HELMINTH OVA CONTROL IN WASTEWATER AND SLUDGE FOR AGRICULTURAL REUSE

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Summary

A new version of the WHO Guidelines for the Safe Use of Waste Water, Excreta and Greywater in Agriculture and Aquaculture has been released in 2006. These guidelines, among other things, establish criteria for the helminth ova content, considering them as one of the main targeted pollutants for developing countries. However, in spite of this breakthrough and the fact that helminth ova have been considered the main health risk
when wastewater is reused for irrigation or aquaculture, relatively little information exists on how to remove and inactivate helminth ova from wastewater and sludge and, consequently, there are few technological options for controlling them. Moreover, it is still common nowadays to find recommendations on how technology can be applied to solving this problem based on data related to the inactivation of thermo-tolerant coliforms, even though it is well known that these are not indicators of helminth ova behavior. Furthermore, treatment methods are unable to produce treated wastewater and sludge with the low helminth ova content required by such criteria due to the high initial content found in the developing world. Due to the great need to apply adequate control methods in developing countries to address helminthiasis problems, this paper presents useful information for environmental and sanitary engineers concerning: (a) the general characteristics of the helminth ova; (b) the common helminth ova genus found in wastewater and sludge around the world; (c) the reason why common water and sludge disinfection methods are not effective at inactivating helminth eggs; (d) the main removal and inactivation mechanisms, (e) the processes that in practice have effectively removed or inactivated helminth ova and (f) how its content is measured in wastewater and sludge.

1. Introduction

In several regions of the world, wastewater (treated or untreated) and sludge, are being used for agricultural works. In 1989, the World Health Organization (WHO) drew attention to diarrheic diseases caused by these practices and the presence of helminth ova. In agreement, WHO set guidelines for its safe reuse. In 1992, the US-EPA published biosolids and sludge criteria (part 503) defining the elimination of helminth ova as a key parameter for sludge revalorization in agriculture. In 2006, WHO once again released guidelines, this time for the safe use of wastewater, fecal material and sludge in agriculture and aquaculture with a recommendation on the helminth ova content to a value that for developing countries will imply using treatment methods with 1-3 log removal or inactivation efficiencies, for which there is almost no information available. In spite of the importance of helminth ova as waterborne vectors, throughout the years, little attention has been paid to them in terms of their characterization and control, in both wastewater and sludge. Helminth ova are still poorly known and understood in the water profession and are often thought to be similar to microorganisms (bacteria, viruses and protozoa) even though they behave very differently. This chapter reviews from a practical point of view: (a) general characteristics of helminth ova; (b) common helminth ova genus found in wastewater and sludge around the world; (c) the reason why common water and sludge disinfection methods are not effective at inactivating helminth eggs; (d) main removal and inactivation mechanisms, and, (e) processes that in practice have effectively removed or inactivated helminth ova. This consolidated information (until now spread across research and internal reports papers) addresses to water professionals dealing with wastewater and sludge problems in developing countries. Information should also encourage researchers to look for more useful information on helminth ova characteristics but as well on removal and inactivation methods.

2. General Information
2.1. Helminthiasis: A Common Disease

Globally there are 5 million people suffering helminthiasis, mainly in developing countries. Helminthiasis is particularly common in regions where poverty and poor sanitary conditions are dominant. Under these circumstances helminthiasis incident rates reach 90%. There are several kinds of helminthiasis, Ascariasis being the most common and endemic in Africa, Latin America and the Far East. Even though the mortality rate is low, most of the people infected are children under 15 years with problems of faltering growth and/or decreased physical fitness. Around 1.5 million of these children will probably never bridge the growth deficit, even if treated. Helminthiasis is transmitted through: (a) the ingestion of polluted crops, (b) contact with polluted sludge, faeces or wastewater, and (c) the ingestion of polluted meat.

