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# **Evaluation of sludge treatments for pathogen reduction**



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**EVALUATION OF SLUDGE TREATMENTS FOR  
PATHOGEN REDUCTION – FINAL REPORT**

**STUDY CONTRACT NO B4-3040/2001/322179/MAR/A2**

**FOR THE EUROPEAN COMMISSION  
DIRECTORATE-GENERAL ENVIRONMENT**

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September 2001**

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Report No.: CO 5026/1

September 2001

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## SUMMARY

The recycling to agricultural land is an important outlet for sewage sludge and other sludges but it must be controlled in order as to obtain agricultural benefit from the sludge whilst protecting human and animal health and the environment at large. Current practices in Europe are based on the requirements of the 1986 Directive on the use of sewage sludge in agriculture (86/278/EEC). Since that time new technologies have become available for sludge treatment, more pathogens associated with the food chain have been identified and the concerns of the public relating to acceptable risk have changed.

This report identifies for a number of different types of sludges those pathogens that may present a risk to human, animal or plant health. It considers the factors in sludge treatment processes, particularly the newer processes, that influence lethal effects on pathogens, and makes recommendations for the parameters to be applied to these processes to produce treated sludges that present minimum risk.

The constraints on the use in agriculture of sludges that have not been subjected to advanced forms of treatment are discussed.

Proposals are made for the measures to be adopted for the quality assurance of treated sludge and for the validation of treatment processes and new treatment plants as follows:

### Quality assurance

It is not practicable to monitor treated sludge for the presence of pathogens and surrogates should be used for routine evaluation of treatment plant performance and sludge quality.

Surrogates should be organisms commonly found in sludges that have similar resistance to treatment as pathogens. *E. coli* (or enterococci) and *Clostridium perfringens* are suggested. Numbers of *E. coli* should not exceed 1000 per gram (dry weight) and it is tentatively recommended that spores of *C. perfringens* should not exceed 3000 per gram (dry weight).

Because of the diversity of microorganisms and hosts it is not possible to suggest suitable surrogates for systems treating vegetable waste. However, a treatment plant designed to hygienise sewage sludge or sludges of animal origin will also destroy plant pathogens.

### Validation of new processes and new treatment plants using advanced processes

New treatment processes and plants will need to be validated.

A reduction of at least 4 log<sub>10</sub> of added *Salmonella* and the destruction of viability of *Ascaris* ova is suggested for plants treating sewage sludge or animal waste by advanced processes.

### Conventional treatment processes

These processes should achieve at least a 2 log<sub>10</sub> reduction in pathogen numbers as evidenced by reduction in numbers of *E. coli*. Use of conventionally treated sludge on the land must be restricted according to the controls set out in the report to further reduce the risk of pathogen transmission.



## 1. INTRODUCTION

### 1.1 Background

The Waste Management Unit of DG Environment of the European Commission is currently revising the provisions contained in Directive 86/278/EEC on the protection of the environment, and in particular the soil, when sewage sludge is used in agriculture.

Since the first third of the twentieth century it has been the practice for potable water treatment to use a dual-barrier approach of processes that clean the water to a high degree followed by disinfection to ensure that the water is microbiologically safe. In the last quarter of that century a somewhat similar approach has been generally adopted for the hygienisation of sewage sludge to be applied to land as a fertiliser. Firstly the sludge has been stabilised, frequently by mesophilic anaerobic digestion, which considerably reduces, but does not eliminate, the pathogen load and then the risks have been further reduced by the application of constraints on the use or harvesting of subsequent crops.

The rate of pathogen die-off during the period of constraint is influenced by a number of factors. Some of these are unpredictable, e.g. sunlight, temperature and moisture. In practice this dual-barrier approach has worked well but has been difficult to police, particularly with respect to casual access to land by persons pursuing recreational activities. Also, wildlife has not been protected.

There is little evidence of disease in man or animals arising from land application of sludge. The few documented cases have occurred when local regulations or codes of practice have not been observed. However, generally the source of isolated cases of infection are not investigated or identified.

During the last decade there has been an increased public awareness of the importance of food hygiene and the food chain has been identified as a major route of transmission of some of the more recently acknowledged pathogens, e.g. *Escherichia coli* type O157. Current thinking, therefore, is to subject sludges to more stringent treatment processes such that the level of pathogens in material applied to land does not increase the numbers already in the environment.

It is envisaged that the scope of the current Directive will be extended to:

- a) All sludges suitable for being used on land; and
- b) Outlets such as silviculture, green areas and land reclamation.

In particular it is proposed to have a description of the technical characteristics of the treatment processes in order to ensure a sufficient degree of hygienisation for those sludges whose use would be unrestricted.

## 1.2 **Objectives**

The objective of the study is to collect and evaluate information available in the literature on pathogens in sludge (bacteria, viruses, parasites, fungi and yeast) and on the processes capable of achieving sludge hygienisation rendering it suitable for unrestricted use.

The use-restrictions needed to minimise the risk from sludge that has been subjected to less demanding treatment processes will also be evaluated. Accounts will be taken of the different climatic conditions and availability of technology throughout the Community.

The study will:

- Identify pathogens (bacteria, viruses, parasites, fungi and yeast) likely to be present in different types of sludge (sewage sludge; septic tank sludge; sludges from meat processing waste; sludges from vegetable processing; tannery sludges; paper sludge; and similar materials);
- Evaluate the parameters (residence time, temperature, pH, moisture etc.) that make sludge treatment processes effective in killing/inactivating pathogens in sludge and assess the need for treatment, according to the different types of sludge;
- Evaluate the use restrictions (e.g. deep injection, ploughing, and grazing and harvesting) for sludge treated to a lesser degree of hygienisation;
- Indicate which micro-organisms could be routinely used as indicators of sludge hygienisation and which micro-organisms could be used to validate a process during its start-up phase.

The results of this study will assist the Commission in proposing the appropriate combination of treatments, use restrictions and pathogen control for safe spreading of sludges in the Community.

## 2. IDENTITY OF PATHOGENS

Pathogens, in the main, are parasites in the wide sense of the word. They derive their nutrients from the products of their host's metabolic system. They have little, if any, beneficial effect to the host and their excretions are generally toxic to the host.

The control of pathogens in man and animals is important to medical and public health professionals. They are of economic importance to the food and pharmaceutical industries and they have been studied widely in the laboratory. Consequently there is a lot of information on their destruction or survival in relatively simple media under defined conditions but their behaviour in complex media or ecosystems is less easy to predict.

### 2.1 Sewage sludge

#### 2.1.1 Sources of pathogens

Sewage sludge can vary in the quality and quantity of its organic and inorganic content, including the types and numbers of pathogens it contains. Such variations occur not only geographically but also over time at the same site.

The nature and concentrations of pathogens in sewage will depend on the health and the size of the population in the catchment. In a large community a small proportion of the population is likely to be infected at any time, and consequently low levels of pathogens will be in the sewage at all times. In a small community, the excreta of a few people who may or may not be infected, represents a large part of the sewage input and in these circumstances the sewage will contain either no pathogens or relatively high concentrations.

The nature of sewage, and hence its sludge, is such that it may contain enteric pathogens, i.e. those which are excreted with faecal material and generally are infective by the oral route. Man is obviously the most likely source but, depending upon local conditions, the sewage may also contain excreta from pets - arising from storm runoff - or from farm animals. The levels and diversity of pathogens in sewage will be dependent upon local conditions, including the incidence of disease in the contributing community at that time.

Pathogens that are excreted by the respiratory route are unlikely to be present in sewage sludge in large numbers although some respiratory viruses are excreted with faeces. Pathogens, which cause skin infections, are unlikely to be present in large numbers.

Washings from vegetable preparation, both domestic and commercial, are the major source of plant pathogens although other routes are possible such as storm run-off from paved and roof areas following aerial deposition of the micro-organisms.

#### 2.1.2 Types of pathogens in sludge

Bacteria are capable of independent existence and can multiply wherever suitable conditions are present. Most are inactivated at temperatures in excess of 70°C over a relatively short period of time. Lower temperatures over longer time periods are equally effective. However, some that produce spores, e.g. *Clostridium* species, require higher temperatures for complete kill. The bacterial pathogens of mammals have optimum growth temperatures around the body

temperature, which is 35-40°C. They are unlikely to multiply rapidly at temperatures below about 25°C. For multiplication they also need suitable nutrients and water levels. As the natural habitat of these organisms is the intestine, nutrients will not be lacking in sewage sludges. Conditions unsuitable for growth are not necessarily lethal to micro-organisms. Some organisms have further evolved structures - cysts or spores - as a means of survival in adverse conditions.

Similar comments can be made about the bacterial pathogens of plants, but the temperatures for optimum growth and at which growth is inhibited are lower than for mammalian pathogens.

Viruses are unable to multiply outside of the living cells of their host, but they can survive in adverse conditions.

There is limited evidence linking wastewater workers to respiratory tract viral illnesses such as colds and influenza. In these cases the route of infection has probably been through the inhalation of aerosols during sludge handling or processing.

