 792 238 110	27401 27401 4126 1303 2837 382 95	4m/h 19678 1519 391 1084 116 89	27420 1971 565 1816 351 130	19090 2445 1045 2029 232 93	17954 4041 533 672 305 276
1 16 31 46 61 76	Sand/ga 24054 1609 644 262 436 0	arnite	media, F 32663 1970 473 353 718 0	===≡== 'low ra	te 4m/h 27127 2403 491 349 176 0		31638 2384 426 85 172 0		25118 1324 359 173 246 0	
1 16 31 46 61 76	Backwas Anthric 1015 155 97 331 102 45	h Meth ite/sa 986 0 140 0 0	od 2 nd media 3293 547 0 0 49 0	Flow 977 0 146 134 0	rate 4 1853 0 269 244 93 120	h/h 1212 348 0 0 164 0	1636 340 0 0 0 100	====== 1879 99 154 0 0 0	996 316 0 57 0 83	2469 135 116 112 0 0
1 16 31 46 61 76	Sand/ga 873 95 0 0 127 0	rnite i	nedia, F 1202 104 167 0 0 0	low rat	te 4m/h 1342 67 0 102 0 0 -205-		2187 61 0 33 74 0		1655 0 81 0 0 0	

Table E.5:Raw data for the mixed media from methods two and three

- -

dej sai	pth of mple	n 1	ru	n 9	r	ר מוו	rı	חו /	ru	n 5
===	=======			======		=======		========	======	======
	Method	2:Wate	er, Effe	ctive	sand si	ze 0.5mm	ı, Flow	rate 4m	ı/h	
1	26260	30043	41370	29649	38060	31697	34593	36012	41390	27836
16	416	353	1034	487	489	256	666	737	1160	747
31	278	223	422	366	181	242	244	254	361	217
46	227	21/	150	223	183	134	175	144	120	144
61	197	286	154	164	16/	150	16/	0	134	99
/6	234 =======	108 ======	811 =======	91 =====	5/ :======	59 =======	89 =======	49 =======	/3 ======	/ ز =====
	Efflue	nt, Eff	ective s	sand s	ize 0.5	mm, Flow	rate 4	µm∕h		
1	38454	3106/	40129	30653	33707	39262	27344	28132	34810	33864
16	1064	1024	491	861	/60	1018	506	609	634	930
31	252	156	455	197	453	229	21/	329	240	329
46	1/3	114	303	183	142	213	132	296	146	229
61 7(112	128	116	106	124	146	142	144	152	1/1 1/0
/0 ===	49 =======	,⊂ ======	102 =======	63 ======	97 ======	108	6/ =======	150	118 =======	14Z =====
	Method	3:Wate	r, Effec	tive	sand siz	ze 0.5mm	, Flow	rate 4m	/h	
1	226/5	24192	27068	26398	29530	30693	35696	38435	33175	33510
10	1641	1359	1225	1196	867	1897	1054	11/4	1351	1170
31	2/8	294	218	603	418	603	396	329	2/1	230
40	205	282	208	202	372	431 201	301	429	309	294
01 76	254	120	200	227	284	201 212	305	250	200	199
/0 ===	204	, , ======	,, ========	21/	105	215 === = ===	97 ======	/ 1	2/1 ======	=====
	Effluen	nt, Eff	ective s	and st	ize 0.5m	um, Flow	rate 4	m/h		
1	39636	40779	38632	38671	28270	30594	26871	21453	29117	33943
16	1617	1791	630	1485	1208	1269	1824	182	1625	1556
31	906	270	424	634	208	337	274	475	445	457
46	274	280	341	244	311	191	233	487	299	327
61	0	213	116	230	167	162	184	169	203	225
/6 ===	211 =======	95 === == =	213 =======	118 == = ===	55 ===== =	87 ========	79 	162 =======	154 -	205
•	Backwas	h Metho	od 2							
	Water,	Effect	ive sand	size	0.5mm,	Flow rat	te 4m/h			
1	3275	2522	43/3	2482	2029	2049	705	1789	839	1535
16	148	/5	146	/1	5/	134	0	53	57	91
31	0	0	30	32	100	0	55	0	45	0
40	0	126	0	26	0	0	20	83	0	0
o⊥ 76	126	126	53	26	0	41	39 0	0	0	0
====	======	======	=======	======	=======	========		=======		=====
1	Effluen 863	t, Effe	ective sa	and ai	ze 0.5m	m, 4m/h	3000	706	1202	3034
16	505	0C2 F N	177	100 67	2017 N	160	2002 A	750	100	5054
1	0	187	- · · / 0	307 30	0	170	n 0	79	61	0
6	õ	138	õ	0	116	1, 5	116	0	0 0	95
51	91	138	Ő	õ	30	ñ	0	Ő	õ	0
² 6	0	0	35	· 0	32	Õ	õ	45	õ	43
	-	-		-	-206	_	Ũ		5	

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Table E.6:Raw data of control experiments from methods two and three

Appendix F: Proof of Total sum of squares

From the estimates of ε_i as calculated by:-

$$e_i = y_i - y_i^1$$
, for $i = 1, 2, 3...n$

the value of Σe_i^2 can be calculated. If Σe_i^2 is small, the regression represents a well fitting line. However, how is large and small quantified?

The total sum of squares, $\Sigma(yi - y)^2$, may be partitioned into two components: the sum of the squared residuals, Σe_i^2 ; and the regression sum of squares, $b^2 \Sigma (x_i - x)^2$.

Consider
$$\Sigma e_i^2 = \Sigma (y_i - a - bx_i)^2$$

 $= \Sigma (y_i - y + bx - bx_i)^2$
 $= \Sigma [(y_i - y) - b(x_i - x)]^2$
 $= \Sigma [(y_i - y)^2 - 2b(x_i - x)(y_i - y) + b^2(x_i - x)^2]$
 $= \Sigma (y_i - y)^2 - 2b\Sigma (x_i - x)(y_i - y) + b^2\Sigma (x_i - x)^2$

but b = $\frac{\Sigma(x_{1} - x)(y_{1} - y)}{\Sigma(x_{1} - x)^{2}}$

Therefore $\Sigma e_i^2 = \Sigma (y_i - y)^2 - b\Sigma (x_i - x)(y_i - y)$

$$\Rightarrow \qquad \Sigma(y_{i} - y)^{2} = \Sigma e_{i}^{2} + b\Sigma(x_{i} - x)(y_{i} - y)$$

$$\Rightarrow \qquad \Sigma(y_{i} - y)^{2} = \Sigma e_{i}^{2} + b^{2} \Sigma(x_{i} - x)^{2}$$

This can be expressed as:-

Total sum of squares = Residual + Regression sum of squares.

The residal sum of squares measures the variation about the line, and the Regression sum of squares measures the variation due to the regression, as explained in section 6.5.