Evaluation of the WHO helminth eggs criteria using a QMRA approach for the safe reuse of wastewater and sludge in developing countries
I. Navarro and B Jiménez

ABSTRACT
An analysis of the actual WHO recommendations to develop standards for the safe reuse of wastewater, excreta or sludge in agriculture using a quantitative microbial risk assessment (QMRA) is presented. The proposed values are defined using a risk-based model for Ascaris lumbricoides infection to assess the human risks associated with exposure to this pathogen from crops irrigated with polluted water, or from crops grown in biosolid-enriched soil. From the results it becomes evident that, with regard to helminth eggs, the WHO guidelines for wastewater reuse in agriculture seem more stringent than are needed in developing countries, while for the reuse of sludge they appear to be the opposite. Although more information is needed to confirm this conclusion, which was derived from a single piece of research, at the very least a more cautious approach is recommended when evaluating excreta or sludge for agricultural purposes in developing countries. Additionally, this work shows that the application of some barriers, other than wastewater and sludge treatment as suggested by WHO, can play an important role in controlling risks.

Key words | agriculture, developing countries, helminth eggs, QMRA, reuse, wastewater

INTRODUCTION
In 1989, the World Health Organization (WHO) established that among the main health risks associated with reusing wastewater or excreta for agricultural irrigation were those caused by helminth eggs, notably Ascaris lumbricoides (A. lumbricoides). The occurrence of helminth eggs in wastewater, faecal sludge (excreta from latrines) and sludge (from wastewater treatment plans) in developing countries differs considerably from that of industrialised countries where helminthiasis infection has much lower prevalence (Jiménez 2009). These differences in health conditions mean that the helminth egg (HE) content in wastewater and sludge (Table 1) can be 7–80 times greater in developing countries than in developed ones, and hence the risk of reusing them is greater.

Based on epidemiological studies, WHO (2006) recommended a criterion of ≤1 helminth egg (HE) L^{-1} for wastewater used for irrigation. In faecal sludge, WHO suggested a value of 1 HE per gram of total solids (g^{-1}TS or total solids). These values were established using epidemiological evidence instead of a risk assessment approach as was made for other parameters (Navarro et al. 2009). Unfortunately, due to the high initial helminth egg concentrations in wastewater, faecal sludge and sludge in many developing countries, treatment with very high efficiencies (< 99%) is needed. These targets often require unaffordable technologies or furthermore are unattainable using most conventional treatment technologies.

In 2006, the WHO Guidelines for the Safe Use of Wastewater, Excreta and Greywater suggested the development and use of health-based targets, with the goal of attaining a predetermined level of health protection in an exposed population. This level of health protection can be achieved by using a combination of risk management approaches (e.g. crop restriction, safer application techniques and human exposure control) (WHO 2006). It is also recognised that in some

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situations it is not possible to fully implement the desired level of protection at a given time. For this reason, the WHO guidelines suggest designing regulation policies that allow progressive implementation of higher levels of protection. This can be achieved over time in an ordered manner, depending on the circumstances and resources of each country. In order to achieve this, each country or government should develop a risk management plan based on the local context. To estimate the risk to human health of local relevant pathogens quantitative microbiological risk assessment (QMRA) can be used. This procedure allows the estimation of the infection or illness rates based on the pathogen densities in wastewater or excreta, the measured or estimated ingestion rates, and appropriate dose–response models for the exposed population.

Considering that QMRA had not been applied before the 2006 WHO Guidelines to assess helminth egg risks, the aim of this paper is to apply this methodology using a risk-based model for *A. lumbricoides* infection to show how a level of health protection may be developed. Two examples are presented based on local circumstances in one developing country. Consideration is also given to some health control barriers to assess human risks associated with the consumption of raw crops irrigated with untreated wastewater and those grown in biosolid-enriched soil.

### METHOD

**Dose–response model for *Ascaris lumbricoides***

A dose–response function has been developed for other pathogens based on outbreak data (Haas et al. 1993; DuPont et al. 1995). For developing countries, it is difficult to obtain outbreak data of helminth diseases mainly due to the endemic nature of helminth infections, such as ascariasis, trichuriasis, schistosomiasis, etc. (Table 2); and, therefore, delays are observed in the population between exposure to pathogens and symptomatic responses. Despite these limitations, the above mentioned dose–response curve was developed using available information from epidemiological and wastewater quality studies.