Worms, e.g. *Taenia* spp. and *Ascaris* spp., have evolved an egg stage as a means of transferring from one host to another. Pathogenic protozoa e.g., *Cryptosporidium* and *Giardia* have evolved a cyst stage for the same purpose. These structures are extremely resistant to the stresses of the ambient environment. However, these organisms cannot reproduce outside a suitable host and this means that their concentrations in sludges are relatively low.

A number of workers, notably Strauch (1998) and the US Environmental Protection Agency (US EPA, 1999), have collated the pathogens that could be present in sewage sludge and these are summarised in Table 2.1.

This list is comprehensive and the risk from some organisms is very small. For example, in waters with a high organic content *Shigella* spp. will only survive a few days and *Vibrio cholera* will not be detected after about a week. On the other hand, some serovars have in recent years been demonstrated to pose a greater risk than most members their genera. The emergence of *E. coli* O157 is of particular importance in this context. The development of multiple resistances to antibiotics by some salmonellae may present problems in the treatment of infections arising from sewage sludge and similar wastes.

The relative importance of an agent will depend upon a number of factors such as the numbers of organisms present, the prevalence in the local community and the ease with which the disease may be treated. In the case of food animals or crops there may also be economic implications. In sewage sludge and similar wastes most of the pathogens will be in hazard category 2 (Directive, 1993), but a few, *Salmonella typhi*, *E. coli* O157 and some mycobacteria, for example, have a higher rating of category 3.

In practice the evaluation of sludge treatment processes can be narrowed down to those organisms that are prevalent and likely to cause infection, and those pathogens that are more resistant to hygienisation processes.

The dangerous properties of some pathogens arise from their ability to produce stable exotoxins e.g. *Clostridium botulinum*. Advanced and Conventional sludge treatment systems produce a stabilised final product where such organisms are unlikely to multiply to large numbers and there are unlikely to be problems arising from bacterial toxins during sludge storage or application.

**Table 2.1 Pathogenic micro-organisms that may be found in sludge derived from faecal material**

<p><b><u>Bacteria</u></b></p> <p><i>Salmonella</i> spp.  <i>Shigella</i> spp.  <i>Escherichia coli</i> (enteropathogenic strains)  <i>Pseudomonas aeruginosa</i>  <i>Yersinia enterocolitica</i>  <i>Clostridium perfringens</i>  <i>Clostridium botulinum</i>  <i>Bacillus anthracis</i>  <i>Listeria monocytogenes</i>  <i>Vibrio cholera</i>  <i>Mycobacterium</i> spp.  <i>Leptospira</i> spp.  <i>Campylobacter</i> spp.  <i>Staphylococcus</i>  <i>Streptococcus</i></p> <p><b><u>Viruses</u></b></p> <p>Poliovirus  Coxsackievirus  Echovirus  'New' enterovirus  Adenovirus  Reovirus  Hepatitis A-virus  Rotavirus  Astrovirus  Calicivirus  Coronavirus  Norwalk-like calicivirus  Small round viruses  Parvovirus  Adenoassociated viruses  Influenza virus</p>	<p><b><u>Protozoa</u></b></p> <p><i>Entamoeba histolytica</i>  <i>Giardia lamblia</i>  <i>Toxoplasma gondii</i>  <i>Sarcocystis</i></p> <p><b><u>Helminths</u></b></p> <p><i>Taenia saginata</i>  <i>Taenia solium</i>  <i>Diphyllobothrium latum</i>  <i>Echinococcus granulosus</i>  <i>Ascaris lumbricoides</i>  <i>Ancylostoma duodenale</i>  <i>Toxocara canis</i>  <i>Toxocara cati</i>  <i>Trichuris trichura</i></p> <p><b><u>Yeast</u></b></p> <p><i>Candida albicans</i>  <i>Candida krusi</i>  <i>Candida tropicalis</i>  <i>Candida guilliermondii</i>  <i>Cryptococcus neoformans</i>  <i>Trichosporon</i></p> <p><b><u>Fungi</u></b></p> <p><i>Aspergillus</i> spp.  <i>Aspergillus fumigatus</i>  <i>Phialophora richardsii</i>  <i>Geotrichum candidum</i>  <i>Trichophyton</i> spp.  <i>Epidermophyton</i> spp.</p>
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## **2.2 Septic tank sludge**

Those wastes entering septic tanks are essentially the same as those entering sewage treatment plants in that a large component, in microbiological terms, is derived from faecal material. The genera of pathogens that may be present will be similar to those listed in Table 2.1. Because septic tanks are usually associated with individual families or similar closely related communities the range of pathogens present at any one time will be small. However, it is likely that if one individual becomes infected the whole community will be infected. Therefore the concentration of pathogens will either be very low or relatively high.

### **2.3 Waste from meat processing**

The waste from meat processing will arise from abattoirs and related establishments. In practice the waste can be considered as two streams. The material arising from the herding, slaughter and initial dressing of the carcasses, and that arising during the butchery activities to reduce the carcass to manageable pieces.

The main risk to animals and humans from the ultimate return of these materials to land will be from the intestinal contents and will consequently contain a similar range of organisms to sewage sludge. Animals are normally kept in herds, which, by human standards, have low levels of hygiene. Consequently the level of infection in a single herd is either low or high, with a single or low number of species of pathogens. Animals from many herds pass through an abattoir in a short space of time so the range of pathogens is potentially broad.

The waste produced during the cutting of carcasses may possibly contain the prions of Bovine Spongiform Encephalopathy (BSE). Regulations relating to the butchery techniques to be used, the age of animals allowed to enter the food chain, and specific requirements for the treatment of this type of waste (e.g. Directive 90/667/EEC and related regulations) should minimise the numbers of prions released into the environment. At present little has been confirmed on the survival of prions, the infective dose or routes and sources of infection.

### **2.4 Tannery waste**

The chemicals and other treatments used during the processing of hides effectively disinfect the waste, with the possible exception the anthrax bacillus, *Bacillus anthracis*. Anthrax infection in tannery workers has occurred in the past after contact with infected hides. There is no evidence to implicate sludge in any cases of infection in the last forty years. Anthrax in farm animals and man is now extremely rare. Regulations are in place to minimise risks from cross-border shipments of hides, for example, Directive 92/118/EEC and related Directives.

### **2.5 Vegetable processing waste**

It is extremely unlikely that pathogens of humans and animals would be present in waste from vegetable processing. Some micro-organisms that are pathogenic to plants are so widespread that any reduction in numbers would have a minimal effect on the total environmental load. However there are plant pathogens that are of concern to commercial growers of vegetables. These include, amongst others, the potato cyst nematode, the rhizomania disease of beet, and brown rot of potatoes. To some extent the spread of these diseases can be minimised by local codes of practice, such as those in place in the UK which restrict the amount of soil taken into the processing plants, and require as far as possible the returning of the soil to the field of origin.

Plant pathogens generally have low optimum growth temperatures and their lethal temperature will be lower than those of mammalian pathogens. Consequently the conditions required to hygienise such waste will be less stringent than those required for human or animal waste.

There is a wide diversity of micro-organisms that are pathogenic to plants most of which are specific to a narrow range of hosts. Although they are of economic importance it is extremely difficult to treat plants for microbiological infections and consequently the amount of

information relating to their characteristics is considerably less than for organisms of public health significance. Bohm *et al.*(1999) have identified the more important organisms and these are listed in Table 2.2.

**Table 2.2 Pathogens that may be present in vegetable waste**

	Typical host		Typical host
<b><u>Bacteria</u></b>		<b><u>Fungi</u></b>	
<i>Xanthomonas campestris</i>	Cabbage	<i>Plasmodiophora brassicae</i>	Cabbage
<i>Pseudomonas marginalis</i>	Salad	<i>Phoma apiicola</i>	Cabbage
<i>Pseudomonas phaseolicola</i>	Beans	<i>Peronospora brassicae</i>	Celery
<i>Pseudomonas lacrimans</i>	Cucumber	<i>Peronospora spinaciae</i>	Spinach
<i>Corynebacterium michiganense</i>	Tomato	<i>Peronospora destructor</i>	Onion
<i>Corynebacterium sepedonicum</i>	Potato	<i>Marssonina panattoniana</i>	Salad
<i>Erwinia phytophthora</i>	Potato	<i>Sclerotinia minor</i>	Salad
<i>Agrobacterium tumefaciens</i>	various	<i>Botrytis cinerea</i>	Salad
		<i>Bremia lactucaae</i>	Salad
		<i>Cercospora beticola</i>	Turnip
		<i>Aphanomyces raphani</i>	Radish
		<i>Alternaria porri</i>	Carrot
		<i>Septoria apii</i>	Celery
<b><u>Viruses</u></b>		<i>Turburcinia cepolae</i>	Onion
Potato virus X	Potato	<i>Sclerotium cepivorum</i>	Onion
Potato virus Y	Potato	<i>Botrytis allii</i>	Onion
Aucuba-virus	Potato	<i>Uromyces appendiculatus</i>	Bean
Tobacco ring-spot virus	Potato	<i>Mycosphaerella piodes</i>	Peas
Rattle-virus	Potato	<i>Ascochyta pinodella</i>	Peas
Tabocco mosaic virus	Tomato	<i>Eryiphe polygoni</i>	Peas
Horse bean mosaic virus	Beans, Peas	<i>Cladosporium cucumernum</i>	Cucumber
Pea mosaic virus	Beans	<i>Sclerotinia sclerotiorum</i>	Cucumber
Bean mosaic virus	Beans	<i>Septoria lycopersici</i>	Tomato
Yellow bean mosaic virus	Beans	<i>Alternaria solani</i>	Tomato
Cauliflower mosaic virus	Cabbage spp.	<i>Didymella lycopersici</i>	Tomato
Cucumber mosaic virus	Cucumber	<i>Rhizoctonia solani</i>	Potato
Aucuba mosaic virus	Cucumber	<i>Phyophthora infestans</i>	Potato
Cabbage ring spot virus	Cabbage spp.	<i>Synchytrium endobioticum</i>	Potato
Lettuce mosaic virus	Salad	<i>Verticillium albo-atrum</i>	Potato
Beet mosaic virus	Beet		
Onion mosaic virus	Onion		