2.2. Helminths’ Life Cycle

Helminthiasis infective agents are the eggs, not the worms. Worms cannot live either in wastewater or in sludge because they need a host. Therefore, part of the control strategy for helminthiasis is to remove the eggs from wastewater and later inactive them in the sludge produced from wastewater treatment. Helminths are pluri-cellular worms; they are not microbes although their eggs are microscopic. Helminths come in different types and sizes (from 1 mm to several m in length), with several life cycles and ideal living environments. Besides humans some of them have intermediary hosts (such as \textit{Schistosoma} spp. that live temporarily in a snail). Helminths’ life cycle is very complex and different from that of bacteria and protozoan, which are well-known microbes in the wastewater treatment field. The \textit{Ascaris lumbricoide’s} life cycle illustrates this complexity well. When a person ingests \textit{Ascaris} eggs (1-10), they adhere to the duodenum where the larva leaves the shell, crossing the wall into the blood stream. Through the blood \textit{Ascaris} travels to the heart, lungs and bronchus tubes where it breaks the walls remaining in the alveolus around 10 days. The larva then travels to the trachea from where it is ingested again returning to the intestine. Back in the intestine, \textit{Ascaris} reaches its adult phase, and, if female, produces up to 27 million eggs. During, its migration, \textit{Ascaris} provokes allergic reactions (fever, urticaria and asthma); it may also sometimes lodge in the kidney, bladder, appendix, pancreas or liver forming cysts that can only be removed through surgery. In the intestine, \textit{Ascaris} produces abdominal pain, meteorism, nausea, vomiting, diarrhea and undernourishment. Helminthiasis diseases have different manifestations but in general they cause intestinal wall damage, hemorrhages, deficient blood coagulation and undernourishment. Helminthiasis can degenerate into cancer tumors.

2.3. Classification

There are three different types of helminths: (a) Plathelminths, or flat worms, (b) Nemathelminths, Nematodes or round worms, and (c) Annelids (see Figure 1). In the sanitary engineering field only the first two are of importance. A common characteristic of helminths is that they reproduce through eggs. The eggs of different helminths differ in shape and size (see Figure 2). As can be seen in Figure 1 it is improper to use the terms nematodes, \textit{Ascaris} and helminths as synonyms, as frequently happens in the sanitary engineering literature. This misunderstanding comes from the fact that \textit{Ascaris}
(a nematode) is the most common helminth egg in wastewater and sludge (see Figure 3).

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Soil Absolute Maximum</th>
<th>Soil Common Maximum</th>
<th>Crops Absolute Maximum</th>
<th>Crops Common Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacteria</td>
<td>70 days</td>
<td>20 days</td>
<td>30 days</td>
<td>15 days</td>
</tr>
<tr>
<td>Viruses</td>
<td>100 days</td>
<td>20 days</td>
<td>2 months</td>
<td>15 days</td>
</tr>
<tr>
<td>Protozoan Oocytes</td>
<td>20 days</td>
<td>10 days</td>
<td>10 days</td>
<td>2 days</td>
</tr>
<tr>
<td>Helminth Ova</td>
<td>Many months</td>
<td>Many months</td>
<td>2 months</td>
<td>1 month</td>
</tr>
</tbody>
</table>

Table 1: Survival time of different pathogens in soil and crops

Note: Periods of time may vary according to weather conditions.

Normally, eggs contained in wastewater are not infective. To be infective they need to develop larva, for which a certain temperature and moisture are required (26°C and 1 month in laboratory conditions. Conditions usually found in soil or crops are suitable for the development of larvae in 10 days, hence the risk of using polluted wastewater or sludge in agricultural fields. According to information that is now several years old and obtained, using a much less sensitive helminth ova analytical technique than the one available nowadays, it was found that they live in water, soil and crops for several months, a period of time that it is much longer than the one for microorganisms (1-2 months in crops and many months in soil, see Table 1).

3. Helminth Ova in Wastewater and Sludge

3.1. Type and Content

Due to differences in health and conditions according to the little literature available on the subject, helminth ova content in wastewater and sludge is very different (Table 2) in developed and developing countries. Moreover, the distribution genera presented vary from country to country, reflecting local health conditions. Different helminth ova genus reported as detected either in wastewater or sludge are mentioned in Figure 1, while a general distribution of genus, but not representative for all countries is presented in Figure 3.