The dose–response relationship developed for *A. lumbricoides* infection was a Beta-Poisson model with values for $\alpha = 0.104$ and $\beta = 1.1$ (Equation (1)). This relationship was

<table>
<thead>
<tr>
<th><em>Helminthiases causative agent</em></th>
<th><em>Prevalence of infection (%)</em></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cameroon</td>
</tr>
<tr>
<td><em>Ascaris lumbricoides</em></td>
<td>42.3</td>
</tr>
<tr>
<td><em>Trichuris trichiura</em></td>
<td>54.7</td>
</tr>
<tr>
<td><em>Schistosoma mansoni</em></td>
<td>52.1</td>
</tr>
<tr>
<td>Hookworm spp.</td>
<td>16.6</td>
</tr>
</tbody>
</table>

the end result of fitting the observed infectivity and the exposure dose estimated for the Mezquital Valley in Mexico (Navarro et al. 2009). The epidemiological data included the prevalence of *A. lumbricoides* infection (10–17%) within the population group of concern (children under 15 years of age) (Blumenthal et al. 1996; Cifuentes et al. 1991).

The estimation of the dose was based on the content of *A. lumbricoides* eggs (6–23 eggs/L) in the untreated wastewater used for irrigation (Jiménez et al. 1992), as well as the assumption of 10 mL of wastewater remaining per 100 g of produce (Shuval et al. 1997), and a consumption of raw crops of 100 g per week per child during one year. Therefore, the probability that an individual might be infected if he/she is exposed to a dose (d) containing *A. lumbricoides* eggs may be estimated with the application of this model to specific scenarios.

\[
P(d) = 1 - \left(1 + \frac{d}{1.1}\right)^{-0.104}
\]  
(1)

**Scenarios to estimate the level of health protection for reuse practices**

Some developing countries may not be able to afford wastewater and/or excreta or sludge treatment to achieve the WHO health-safety values of ≤ 1 egg L\(^{-1}\) for wastewater or < 1 egg g\(^{-1}\)TS for sludge reuse. In such circumstances, it is recommended to initially set an affordable standard using QMRA based on the local incidence of helminthiases. Subsequently, this limit can be progressively reduced to become more stringent as the helminthiasis rate in the population decreases as a result of partially controlling the risk. Thus, the standard health-safety goal recommended by WHO can eventually be achieved. To show how an intermediate standard value might be developed, two examples, one for wastewater and another for sludge, are presented. These scenarios were developed considering that the local population in the Mezquital Valley already has an ascariasis endemic infection rate of 10–17%. One scenario describes the consumption of raw spinach irrigated with untreated wastewater and the other the consumption of raw spinach grown on biosolid-amended soil. For both cases the assumptions made for the microbial risk estimates were:

- Children 5 to 15 years of age acted as the exposure group.
- To analyse the risk variability, several spinach ingestion rates were considered: (a) a mean intake range of 30–54 g per event for child (U.S. EPA 1997, 2002); (b) a mean consumption of 65 g per eating event, and (c) the 90\(^{th}\) and 99\(^{th}\) percentile intake distribution, 140 and 270 g, respectively. These values corresponded to lettuce consumption rates due to limited data in the literature concerning spinach consumption.

- A spinach consumption frequency of once per week during a year.
- An *A. lumbricoides* content reduction of one (WHO 2006) and two log\(_{10}\) units (as a best case scenario) due to crop washing before being eaten raw were considered.
- For both scenarios, the risk was calculated using the dose-response model for *A. lumbricoides* (Equation (1)) associated with a single week of exposure to spinach eaten raw. Equation (2) was applied to estimate the annual risk considering n = 52.

\[
P_{\text{annual}} = 1 - [1 - P(d)]^n
\]  
(2)