## **2.6 Paper processing sludges**

These wastes are inherently composed of poorly biodegradable cellulose and lignin. They can be regarded as free of pathogens and present no risk to the health of man or animals.

### 3. PARAMETERS AFFECTING THE KILLING OR INACTIVATION OF PATHOGENS

The survival of micro-organisms is a function of a number of factors, which include the local temperature, local antagonistic conditions, and the characteristics of their species or genera. The lethal effect on the population of specific micro-organisms is a gradual one and generally the kinetics follow approximately the exponential law of disinfection (Chick's law);

where the surviving fraction,  $X_t/X_o$ , of the original population  $X_o$ , after the time interval  $(t-t_0)$ , is given by

$$X_t/X_o = e^{-k(t-t_0)}$$

when  $k$  is the specific decay rate.

#### 3.1 Heat

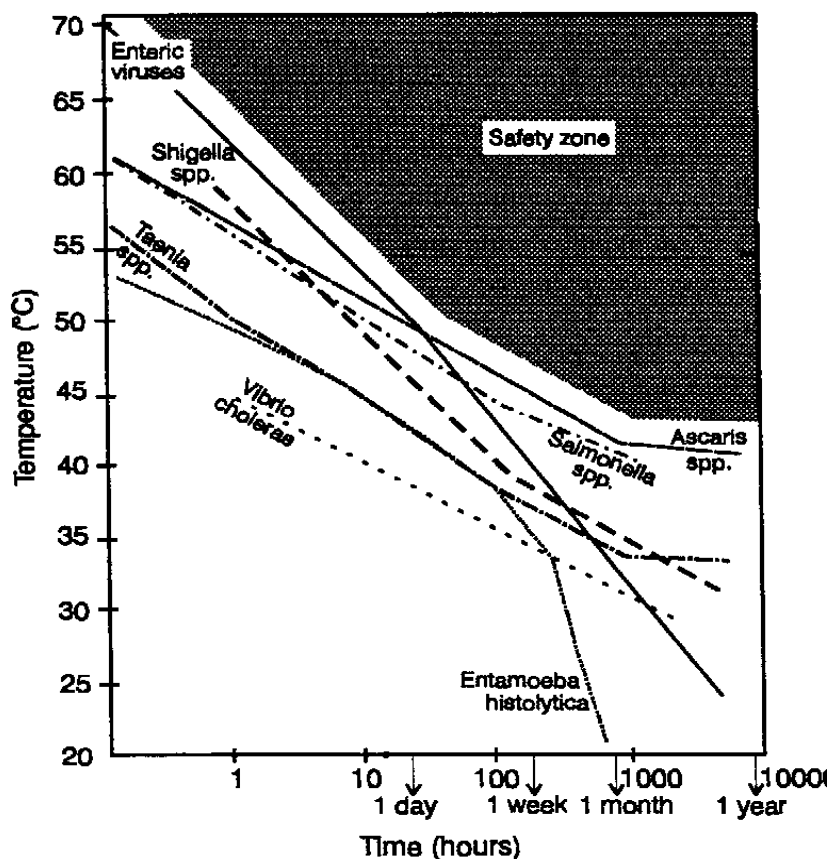
##### 3.1.1 Thermophilic temperatures

Pathogens are inactivated during exposure to heat, which to be effective and relatively rapid must be above their optimum growth temperature. The period of exposure is dependent on the temperature and on the species of the organism. Feacham *et al.* (1983) determined the thermal death curve of a number of enteric pathogens. These have been collated into a single graph by Strauch (1991 and 1998), which indicates a 'safety zone' where, if the operating parameters were above the minimum requirements, the resultant sludge would be virtually pathogen-free (Figure 3.1).

These data suggest that the operating parameters should be in excess of 7 minutes at 70°C; 30 minutes at 65°C; 2 hours at 60°C; 15 hours at 55°C or 3 days at 50°C.

A recent review for UK government departments of reported practical studies, much of which were carried out as part of the COST 681 programme in the 1980's, concluded that 70°C for 30 minutes or 55°C for 4 hours would produce a sludge that is virtually pathogen-free (Carrington *et al.*, 1998).

The differences in the suggested exposure times arise from differences in the design of the two studies. The data of Strauch was based studies using pure cultures of the micro-organisms, whereas the studies reported by Carrington allowed sufficient time to ensure the efficient warming of the sludge and also took into account the movement of the sludge through a continuous flow system. The UK workers thought that 30 minutes was the minimum residence time that would ensure that all sludge had been exposed to lethal time-temperature conditions.



**Figure 3.1** Time-temperature requirements to produce sludge, which is virtually pathogen-free

The US EPA have produced formulae to determine the time-temperature regimes for thermally treated "Class A" sludge, (US EPA, 1999). These are dependent upon the solids content of the sludge and are:

If the solids content of the sludge is 7% or more

$$D = (131.7 \times 10^6) / 10^{0.14t}$$

where D is time in days and t is temperature in °C. Contact time should be for not less than 30 minutes and the temperature should be not less than 50°C. If the sludge particles are small and are heated by hot gas or immiscible fluid the heating time should be not less than 15 seconds;

If the solids content of the sludge is less than 7% the exposure is calculated from

$$D = (50.07 \times 10^6) / 10^{0.14t}$$

Contact time should be for not less than 30 minutes and the temperature should be not less than 50°C.

Whilst the formulae are in agreement with the graph of Strauch at the higher temperatures there is a wide divergence at lower temperatures. For sludges of less than 7% solids the US EPA time-temperature conditions are: 30 minutes at 67°C; 1 hour at 65°C; 4.8 hours at 60°C; 1 day at 55°C; and 5 days at 50°C. For sludges with a high solids content the requirements range from 30 minutes at 70°C to 13 days at 50°C.

The differences seem to arise because the US EPA developed a linear model, using a logarithmic scale for time, and incorporated a margin of safety, whereas it can be seen from the work of Feacham *et al.* and of Strauch (Figure 3.1) that the boundary of the safety zone is an irregular curve influenced by different sensitivities of different micro-organisms and is not directly proportional to either time or temperature.

### 3.1.2 Mesophilic temperatures

The sludge treatment processes that operate at mesophilic temperatures (33-37°C) were developed for the stabilisation of sludge. The mean retention time in such plants is usually in the region of 12-15 days, but to enable the process to operate on a continuous basis there must be an addition of fresh material in the form of raw sludge, normally at least once every day. For maximum effect of the reduction of numbers of pathogens the removal of treated sludge must take place before the addition of raw sludge. This still means, however, that a fraction of the withdrawn sludge will have been exposed to the digesting conditions for 24 hours or less. The implications of mean retention time are discussed later.

It can be seen from Figure 3.1 that prolonged exposure to mesophilic temperatures will have an effect on the survival of pathogens. The simple effect of temperature will be enhanced by the acids and other antagonistic compounds developed during the digestion process. Studies carried out in several European countries, and reported to the COST 681 programme and elsewhere, indicate that reduction in numbers of pathogens of 2-3 log<sub>10</sub> can be achieved (Carrington *et al.*, 1998). However because different micro-organisms have different survival times at these temperatures and because the pathogen load in the local population varies with their state of health it is not possible with mesophilic systems alone to produce a sludge that is virtually free of pathogens consistently.

### 3.1.3 Ambient temperatures

The survival of pathogens at ambient temperatures is a function of their phyla, local conditions and the nature of the media in which they are suspended. The data presented in Figure 3.1 suggests that whilst the cysts of *Entamoeba histolytica* would survive about one month and enteric viruses may survive about one year in temperatures that may be experienced in Mediterranean countries, the ova of *Ascaris* would persist for a considerable period. In conditions where the daily temperatures are considerably lower the survival periods will be considerably longer.