<table>
<thead>
<tr>
<th>Country/region</th>
<th>Municipal wastewater HO L⁻¹</th>
<th>Sludge HO g⁻¹ TS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Developing countries</td>
<td>70-3000</td>
<td>70-735</td>
</tr>
<tr>
<td>Brazil</td>
<td>166–202</td>
<td>75</td>
</tr>
<tr>
<td>Egypt</td>
<td>No data</td>
<td>Mean: 67; Maximum: 735</td>
</tr>
<tr>
<td>Ghana</td>
<td>No data</td>
<td>76</td>
</tr>
<tr>
<td>Jordan</td>
<td>300</td>
<td>No data</td>
</tr>
<tr>
<td>Mexico</td>
<td>6–98 in cities</td>
<td>73-177</td>
</tr>
</tbody>
</table>
Table 2: Helminth ova content in wastewater and sludge from different countries

<table>
<thead>
<tr>
<th>Country</th>
<th>Helminth Ova Content</th>
</tr>
</thead>
<tbody>
<tr>
<td>Morocco</td>
<td>Up to 330 in rural</td>
</tr>
<tr>
<td></td>
<td>and peri-urban areas</td>
</tr>
<tr>
<td>Ukraine</td>
<td>840</td>
</tr>
<tr>
<td>France</td>
<td>60</td>
</tr>
<tr>
<td>Germany</td>
<td>No data</td>
</tr>
<tr>
<td>Great Britain</td>
<td>No data</td>
</tr>
<tr>
<td>United States</td>
<td>1-8</td>
</tr>
<tr>
<td></td>
<td>2-13</td>
</tr>
</tbody>
</table>

Figure 1: Helminth classification and common genus found in wastewater and sludge.

3.2. Fecal Coliforms as Indicators

Fecal coliforms are the bacterial pollution indicators most extensively used, and it is frequently, and wrongly, assumed that they are indicators of any kind of biological pollution. Even though fecal coliforms are useful indicators of fecal pollution in developed countries, this is not the case in developing ones owing to the presence of a wide variety and larger quantities of microorganisms. This does not mean that fecal coliforms are not useful in developing countries, simply that care must be taken to select additional indicators for specific purposes, such as agriculture and aquaculture wastewater and sludge reuse, which is where helminth ova fit in, given that they are one of the main associated health risks displaying a much higher resistance to environmental conditions than viruses, bacteria and protozoa. Actually, in contrast to fecal coliforms, helminth ova cannot be inactivated with chlorine, UV light or ozone (in the latter case at least not with economical doses because >36 mg O₃ L⁻¹ are needed with 1 hour contact time.)
3.3. Helminth Ova Criteria

As shown in Table 2, not all wastewater and sludge contain significant amounts of helminth ova. For this reason they are not considered in all wastewater and sludge countries’ norms as is the case of BOD or fecal coliforms, which are universal parameters. Based on epidemiological studies WHO has set a recommended limit of $\leq 1 \text{ HO L}^{-1}$ for the irrigation of crops that are eaten uncooked for both restricted and unrestricted irrigations, but for drip irrigation of high growing crops (crops not growing down or on the soil), there is no recommendation. For fish culture with wastewater, the trematode eggs (Schistosoma spp., Clonorchis sinensis and Fasciolopsis buski) maximum content has been set as zero, as these worms multiply by tens of thousand producing millions of eggs in their first intermediate aquatic host (a snail) before infecting fish and humans.

In sludge or biosolids intended for agriculture, based on the value of $\leq 1 \text{ HO L}^{-1}$ set for wastewater, some authors have calculated a limit criterion of 3 to 8 HO g$^{-1}$ TS for sludge depending on its application rate. This value is much higher than the 0.25 HO g$^{-1}$ TS set in the United States as standard or the 1 HO g$^{-1}$ TS set as criteria by WHO (for fecal sludge). The US EPA value has been set based on the inactivation removal achieved by most of the available treatment technologies (with efficiencies of around 90%) to treat sludge with a maximum helminth ova content of 10 HO g$^{-1}$ TS. Anyway, in practice, both, standard and criteria mean very high inactivation efficiencies ($< 99\%$).
for sludge and fecal sludge (due to high initial content normally found in developing countries) not affordable in practice.