**QUANTITATIVE MICROBIAL RISK ASSESSMENT (QMRA) RESULTS**

**Scenario 1: vegetables irrigated with untreated wastewater**

When applying Equations 1 and 2 to assess the consumption of raw vegetables irrigated with untreated wastewater the expected risk of *A. lumbricoides* infection varies from \(5 \times 10^{-2}\) to \(9 \times 10^{-1}\) per child per year. The variation depends on the wastewater application rate and on the amount of spinach consumed by the exposed population. However, these risks may be reduced if the washing procedure after harvesting is enhanced in order to remove 1 log of eggs (Figure 1). This control barrier implies the assumption of a 90% reduction of *A. lumbricoides* eggs content in crops. Under these circumstances the estimated risk range is reduced to \(5 \times 10^{-3}\)–\(2.5 \times 10^{-1}\) per child per year, resulting in an expected infection rate of less than 17%. Moreover, the infection rates may be reduced to less than 3% if an additional barrier is assumed at the food preparation points by improving the washing efficiency to remove 2 log, even though the overall risk of infection is greater than \(10^{-4}\) (\(6 \times 10^{-4}\)–\(3 \times 10^{-2}\) per child per year). Consequently, considering the factors influencing the health risk estimates (the wastewater application and the consumption rates) and the barriers, an intermediate standard of ≤ 25 *A. lumbricoides* eggs/L can be
set provided that mean consumption is less than 140 g/d per week-year (Figure 1) and the washing procedure barriers are enhanced to remove 1 to 2 log eggs. Under such conditions the infection rate estimates will vary from 10% to 1%, respectively. This would produce an infection rate significantly lower than that actually observed in the Mezquital Valley (10–17%).

Scenario 2: vegetables grown on biosolid-amended soil

Results from an experimental study performed with spinach grown in soil fertilised with biosolids were used to calculate the number of Ascaris eggs in spinach (Jiménez et al. 2006). The A. lumbricoides dose was calculated based on the use of four different initial helminth eggs content in biosolids (HE/gTS): 0.25 egg/gTS and 1 egg/gTS, and the minimum and maximum concentration values obtained in treatment processes affordable to developing countries, 4 and 37 eggs/gTS, respectively.

The annual risk of consuming uncooked spinach grown on biosolid-amended soil, after harvesting without any control barrier, is 1. If the washing procedure, as an initial barrier, is enhanced to remove 1 egg log, the annual risk decreases but still it is greater than $6 \times 10^{-1}$ per child per year, resulting in an annual infection rate greater than 58%. This annual infection rate could be reduced if the washing procedure was improved to remove 2 eggs log. However, under such circumstances, merely for a biosolid content of 0.25 eggs/gTS and a consumption rate $r = 65$ g/d once per week during a year (Figure 2), the infection rate achieved is very similar to the actual ascariasis incidence rate in the region of 10–17%. In other words, risks associated with the spinach exposure pathway when using biosolids are higher than those arising from the reuse of untreated wastewater. In this case, the value advised to control risks is less than the limit proposed by WHO of 1 egg/gTS.

The above results suggest that WHO limits for wastewater and sludge reuse in agricultural production are restrictive limits for developing countries. They illustrate that thresholds could be higher by an order of magnitude for these countries. Although higher expected infection rates were estimated and the risk estimates are greater than $10^{-4}$, the change of such standards to those that predict infection rates less than the local endemic prevalence may induce a gradual improvement in population health conditions. Finally, control regulations should be implemented in an integrated framework for risk management, where additional health protection measures,

![Figure 1](https://iwaponline.com/wst/article-pdf/63/7/1499/445650/1499.pdf)

Figure 1 | Annual risk estimate from exposure to spinach irrigated with different A. lumbricoides egg concentrations in wastewater for several consumption rates (based on Jiménez & Navarro 2009).

![Figure 2](https://iwaponline.com/wst/article-pdf/63/7/1499/445650/1499.pdf)

Figure 2 | Annual risk estimate from exposure to spinach grown on biosolid-amended soil (based on Navarro et al. 2009).
such as improvements in the washing of produce, may be considered.

**DISCUSSION**

The QMRA model and the scenarios presented here provide a useful basis for managing risks associated with flooding irrigation employing untreated wastewater for specific crops. This can be adjusted to consider different scenarios; for example, different wastewater treatment methods, different crops, or other interventions or strategies to control the health risk. The uncertainty in this model may be minimised by gathering improved input data (local spinach ingestion rates, consumption frequencies, wastewater volume remaining in different produce, etc.).