The main processes for the reduction of pathogen numbers during sludge treatment at ambient temperatures, particularly in colder climates, will be the production of fatty acids and other anti-microbials during the stabilisation of the sludge, and the activity of the micro-flora,

such as protozoa. Because of the many uncontrollable variables it would never be possible to produce sludge that was virtually pathogen free.

The US EPA (1989) did sanction an ambient process for air drying sludge to their "Class B" standard, but it had several constraints. The beds had to be underdrained, the sludge had to be at least partially digested before being applied to the beds and applied to a depth not exceeding 23cm. The minimum drying period was 90 days and the average daily temperature had to be above 0°C for 60 days during which the beds had to be exposed (i.e. not covered with snow). They estimate that the pathogen reduction would be approximately 1 log<sub>10</sub>.

### **3.1.4 Thermal drying**

In recent years there have been a number of developments in the field of thermal drying of sludges. Variations of the process include convection dryers, contact dryers and vacuum dryers. Sludge is dried to a water content of less than 10% at a temperature in excess of 80°C. The data in Figure 3.1 indicate that a short period, i.e. less than 10 minutes, at this temperature should be lethal to most pathogens. The hygienisation of the sludge will be further enhanced by the reduction in water content, which will inhibit the re-growth of bacteria. However there is little peer-reviewed evaluation of these processes in terms of pathogen control, and there is some evidence that a too rapid reduction of water content could reduce the sensitivity of organisms to heat treatment.

## **3.2 Moisture**

In general simple drying is not an effective means of minimising the pathogen content of sludges, although reducing the moisture content can enhance the effects of other treatments. Drying at ambient temperatures may be practicable in the Mediterranean areas of the European Community. To be most effective the initial reduction in water content should be carried out relatively slowly. Most micro-organisms cannot grow in substrates where the water activity has been reduced to below 0.9 (Singleton and Sainsbury, 1987). Water activity ( $A_w$ ) is a measure of the water available to the microorganisms and equates to 1/100th of the relative humidity within the sludge particle and not the water content of the body of the sludge. In practice it is very difficult to measure water activity in sludge particles and is not a suitable parameter for the routine monitoring of sludge from treatment processes such as thermal drying.

## **3.3 pH**

### **3.3.1 Slaked lime**

The raising of the pH of sludge to at least pH 12 by the use of slaked lime has the effect of suspending microbiological activity and reducing the hazard from pathogens by 2 or more log<sub>10</sub>. Carrington *et al.* (1998), reviewing the work of others suggest a pH of 12 for 2 hours will reduce many pathogens to insignificant numbers. Because the activity of other organisms is only suspended, offensive odours may be produced, and such sludge should be utilised within two days. Strauch (1998) suggests that the sludge should be raised to pH 12.6 and stored for three months.

### 3.3.2 pH and temperature

When quicklime or similar substances, (e.g. pulverised fuel ash or cement kiln dust), are mixed with sludge, in addition to the raising of the pH there is an autothermic effect and the temperature of the mixture is raised to between 55°C and 70°C. Strauch (1998), describing earlier studies found that a combination of pH 12 and temperatures in the range 60-70°C destroyed *Ascaris* ova and a range of viruses within 24 hours.

### 3.4 Residence time

In thermophilic processes the requirements for sludge stabilisation mean that the digestion process will take several days, while the inactivation of pathogens can be achieved by a minimum of a few hours per day at elevated temperatures. Therefore in the thermophilic processes the duration of the residence time has negligible effect upon the pathogen kill.

Other stabilisation processes, although working at relatively low temperatures, produce products that are deleterious to the survival of pathogens. For example, mesophilic anaerobic digestion operates at about 35°C, which is the optimum growth temperature for most enteric bacteria, but the digestion process produces amounts of fatty acids and other products that are lethal to many pathogenic organisms.

The time-temperature exposure conditions considered earlier hold good for batch reactors or plug flow reactors. For intermittently fed reactors where there is a frequent influx of new material and a corresponding outflow (e.g. digesters) the exposure conditions may need to be amended. The fraction of the pathogens in the feed surviving and passing into the outgoing sludge can be calculated from

$$C/C_o = R / (10^{DR_\theta} - 1 + R)$$

where  $C/C_o$  is the fraction surviving,  $R$  is the fraction of the reactor content replaced each day,  $R_\theta$  is the interval between feeds (expressed as days) and  $D$  is the decimal decay rate.

An example of the reduction in numbers of a hypothetical pathogen during mesophilic primary digestion is given in Table 3.1. For this example it has been assumed that the concentration of the pathogen in the sludge is consistent and that it has a decimal decay rate of 1 d<sup>-1</sup>, which is an arbitrary figure but not unrealistic. The calculations have been made for situations using three different intervals between feeding and for two different mean retention periods.

**Table 3.1 Examples of log<sub>10</sub> reduction of numbers of a hypothetical pathogen during a range of mesophilic digestion situations**

Interval between feeds (hours)	Mean retention period of digester	
	12 days	15 days
2.4	0.61	0.69
24	2.04	2.10
72	4.01	4.18

These figures show that increasing the retention period does not proportionately increase the kill.

If it is assumed that the decimal decay rate at ambient temperatures is  $0.1 \text{ d}^{-1}$  sludge from Table 3.1 would undergo a further reduction of about  $2.5 \log_{10}$  if it was subjected to secondary digestion in a completely mixed batch reactor for 14 days. However, if additions and withdrawals were made on a daily basis during the secondary digestion period the reduction in numbers would be about  $0.6 \log_{10}$ . However conditions during secondary digestion will vary considerably both geographically and with time. The effective decay rate will be dependent upon the temperature and the activity within the sludge. The continuing digestion will produce fatty acids and other material deleterious to the pathogens, but for this to have maximum effect the sludge would have to be continuously stirred. Secondary digestion is normally carried out at ambient temperatures in unstirred tanks.

### **3.5 Other forms of sludge treatment**

At the present time the most cost-effective forms of sludge treatment are the use of heat. In most cases this may be because the technology is already well established. Other processes may be developed in the future; irradiation has a high potential. The US EPA have approved irradiation as a means of producing "Class A" sludge and quite a lot of research has been carried out in Europe (e.g. IAEA, 1975). However, because of the high costs and the perception of risk by the public it is unlikely to be accepted in the near to medium term. Lime treatment is already well established and other chemical treatments may be developed. However such methods are likely to increase the total volume of treated sludge with consequential problems of transport, storage and the need to increase application rates to achieve similar fertilising value to heat-treated sludges.

## **4. TREATMENT NEEDS OF DIFFERENT SLUDGE TYPES**

### **4.1 Treatment needs**

There are two aspects to sludge treatment; the reduction of putrescibility, i.e. stabilisation, and the reduction of the levels of pathogens, i.e. hygienisation. These may be achieved in a single process, e.g. thermophilic digestion, or as a two-stage process, such as pasteurisation followed by digestion.

The DG Environment Waste Management Unit in considering revisions to the Directive relating to the land application of sludges (Directive, 1986) is suggesting two possible levels of treatment; Advanced and Conventional. The Advanced treatments will reduce the pathogen content to insignificant levels. "Insignificant levels" can be considered to mean where there is minimal risk to human, animal and plant life in the context both of farming and of natural ecosystems.

Several approaches could be used to define the quality of sludge after treatment. However, treatment methods must be understood by, and not be excessively onerous on, the sludge producers. The principle adopted in these recommendations is to not increase the risk when applied to land by reducing the pathogen content of sludges to those of the ambient surroundings. To be adequately monitored the quality of the treated sludge needs to be defined and in practice this means setting limits for the numbers of certain representative organisms that may be present.

### **4.2 Sewage sludge**

#### **4.2.1 Sludge treatment**

Sewage sludge by its nature will contain a wide diversity of pathogens, some of which may be present in large numbers. These pathogens have evolved a range of protective characteristics to enable them to survive in the mammalian intestine, and in some cases in the external environment. However it can be seen from Figure 3.1 and from the discussion in section 3 that if the most resistant pathogens are destroyed the less resistant will be also be destroyed. Consequently only the most resistant organisms need to be considered when defining plant-operating parameters. For example, if plants using heat treatment processes operate in the 'safety zone' of Figure 3.1, all pathogens whose heat tolerance is to the left of the shaded area will be destroyed.

The data in Figure 3.1 was derived from many sources where cultures of organisms were exposed to heat in batch conditions and these indicate that if sludge is to be hygienised at 55°C the undisturbed exposure time should be about 15 hours. However, for a sludge to be aesthetically acceptable for land use it must also be stabilised, i.e. it must not putrefy further. Studies have shown (Carrington *et al.*, 1998; US EPA, 1999), that digesting conditions such as fatty acid production at thermophilic temperatures stabilise sludge and enhance the rate of inactivation of pathogens. In a continuously operating, but intermittently fed system, pathogen levels in the withdrawn sludge will be insignificant provided that certain conditions are met. These are; that immediately prior to withdrawal the sludge has been exposed to 55°C or more for a least 4 hours; that sludge is withdrawn before raw sludge is added; and that the mean

retention period of the digester is sufficient to stabilise the sludge. Ideally the contents would be completely mixed during the whole digestion period and the temperature would be recorded near the centre of the system.