The US-EPA standard value was defined for biosolids Class A (sludge with no restriction on use), while for biosolids Class B there are no helminth ova limits, although sludge can be reused in agriculture with some restrictions. Unfortunately, for sludge with greater content than those reported for the USA, very few economical feasible options are available for inactivating helminth ova in sludge or fecal sludge with low content values. Not applying any limit as is done for class B in US-EPA would be dangerous for the revalorization of sludge in developing countries.

4. Helminth Ova Characteristics

An important characteristic of helminth ova is that they are covered by 3-4 layers. The 1-2 outer layers are formed with mucopolysacharides and proteins. The middle layers consist of chitinous and serve to give structure and mechanical resistance to the eggs.

Finally, the inner layer is composed of lipids and proteins and is useful to protect eggs from desiccation, strong acid and bases, oxidants and reductive agents as well as detergent and proteolytic compounds. Thus the combination of all these layers is responsible for making eggs very resistant to several environmental conditions.

Helminth ova of concern in the sanitary field measure between 20 and 80 μm with a density of 1.06-1.15 and are gelatinous which makes them very sticky. All these properties determine helminth ova’s behavior during wastewater and sludge treatment. First, it is very difficult to inactivate them unless the temperature is raised above 40°C or moisture is reduced to below 5% (TS > 95%).

But details about the contact time under these conditions and other related environmental factors are generally not well-defined for every type of helminth ova genus or for high helminth ova content. Only for *Ascaris* has a contact time of 10-20 days at temperatures above 40°C been reported.

In wastewater treatment, the inactivation conditions mentioned can hardly be achieved while in sludge treatment they are feasible. Thus, helminth ova are normally removed from wastewater and inactivated in sludge.

5. Helminth Ova Removal from Wastewater

Basically, to remove helminth ova from wastewater it suffices to realize that they are in fact particles forming a fraction of the suspended solids. This is why the helminth ova content is related to the total suspended solids content (TSS) in wastewater-specifically, to the amount of particles measuring 20-80 μm (Figure 4).

As helminth ova are particles, mechanisms used to remove suspended solids are also useful removing helminth ova from wastewater. These mechanisms are sedimentation, filtration and coagulation-flocculation.
Figure 4: Correlation between helminth ova content in Mexico City’s wastewater and (a) the TSS content and (b) the 20-80μm particles content

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Biographical Sketch

**Blanca Jimenez** was born in Mexico City, where she obtained a *Bachelor’s degree in Environmental Engineering*. She has Masters and a PhD degree in Wastewater Treatment from the Institut National de Sciences Appliquées, Toulouse, France. She works since 1985 at the National Autonomous University (UNAM) where she is Senior Researcher. In 1992 she founded the graduate program in Environmental Engineering in the state of Morelos and in 1994 launched the prestigious UNAM Group of Wastewater Treatment and Reuse.

Dr Jiménez has published more than 180 international papers and has 4 patents. She has published the book: “Environmental Pollution in Mexico. Causes, Effects and Technology”: She has been responsible for more than 117 research projects for several public and private institutions. Due to her professional reputation Dr. Jiménez has been invited to lecture more than 100 conferences in several countries. She has been awarded several prizes like the National Ecology Award (as best academic in the environmental) 2006, the Environment and Ecology Award “Miguel Alemán Valdés” (2001), the Award for Scientific Research in the area of Technology Research, granted by the Mexican Academy of Sciences, (1997) and the Ciba Award for Technological Innovation in Ecology (1993). She is the chairperson of the Water Reuse Specialty Group in the International Water Association. She was President of the Mexican Association of Environmental Engineers, the Mexican Federation of Sanitary Engineering and Environmental Sciences (the oldest environmental professionals association in the country), and belongs to the Executive Committee of the International Water Association.