The results of this study suggest that a concentration of 23 *A. lumbricoides* eggs/L (intermediate standard) in untreated wastewater for irrigation would produce an infection rate in the Mezquital Valley significantly lower (1–10%) than the actual observed rate (10–17%), provided the mean consumption per child is less than 100 g/d per week-year and the washing procedure, as a barrier, achieves a removal of 1 to 2 log concentration. The value of the proposed intermediate standard is not very different from the value of 10 eggs/L (*Ayres et al. 1993*) suggested as a standard for the unrestricted use of wastewater in agriculture. This contrasts with the WHO recommendation of a maximum of 1 nematode egg/L.

Although this study was unable to provide conclusive evidence that an *A. lumbricoides* concentration of 23 eggs/L should be a preliminary health target for wastewater reuse, it does contribute to the understanding of the risk of infection in an area of endemic disease. This value is of the same magnitude as that reported by *Ensink et al. (2008)* in a study in India, where no increased risk of hookworm infection was detected when wastewater with a mean concentration of 15 eggs/L was used. Throughout this procedure, further scenarios with regard to local consumption patterns (type of crops, way of preparing food, frequency of ingestion, whether wastewater or sludge is used, etc.) should also be considered with the purpose of defining an intermediate standard.

One aspect to highlight is that the procedure presented only considers biological risk, and not chemical risks. Chemical values for standard settings could be developed following a risk approach. For this purpose, the WHO (2006) guidelines offer dose criteria for chemical parameters, largely heavy metals. For other compounds that could be of concern, few references are available in the literature.

**Other health protection measures**

The results of the two scenarios analysed here can be used in combination with other methods to reduce the risk arising from helminth eggs. For instance, according to *Mara & Sleigh 2010*, the use of wastewater treatment employing an anaerobic pond with a 1-day retention time and facultative pond with a 5-day retention time can achieve a 2-log unit reduction of *Ascaris* eggs. WHO encourages the development of risk reduction strategies through multiple pathogen barrier approaches at various critical control points before vegetables are consumed. The approach described here may therefore cover both conventional and non-conventional methods as well as other health-protection measures to meet health targets. Health-protection measures include improved irrigation methods, cessation of irrigation before harvesting, and the washing of produce (*Keraita et al. 2008*). For example, for agricultural irrigation in the Mezquital Valley, where flooding is a very high risk option, the suggested methodology could be used to compare the effect of replacing it with drip irrigation. Another example that could be analysed is the cessation of irrigation before harvesting. There is evidence that this protection measure has a great influence on the reduction of pathogen content in crops.

From the results obtained it is evident that washing produce is an effective way to control risks. Washing methods may include the use of some kind of disinfectant, the use of water only, or a combination of both. Previous work (*Amoah et al. 2007*) has shown that poor people in some developing countries tend to use only water or water with salt, soap and lemon juice. If data were obtained for these practices with regard to the reduction of pathogen content then the actual reduction of risks could be evaluated. However, considering that the efficacy of the method used depends not only on the washing procedure but also the type and physiology of the targeted microorganisms (*Parish et al. 2003*), microorganisms of concern should be tested for. For example, *Amoah et al. (2007)* found that, independently of the method or disinfectant used, washing in a bowl reduced the helminth egg concentration by half or more. Washing under a running tap (without any sanitiser) appeared to be even more effective, reducing helminth egg contamination levels from about 9 to 1 egg per 100 g wet weight of lettuce leaves.

The feasibility and efficacy of any combination of these or other health-protection control measures will depend on several factors (availability of resources, agricultural practices, market demand, capacity of governmental agencies, etc.). These must be carefully considered before measures are put into practice.
CONCLUSIONS

New WHO wastewater and sludge reuse guidelines offer a different and interesting approach for developing countries to set affordable regulations that can be used progressively to improve current health conditions. Nevertheless, one of the main findings is that by applying QMRA it becomes evident that the guidelines provided by WHO to reuse wastewater for agriculture seem more stringent when it refers to *A. lumbricoides* than is needed in developing countries, while for sludge reuse it is the opposite. Even though this conclusion is drawn from the results of a single piece of research, and therefore requires further investigation, a cautious approach is recommended when evaluating sludge for agricultural purposes.

REFERENCES