It can also be seen from section 3 that to produce sludge that is virtually free of pathogens it must be exposed to thermophilic temperatures; thermophilic temperatures and active digestion; or a combination of high pH and relatively high temperature.

In practice this means:

*If the sludge is to be thermally dried:*

Sludge should be raised to at least 80°C for 10 minutes and moisture content reduced to less than 10%.

*If the sludge is to be treated by moist heat and then stabilised:*

Sludge should be raised to temperature-time conditions inside safety zone e.g.

80°C - 10 minutes

75°C - 20 minutes

70°C - 30 minutes

followed immediately by digestion, which may be at mesophilic temperatures.

*If the sludge is to be hygienised and stabilised by thermophilic (anaerobic or aerobic) digestion:*

Sludge should be raised to at least 55°C for a continuous period of at least 4 hours after the last feed and before the next withdrawal. Plant should be designed to operate at a temperature of at least 55°C with a mean retention period sufficient to stabilise the sludge.

*If the sludge is to be composted, with or without other material:*

Composting should be carried out in batches or in a 'plug-flow' system over a period sufficient to stabilise the sludge and produce an acceptable product. All material should be maintained at a temperature of at least 55°C for at least 4 hours between each turning. The number of turnings for a windrow system should be at least three, but more may be necessary to produce a stabilised compost .

*If the sludge is to be treated with quicklime:*

The sludge and lime should be thoroughly mixed and the pH should be at least 12 and the temperature at least 55°C not less than 2 hours after mixing.

The process requirements for advanced treatment are summarised in Table 4.1

**Table 4.1 Summary of advanced treatments for sewage sludge**

Process	Parameters
<b>Windrow composting</b>	Batches of sludge (+/- bulking agent) to be kept at 55 <sup>0</sup> C for 4 hours between each of 3 turnings, followed by a maturation period to complete the composting process.
<b>Aerated pile and in-vessel composting</b>	The batch to be kept at a minimum of 40 <sup>0</sup> C for at least 5 days and for 4 hours during this period at a minimum of 55 <sup>0</sup> C. This to be followed by a maturation period to complete the composting process.
<b>Thermal drying</b>	The sludge should be heated to at least 80 <sup>0</sup> C for 10 minutes and moisture content reduced to < 10%.
<b>Thermophilic digestion (aerobic or anaerobic)</b>	Sludge should achieve a temperature of at least 55 <sup>0</sup> C for a minimum period of 4 hours after the last feed and before the next withdrawal. Plant should be designed to operate at a temperature of at least 55 <sup>0</sup> C with a mean retention period sufficient to stabilise the sludge.
<b>Heat treatment followed by digestion</b>	Minimum of 30 minutes at 70 <sup>0</sup> C followed immediately by mesophilic anaerobic digestion at 35 <sup>0</sup> C with a mean retention time of 12 days.
<b>Treatment with lime (CaO)</b>	The sludge and lime should be thoroughly mixed to achieve a pH value of at least 12 and a minimum temperature of 55 <sup>0</sup> C for 2 hours after mixing.

Other forms of sludge treatment, such as mesophilic anaerobic digestion, use of slaked lime, and storage in various forms, will not produce a sludge that can be considered to be free of pathogens.

#### 4.2.2 Sludge quality

The quantity and species of pathogens in sewage sludge can vary considerably both with time and location depending upon local circumstances and the current health of the local population. The published information shows a wide range of concentrations may be present, for example data collated by Davis *et al.* (1999) from a range of sources, is summarised in Table 4.2.

**Table 4.2 Typical concentrations of micro-organisms (wet weight) in untreated sewage sludge**

<b>Bacteria</b>	<i>Escherichia coli</i>	$10^6 \text{ g}^{-1}$
	<i>Salmonella</i>	$10^2\text{-}10^3 \text{ g}^{-1}$
<b>Viruses</b>	Enterovirus	$10^2\text{-}10^4 \text{ g}^{-1}$
<b>Protozoa</b>	<i>Giardia</i>	$10^2\text{-}10^3 \text{ g}^{-1}$
<b>Helminths</b>	<i>Ascaris</i>	$10^2\text{-}10^3 \text{ g}^{-1}$
	<i>Toxocara</i>	$10\text{-}10^2 \text{ g}^{-1}$
	<i>Taenia</i>	$5 \text{ g}^{-1}$

These data support the suggestion of Strauch (1998) that a plant producing hygienised sludge should be able to demonstrate a  $10^4$  reduction in concentration of naturally occurring or added *Salmonella* and that helminth ova are rendered non-viable. The enterovirus classification covers a wide range of types and although in total there may be as many as  $10^4 \text{ g}^{-1}$  in the sludge, a  $4 \log_{10}$  reduction would reduce the concentrations of the individual types to insignificant levels.

### 4.3 Sludge from meat processing

In general the range of pathogens in sludge arising from the treatment of meat processing waste will be similar to those in sewage sludge so the requirements for hygienisation in terms of heat and/or pH will be similar, although the process parameters to achieve stabilisation may differ because the nature of the waste.

There is little published evidence relating to the range and concentrations of pathogens in these sludges. Farming practices keep animals in close contact in herds, which are ideal conditions for the spread of infectious diseases. The use of drugs, to treat infectious diseases or prophylactically, is generally on a herd-wide basis. This means that most animals from the same source are either free of specific diseases or may be infected. Consequently the level and range of pathogens is likely to vary considerably between batches of animals and from day to day.

One literature review, (Carrington, 1997), suggests that the faeces of farm animals contain about 10 fold less *E. coli* than human faeces. However human waste (i.e. sewage) is likely to contain more non-faecal material than the animal waste. In the absence of more definitive information this would suggest that the parameters for sewage sludge treatment in section 4.2.1 are equally applicable to these sludges.

Both sewage sludge and animal processing waste are largely derived from mammalian faecal material and thus the same quality criteria for treated sludges (section 4.1.2) should be applied to both forms of sludge.

Bovine Spongiform Encephalopathy (BSE) is a neurodegenerative disease of cattle, colloquially known as 'mad cow disease'. The infective agent is a prion that is found in high concentrations in the brain and spinal chord of infected animals. Little is known about the infective dose or the prion's survival in the environment, but it is extremely resistant to heat. In

the UK and other countries there are controls to eliminate the disease from the cattle population and to prevent animal parts that may contain high concentrations of prions being treated with other waste and entering the food chain. A study sponsored by the Environment Agency of England and Wales (DNV Technica, 1997) of the theoretical risk of BSE to the human population *via* environmental pathways, but not the food chain, concluded that the estimated risk was very low. The maximum individual exposure by this route was estimated to be one-millionth of an infective dose per year.

At present little is known regarding the persistence in the environment or the infective dose of the BSE prion. At a meeting held on 24-25 September 1998 the Scientific Steering Committee of the Commission adopted the opinion that in the absence of more definitive information wastes likely to contain the BSE prion should not be incorporated into organic fertilisers used where cattle may have access. Unless sludge has been treated at the high temperatures known to destroy the prion (moist heat at 130°C (3 bar) for 20 minutes), it would be logical to apply the same advice to sludge applied to land.

#### **4.4 Vegetable processing waste**

The wastes from vegetable processing are unlikely to pose a major risk to human or animal health. However, they are likely to contain micro-organisms that are pathogenic to plants. Generally crops are not treated to eliminate pathogens so such pathogens are widespread in the environment. There are however some serious pathogens that have limited distribution, and it is desirable that these are not allowed to spread further. As an example, the potato cyst nematode is common in some areas where potatoes are grown as a food crop. But it is undesirable to allow it in areas where basic seed potatoes are grown, as not only would it have an undesirable effect on those crops but it may well get carried in the seed tubers to previously uninfected areas.

Because of the wide range of pathogens and the diversity of genera of hosts there are no comprehensive reviews from which a 'safe zone' can be derived in the same way as for mammalian pathogens. However, as plants grow at ambient temperatures it is unlikely that plant pathogens would tolerate temperatures in excess of the upper end of the mesophilic temperature range (about 40°C).

There is anecdotal evidence of human infection with cholera and *E. coli* O157 from handling vegetables originating from South America or Africa that had been irrigated with untreated sewage.

Some of the process described as 'conventional treatment' in the third draft of the discussion document on the revision of the sludge directive (EC, 2000) would be appropriate technology for the treatment of these wastes. In particular the use of lime to raise the pH to 12 and to maintain the pH at that level for 24 hours. If sufficient organic material were present, mesophilic anaerobic digestion at 35°C with a mean retention period of 15 days is a possibility, as is composting with the temperature above 40°C.

If the vegetables originate outside of the EC and there is any doubt as to the source of any irrigation water, the sludge processing plant should demonstrate a 2 log<sub>10</sub> reduction in the numbers of *E. coli* during the treatment process.

#### **4.5 Sludges from paper processing and tanneries**

The discussion in Sections 2.4 and 2.6 indicate that the risk of pathogens being present in these sludges is extremely small and that no special measures are necessary for the destruction of human or animal pathogens.

## **5. CONSTRAINTS ON THE USE OF SLUDGES IN AGRICULTURE**

Sewage sludge and sludge from meat processing plants treated to the standards suggested in Section 4.2 will not significantly increase the pathogen load the environment where they are utilised, and in the context of pathogens, present no risk to animals, humans or plants. Sludge arising from the wastes of vegetable processing plants, paper wastes or tannery wastes present little risk if they are treated at mesophilic temperatures or the pH raised to at least 12 for 24 hours or have been composted by a suitable process.

Sludges from sewage or meat processing plants that have not been treated to standards as high as those recommended in Section 4.2.1 may present some risk to the food chain. This can be minimised by applying constraints on grazing or harvesting of subsequent crops. During this period of constraint the viability of any pathogens present will decline to minimal levels.

### **5.1 Factors influencing the survival of pathogens**

During a period of constraint a number of factors will enhance or retard the decay of micro-organisms. In reviewing the microbiological hazards of landspreading of wastes Carrington (1997) has drawn together data from a number of sources, which is summarised in Table 5.1. A major disadvantage of determining and applying periods of constraint is that most of the lethal factors cannot be controlled or predicted with certainty, e.g. temperature, and duration and intensity of sunlight.

### **5.2 Constraints on planting, grazing and harvesting**

Sludges of faecal origin that have only been treated by conventional processes will have a reduction in the pathogen concentration of about  $2 \log_{10}$ . This means that a further  $2 \log_{10}$  reduction should take place before the material to which they have been applied can enter the food chain. It can be seen that for sludges spread on surface the rate at which this reduction takes place is related to the local climatic conditions.

The European Community covers a range of climatic conditions and latitude, and an example of this diversity is shown in Table 5.2. Consequently because of the wide range it is difficult to make recommendations that cover the entire Community.

**Table 5.1 Factors influencing the survival of pathogens in wastes spread on land**

<b>Factor</b>	<b>Effect</b>
<b>Microbial structure</b>	
Bacteria	Spore forming bacteria (e.g. <i>Clostridium</i> spp.) are more resistant to effects of environmental pressures than vegetative bacteria (e.g. <i>Salmonella</i> spp.).
Virus	Non-enveloped viruses (e.g. enteroviruses) are more resistant to pH change and dehydration than enveloped viruses.
Parasites	Most helminths and parasitic protozoa have developed a lifecycle stage, (ova or cyst), that is resistant to environmental pressures.
<b>Environmental factors</b>	
Sunlight	All organisms are sensitive to ultraviolet irradiation at 265nm
Temperature	Survival times are longer at cooler, (but above freezing) temperatures. Enteric bacteria may multiply at summer temperatures.
Moisture	Most organisms are not resistant to desiccation. Rainfall may reduce concentrations through runoff or leaching.
pH	Survival times are generally shorter at low (<4) or high (>10) pH values.
<b>Quality of waste</b>	
Pathogen levels	Die-off usually follows a logarithmic curve, higher concentrations give longer ultimate survival times.
Organic content	Suitable organic content will allow the growth of enteric bacteria, if other conditions are satisfactory. However, it will also allow growth of indigenous organisms, or ones migrating from adjacent soil, that are more likely to be adapted to ambient conditions and more successfully compete for nutrients, oxygen and space.
Competing organisms	Waste and/or adjacent soil will support protozoa and nematodes, which are predators to bacteria and viruses.
Toxic substances antimicrobials	These are unlikely to be present in wastes in effective concentrations. Naturally occurring anti-microbials may be present in adjacent soils.
<b>Sludge spreading</b>	
Application rate	Penetration of ultraviolet irradiation, heat, moisture and predators from soil are reduced as the thickness of the sludge blanket increases, as is the loss of moisture by evaporation.

**Table 5.2 Diversity of climate and latitude within the EU**

	Latitude	Temperature <sup>a</sup>	Precipitation <sup>b</sup>
<b>Finland</b>			
Helsinki	60N	-9 to +16°C	200 to 1100 mm
<b>Greece</b>			
Athens	38N	+10 to +28°C	38 to 1270 mm
<sup>a</sup> Range of average monthly temperatures for specified city			
<sup>b</sup> Range of average annual precipitation across the country. Precipitation occurs most days in Finland and there is a snow cover for 4-7 months. Most rain in Greece falls in the winter, snow is rare in the lowlands.			

In the final summary of the presentations to a conference on health risks of sludge disposal, Wallis and Lehmann (undated, *circa* 1983) identified factors influencing pathogen survival similar to those listed in Table 5.1, but also derived from the presented data, an indication of the survival times (not 2 log<sub>10</sub> reduction) of *Salmonella* and *Ascaris ova*, which is shown in Table 5.3.

**Table 5.3 Relative survival times of *Salmonella* and *Ascaris ova* in sludge applied to the soil surface**

Moisture content	Temperature	Bare soil		Moderate vegetation	
		<i>Salmonella</i>	<i>Ascaris</i>	<i>Salmonella</i>	<i>Ascaris</i>
< 8%	>10°C	days	weeks	days	weeks
>8%	<10°C	days	months	3 weeks	years
<8%	<10°C	days	months	6 months	15 months

These data would suggest that during the summer for most of Europe, and for the whole year in southern Europe, a 3-week period of 'no-grazing' or prohibition of working of bare soil should be adequate to reduce pathogen numbers by 2 log<sub>10</sub>. These figures are in agreement with those presented by Carrington *et al.*, (1998) who reviewed a range of literature, much of which had been presented at COST 681 symposia.

Where crops are consumed directly by humans, particularly where they may be eaten raw, a longer period between application and harvesting should be applied. A prohibition of application during the growing season would seem appropriate. This would allow application when vegetation is minimal or non-existent and maximises the deleterious effect (to pathogens) of the climate. The original literature reviews for the UK *Code of practice for agricultural use of sewage sludge land* (unpublished) identified a 10-month period before harvest during which at least a 2 log<sub>10</sub> reduction in numbers should occur. This data confirms the recommendation in Directive 86/278/EEC. Similar constraints were made in German and Danish regulations requiring compliance with the Directive.

### **5.3 Deep injection and plough down**

It can be seen from Table 5.1 that where deep injection is used the deleterious effects of weather are minimal. However, the risk from casual contact with the sludge by wildlife and recreational users of the land is reduced to one that is very small, and in addition the effect of the natural predators in the soil is enhanced by the more intimate contact between sludge and soil.

Although deep injection has the advantage to the farmer that for grassland there is little disturbance to the sward, care must be taken that sludge material is not spilt on the land surface and that the injection channels do not allow the sludge to enter land drains or water courses.

Ploughing down has the advantages that it is more aesthetically pleasing than surface application, minimises the loss of ammonia, and places the fertilising effect immediately in intimate contact with the soil. It also minimises potential runoff of residues into surface waters. However, it does reduce the effects of environmental factors on the survival of pathogens.

There is information on the survival of pathogens in soil, but little information on the die-off rates.

Injection and ploughing down minimises the risks to wildlife and non-agricultural users of land. However, because of the risks of splashing and spillage during injection and the unknown factors relating to survival in soil, it would seem appropriate that similar grazing and harvesting constraints are used, particularly for crops eaten raw, whether conventionally treated sludges are incorporated into the soil or applied to land surfaces.

### **5.4 Constraints on sludge use**

The constraints on the land application of sludge will vary according to the treatment to which the sludge has been subjected and the crops which are produced subsequent to the sludge application. The constraints apply particularly to conventionally treated sludges where there is a greater risk of pathogens being present. The constraints have to be more stringent the greater the risk to the consumer. For example there is a much higher risk where fruit and vegetables are eaten raw, as with salads, than from processed cereals from arable land. Constraints by sludge treatment and by crop are suggested in Table 5.4. As a precautionary measure, a 30 month no-harvest period is recommended between an application of sludge to the land and the subsequent harvesting of any fruit and vegetable crops in ground contact and to be eaten raw. In reality, sludge would not be used on land growing salad crops and this guidance would apply in situations where a farmer growing, for instance, arable crops fertilised with sludge plans to change the land use to salad crops without sludge. Evidence to justify the 10-month no-harvest recommendation for vegetables in ground contact was presented by Carrington *et al.* (1998)

In producing these recommendations it is assumed that the sludge will also meet other requirements that may be defined in the revised Directive which are not the subject of the terms of reference of this report, e.g. heavy metal content and organic content. Local conditions, such as those discussed in section 5.2, may also have to be taken into account.

There are circumstances where sludge should not be used because the risk of not transferring pathogens into areas free of specific pathogens cannot be guaranteed. For example, the

potato cyst nematode, which also infects bulbs, persists in contaminated soil for many years. To reduce the spread of this pathogen Regulations in the UK and elsewhere require that fields where basic seed potatoes are grown are demonstrated to be free of the cyst stage of the nematode. There is no satisfactory method of testing the viability of cysts found in the soil. As a result, application of any sludge is prohibited where there is an intention to grow basic seed potatoes or seed bulbs, even though the cysts are rendered non-viable by some sludge treatment processes

**Table 5.4 Suggested constraints on land use of treated sludge**

<b>Crop</b>	<b>Advanced treatment</b>	<b>Conventional treatment</b>
Pasture	Yes	Injection and 3-week no-grazing
Forage	Yes	3-weeks no-harvest
Arable	Yes	Injection or plough-in
Vegetables in ground contact	Yes	10-month no-harvest
Fruit & vegetables eaten raw [salads]	Yes	30-month no-harvest
Fruit trees, vineyards	Yes	Injection and 10-month no-access
Parks & urban open spaces	Well stabilised and odourless	No
Land reclamation	Yes	10-month no-access

## **5.5 Other risks associated with agricultural use of sludges**

### **5.5.1 Transport and distribution of sludge**

Those workers who treat, transport and distribute sludge are at some risk due to the nature of their employment. These risks should be minimised by the application of local and Community legislation, e.g. the Directive on biological agents at work (Directive, 1995), and related legislation. The principles that should be applied are that: the risk is explained to the workers; suitable protective clothing is provided; washing facilities for at least hands and face should be readily available; and adequate provision must be made for meal breaks to be taken away from sludge.

### **5.5.2 Wildlife and general public**

The principle that the pathogen content of treated sludges is reduced to ambient levels minimises the risk to wild animal health. The risk of increased contamination at secondary sites by animals transferring sludge on their bodies or on carrion is minimised. The principle also protects the general public who may use the treated land.

## 6. QUALITY ASSURANCE AND VALIDATION OF PROCESSES

### 6.1 Quality assurance

#### 6.1.1 Problems associated with routine analysis

The discussion on levels of pathogens in the sludges that may present the most risk to humans and animals, (sections 2.1 and 2.2), demonstrates that the distribution of pathogens will vary both temporally and geographically. Such variations mean that analytical results apply only to the sample examined and cannot be extrapolated with confidence to the whole output of the treatment plant, even when regular monitoring is carried out. In addition there is the complication that pathogens from a wide range of genera, from a range of phyla may be present.

The routine monitoring of the microbiological quality of sludges has been discussed by Cooper and Riggs (1994) and by Pike (1984). The difficulties they identified include:

- many tests for pathogens were developed for clinical material where the pathogen is likely to be present in large numbers relative to other organisms, but the converse applies to sludges;
- most clinical requirements are only for 'presence or absence' of a pathogen and to modify these for enumeration (e.g. most probable number tests) is expensive in materials and technician time;
- tests can be expensive, particularly if enrichment procedures are required;
- 'most probable number' techniques are inaccurate;
- most genera of bacteria require specific media, and most virus types require specific cell lines;
- some pathogens cannot be readily enumerated;
- the recovery of ova of helminths and cysts of protozoa is tedious, inefficient and viability cannot be readily determined;
- definitive results are not available for at least one day and in some cases (e.g. viability of *Ascaris* ova) for several weeks.

Pike also pointed out that a test indicates the pathogen numbers at the time of analysis and does not detect any growth or decay between the time of monitoring and the application to land.

Pike *et al.* (1988b) found it necessary to confirm presumptive results as the presence of indigenous heat-tolerant organisms in the sludge could lead to false positive results.

The alternative to routine monitoring for pathogens is to specify the sludge treatment conditions and monitor the process on a regular or continuous basis. Strauch (1998) has

discussed this point and suggests process controls for a number of treatment processes. Examples of these are shown in Table 6.1.

**Table 6.1 Examples of process controls to determine pathogen kill**

Process	Parameters to be monitored
Pasteurisation	<ul style="list-style-type: none"> <li>• temperature within reactor by continuous monitoring</li> <li>• retention time</li> </ul>
Aerobic thermophilic stabilisation	<ul style="list-style-type: none"> <li>• temperature in at least 2 positions with recording instruments</li> <li>• pH values of raw and treated sludge</li> <li>• daily volume flow (and consequently retention times)</li> </ul>
Lime treatment [slaked lime]	<ul style="list-style-type: none"> <li>• initial pH value</li> <li>• reaction time</li> </ul>
Lime treatment [quicklime]	<ul style="list-style-type: none"> <li>• ratio of lime to sludge dry matter</li> <li>• initial pH value</li> <li>• temperature &gt; 2 hours after mixing in 3 positions including one in the outer zone</li> </ul>
Composting	<ul style="list-style-type: none"> <li>• initial water content of mixture</li> <li>• temperature, daily in 3 positions including central part and outer zone</li> <li>• reaction time and record of turnings</li> </ul>

In addition to the appropriate monitoring of treatment plants by the parameters outlined above it is desirable that at intervals it is demonstrated that the treated sludge is meeting the standards of hygienisation. Strauch (1998) suggests that immediately after treatment no *Salmonella* should be detected and that the concentration of enterobacteriaceae should not exceed 1000 in one gram of treated sludge.

It is implicit in the US EPA "Class A" sludge regulations that pathogens should be below the levels of detection, which are defined as:

*Salmonella* spp. - less than 3 MPN<sup>1</sup> per 4 grams of solids (dry weight);

enteroviruses - less than 1 PFU<sup>2</sup> per 4 grams of solids (dry weight);

viable helminth ova - less than 1 viable helminth ova per 4 grams of solids (dry weight).

<sup>1</sup> MPN - most probable number, a form of estimation of bacterial numbers

<sup>2</sup> PFU - plaque forming units, a form of estimation of virus numbers.

At the same time analysis should be carried out using surrogate or 'indicator' organisms to confirm that the treatment would satisfactorily reduce numbers of pathogens, had they been present.

It is also a requirement of the US EPA "Class A" regulations that the number of *E. coli* should not exceed 1000 per gram of dry solids.

### 6.1.2 Characteristics required of indicator organisms

An indicator organism, or surrogate for a pathogen, should ideally have the following characteristics:

Being consistently present in the raw sludge in large numbers;

Being not less resistant to the lethal aspects of the sludge treatment processes, but also not be significantly more resistant, than the pathogens for which it is being used to monitor; and

Being relatively easy to cultivate, and preferably have characteristics that make it easy to identify and confirm.

### 6.1.3 Selection and application of indicator organisms

In practice the presence of protozoa and helminths in sludge is unpredictable. The presence of viruses is inconsistent and they are difficult and tedious to cultivate. Consequently the indicator organisms of choice are bacteria and the candidates, which fit the requirements, are *E. coli* which has characteristics similar to vegetative bacteria, e.g. *Salmonella*, *Shigella*, *Listeria* and *Vibrio*, and also most viruses. Studies in Denmark have suggested that faecal streptococci (enterococci) are more resistant than *E. coli* and most pathogenic viruses and bacteria.

*Clostridium* spp. (e.g. *C. perfringens*) are common in raw sludge and are more resistant to heat and other sludge treatment methods than vegetative bacteria. Langeland and Paulsrud (1985) indicated that numbers similar to those of *E. coli* were present in untreated sludge. Their destruction would confirm that spore bearing bacteria, e.g. *Bacillus* spp., and resistant ova, e.g. *Ascaris*, were destroyed during treatment. However, there are no reported studies on the effects of sludge treatment upon their survival.

It is suggested that the indicators are *E. coli*, and *C. perfringens*, and that the levels of *E. coli* should not exceed 1000 per gram of sludge (dry weight). In the absence of definitive information, a tentative maximum concentration of *C. perfringens* spores of 3000 per gram of sludge (dry weight) is suggested.

The tests should be carried out on a regular basis to confirm that the treatment plant operation is satisfactory. Ideally the tests should indicate the quality of the sludge being applied to land and be carried out shortly before land application. However, this is not practicable because of the time required for testing. The US EPA suggest that for their "Class A" sludges such monitoring should take place within the two weeks prior to the land application.

Because of the diversity of origin of vegetable processing waste it is unlikely that any single pathogen, or even a small number of pathogens, is likely to be present in all or most wastes. Little is known about non-pathogenic micro-organisms that may be associated with plants and plant waste and therefore it is not possible at this time to make recommendations for organisms that may be used as indicators of satisfactory hygienisation of such waste in the context of crop protection.

Böhm *et al.* (1999) suggest that in that absence of other indicators tomato seeds may be used as an indicator that waste of domestic origin has been treated to a satisfactory standard from a phytohygiene point of view. Tomato seeds are almost always present in domestic waste. A standard of not more than 2 viable seeds per litre of treated sludge is suggested. However if sewage sludge has been treated sufficiently to meet the standards based on mammalian bacteria the viability of tomato seeds will have been destroyed and such a test would only be an additional cost and would delay the release of treated sludges.

The development of tests for phytohygienisation are possible but because of the lack of information on suitable 'indicator' organisms such development would entail a long study.

## 6.2 Validation of the processes

Although the best practical approach for quality assurance is to monitor the operating parameters of the treatment plant, it is important that the effectiveness of a plant is initially confirmed by the addition and enumeration of pathogens.

It is suggested in section 4.2.2 that a 4 log<sub>10</sub> reduction in numbers would produce a treated sludge that is virtually free of pathogens. This would entail spiking the raw sludge to a concentration equivalent to 10<sup>5</sup>-10<sup>6</sup> per sample volume. In a full-scale treatment plant it may not be practicable to spike the sludge, but various forms of water-permeable bags to expose the test organism to the treatment conditions have been proposed (e.g. Senkpiel and Ohgke, 1998; Pike *et al.*, 1988a).

*Salmonella* species may or may not be present in the raw sludge, therefore to confirm their destruction during a quality control evaluation they would have to be added (in large numbers) to the treatment plant. *Salmonella senftenberg* has been suggested as it is relatively heat resistant and rarely affects humans. However, it is widespread in poultry and human cases of *S. senftenberg* have been associated with consumption of raw foods that have been fertilised or in contact with poultry wastes (US FDA, 1999; Taorimna *et al.*, 1999). A report from Korea (Ki *et al.*, 1998) indicates that the number of human isolates of *S. senftenberg* is increasing worldwide and at least one large outbreak (104 patients) has occurred in that country.

It is impracticable to evaluate plants against all possible pathogens that may occur in sludges. For sludges originating from wastes containing faecal material both Pike *et al.* (1988a) and Strauch (1998) suggest the use of *Salmonella* and *Ascaris ova* as representative organisms. These organisms are relatively easy to culture and enumerate and consequently a statistically significant number of assays can be carried out. Salmonellae are representative of the vegetative enteric pathogenic bacteria; those which are most likely to be present in high numbers in sludge originating from faecal material. *Ascaris ova* are representative of the more heat resistant organisms (see Figure 3.1). *Salmonella dusseldorf* and *S. senftenberg* have been suggested as suitable test organisms (Pike *et al.*, 1988a; Strauch, 1998; Senkpiel and Ohgke, 1998). Both serotypes have an antigenic structure such that confirmation is fairly easy, and Veermuthu *et al.* (1998) demonstrated that *S. senftenberg* was more heat resistant than

*E. coli* O157. The use of *S. senftenberg* strain W775 is a requirement of the German Biowaste Ordinance. Pike *et al.* (1988a) found that aged cultures of *Salmonella* were more heat resistant than fresh cultures and that careful examination of treated *Ascaris* ova was necessary as temperatures around 45°C did not inhibit cell division but did inhibit development of the larval stage.

It is suggested that new treatment plants and new processes should be validated by determining the destruction of viability of *E. coli*, *C. perfringens*, *S. senftenberg* and the ova of *Ascaris*. The levels of *E. coli* should not exceed 1000 per gram (dry weight) of treated sludge, and in the absence of more definitive evidence a level of 3000 *C. perfringens* spores is suggested. A reduction of at least 4 log<sub>10</sub> of added *Salmonella* and the destruction of the viability of *Ascaris* ova should be demonstrated.

For plants treating sludges from vegetable washing without any faecal material the use of these organisms may result in unnecessarily stringent conditions. Böhm *et al.* (1999) have suggested the use of the Tobacco mosaic virus, the fungus *Plasmodiophora brassicae* and the seeds of *Lycopersicon lycopersicum* (cultivar St Pierre) as suitable test organisms in relation to phytohygienic safety.

Generally it is not practicable to spike all of the feed sludge with pathogens for validation purposes. However several groups of workers have developed 'germ carriers' which enable the test organism to be exposed to the treatment conditions but recovered with relative ease (see supplementary reference list).



## 7. CONCLUSIONS

The conclusions to be drawn from this report can be summarised in the following way.

### ***Identity of pathogens***

A wide range of phyla, genera and species are likely to be present in sludges, particularly those that contain large amounts of faecal material.

There is little evidence to suggest the respiratory diseases may be transmitted by sludges, or that toxins may present a problem.

The identity of the pathogens and their numbers will be dependent upon the health of the contributing population.

Human or animal pathogens are unlikely to be present in sludge from vegetable processing, but plant pathogens may be present.

Pathogens are unlikely to be present in waste from paper processing or tannery waste.

### ***Parameters affecting the kill or inactivation of pathogens***

Thermophilic temperatures are lethal to pathogens if they are exposed for sufficient time such as: 7 minutes at 70°C; 30 minutes at 65°C; or 4 hours at 55°C in digesting conditions. However plant design constraints indicate that the minimum time for exposure should be 30 minutes.

At mesophilic temperatures the products of digestion e.g. fatty acids, enhance the lethal effect of temperature.

The effect of ambient temperatures on sludges which have been spread on land is dependent upon the local climate.

Thermal drying at temperatures in excess of 80°C with the final water content at less than 10% is an effective means of eliminating pathogens.

Generally simple drying is not an effective treatment, except in Mediterranean climates.

The use of quicklime to raise the pH to at least 12 and the temperature to at least 55°C for at least 2 hours will produce a hygienised sludge.

Irradiation has potential but probably is not acceptable.

### ***Treatment needs***

The principle for defining the quality of treated sludge should be on the basis of risk to human, animal and plant life. The level of pathogens should not exceed the ambient levels in the environment. In practice this means for the purposes of quality control realistic limits of defined pathogens must be set.

To produce pathogen-free sludge from sewage sludge or sludge from meat processing waste it needs to be treated for the time-temperature conditions within the 'safety zone' of Figure 3.1; or raised to 55°C for 2 hours whilst the pH is above 12; or raised to 55°C for 4 hours during a thermophilic stabilisation process; or raised to 55°C for 4 hours during a satisfactory composting process.

In windrow composting systems the material should be turned at least three times to ensure that all material is exposed to the time-temperature conditions.

The level of *E. coli* in treated sludge released for land use should not exceed 1000 per gram (dry weight). The tentative level of *C. perfringens* spores of not more than 3000 per gram (dry weight) is suggested.

Advanced treatment process should demonstrate a 4 log<sub>10</sub> reduction in numbers of added *Salmonella*, and *Ascaris ova* should be rendered non-viable.

Plant pathogens will be inactivated by temperatures at the upper end of the mesophilic range (about 40°C).

Sludges from paper processing and tanneries should not need special measures to destroy human or animal pathogens.

### **Constraints on the use of sludges in agriculture**

Sludges treated to the suggested standards for advanced treatment will not add to the pathogen burden of the environment and present no risk to human, animal or plant health.

Sludges that may contain the BSE agent should not be applied to land where animals have direct access.

Sludges from paper waste, vegetable waste and tannery waste should present no risk if treated by mesophilic digestion or a similar standard.

Planting, grazing or harvesting constraints will have to be applied to land that has received sludge which has not been hygienised from sewage treatment plant or from meat processing plants.

These constraints should be sufficient to allow at least 2 log<sub>10</sub> reduction in pathogen numbers as evidenced by reduction of numbers of *E. coli*.

The rate of reduction depends upon local climatic conditions, which vary widely across the European Community.

Constraints on land use after sludge application will depend upon the level of sludge treatment, and the nature of the crop or the use of the land. In most instances 'Advanced' treated sludge can be used with few restrictions but 'Conventional' treated sludge must be distanced from the harvested material by time, and where crop maturation is a short period, by incorporating the sludge into the soil.

Workers treating, transporting or spreading sludge must receive adequate training and briefing on the risks and precautions to be observed.

### **Quality assurance**

It is not practicable to monitor treated sludge for the presence of pathogens and surrogates should be used for routine evaluation of the treatment plant performance and sludge quality.

Surrogates should be organisms commonly found in sludges and have similar resistance to treatment as pathogens. *E. coli* (or enterococci) and *Clostridium perfringens* are suggested. Numbers of *E. coli* should not exceed 1000 per gram (dry weight) and it is tentatively recommended that spores of *C. perfringens* should not exceed 3000 per gram (dry weight)

Because of the diversity of micro-organisms and hosts it is not possible to suggest suitable surrogates for systems treating vegetable waste. However, a treatment plant designed to hygienise sewage sludge or sludges of animal origin will also destroy plant pathogens.

### **Validation of new processes and new treatment plants**

New treatment processes and plants need to be validated.

A reduction of at least 4 log<sub>10</sub> of added *Salmonella* and the destruction of viability of *Ascaris* ova together with a level of *E. coli* not exceeding 1000 per gram (dry weight) of sludge and a level of *Clostridium perfringens* spores not exceeding 3000 per gram (dry weight) in the treated material is suggested for plants treating sewage sludge or animal waste.

For treatment plants receiving solely vegetable waste the use of Tobacco mosaic virus, the fungus *Plasmodiophora brassicae* and the seeds of *Lycopersicon lycopersicum* (cultivar St Pierre) has been suggested.



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