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Estimation of *Ascaris* infection risks in children under 15 from the consumption of wastewater-irrigated carrots

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ABSTRACT

Ascaris lumbricoides, the large human roundworm, infects ~1,200 million people, with children under the age of 15 being particularly at risk. Monte Carlo quantitative microbial risk analyses were undertaken to estimate median *Ascaris* infection risks in children under 15 from eating raw carrots irrigated with wastewater. For a tolerable additional disease burden of 10^{-5} DALY (disability-adjusted life year) loss per person per year (pppy), the tolerable *Ascaris* infection risk is ~10^{-3} pppy, which can be achieved in hyperendemic areas by a 4-log unit *Ascaris* reduction. This reduction can be easily achieved by wastewater treatment in a 1-day anaerobic pond and 5-day facultative pond (2 log units) and peeling prior to consumption (2 log units).

Key words | agriculture, *Ascaris*, children, risk analysis, vegetables, wastewater

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INTRODUCTION

The global burden of ascariasis

In 2003 *Ascaris lumbricoides*, the large human roundworm, infected ~1,200 million people or nearly 20% of the world's population; the main parts of the world affected were, and still are, sub-Saharan Africa, India, China and East Asia (Hotez *et al.* 2006). Children are especially at risk: maximum *Ascaris* prevalence usually occurs before five years of age and the highest intensities of ascariasis (as measured by the number of eggs per g faeces) are in children aged 5–14.

Ascariasis is associated with the malabsorption of food in general and vitamin A in particular, and so often leads to protein-energy malnutrition and growth-faltering. In a 10-year study of 119 slum children in northeast Brazil, children who had had a high burden of diarrhoeal disease in their first two years of life were on average 3.6 cm shorter at age 7 than other children, and those children who had also had an early childhood helminthiasis (commonly ascariasis) were on average a further 4.6 cm shorter at the same age (Moore *et al.* 2001). Early childhood helminthiases and diarrhoeal disease, in conjunction with malnutrition, result in a loss of cognition in later childhood; the areas most affected are verbal fluency, short-term memory and speed of information processing–precisely the areas most needed for people to contribute to economic development (UNICEF 1998; Berkman *et al.* 2002). Furthermore, children with more than one helminthic infection experience worse cognitive outcomes than those with only one (Jardim-Botelho *et al.* 2008). There is also evidence that productivity in adulthood is adversely affected by helminthiases in childhood (Guyatt 2000).

Recommendations in the WHO guidelines for human intestinal nematode eggs

The 2006 WHO guidelines for the safe use of wastewater in agriculture (WHO 2006) make the same recommendation as was made in the 1989 guidelines (WHO 1989): ≤ 1 human intestinal nematode egg per litre of treated wastewater. However, epidemiological studies in Mexico have shown that, while this guideline value protects adults, it does not protect children under the age of 15 (Blumenthal *et al.* 1996). Blumenthal *et al.* (2000) therefore recommended

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lowering the guideline value to ≤ 0.1 egg per litre wherever children under 15 are exposed and the soil conditions are favourable to egg survival. This recommendation was not accepted by the international group of experts who participated in the development and review of the guidelines at a meeting held in Geneva in June 2005, on the grounds that it was too difficult to measure an egg concentration as low as 0.1 per litre. However, if the wastewater is treated in waste stabilization ponds (WSP), which are generally the best wastewater treatment process in developing countries (Mara 2004), the effluent egg concentration can be simply determined from the egg concentration in the untreated wastewater (which is relatively easy to measure) by using the design equation for egg removal in WSP given by Ayres *et al.* (1992).

Since the 2006 WHO guidelines do not protect the health of children under 15 against intestinal nematode disease (unless, additionally, they are dewormed regularly at home or at school), we have investigated by risk analyses how children under 15 can best be protected against *Ascaris* infection, for which dose-response data are now available (Navarro *et al.* 2009), from eating raw carrots irrigated with treated wastewater.

QUANTITATIVE MICROBIAL RISK ANALYSES

Tolerable *Ascaris* infection risk

The 2006 guidelines are based on a tolerable addition burden of disease of $\le 10^{-6}$ DALY loss per person per year (pppy), where DALY is a disability-adjusted life year (used as a metric to compare the disease burden of different diseases and disabilities; DCPP 2008). However, in *Levels of Protection*, one of the documents in the rolling revision of its drinking-water quality guidelines, WHO (2007) states that

> 'in locations or situations where the overall burden of disease from microbial, chemical or radiological exposures by all exposure routes is very high, setting a 10^{-6} DALY [loss] per person per year annual risk from waterborne exposure will have little impact on the overall disease burden. Therefore, setting a less stringent level of acceptable risk, such as 10^{-5} or 10^{-4} DALY [loss] per person per year, from waterborne exposure may be more realistic, yet still consistent with the goal of providing high-quality, safer water and encouraging incremental improvement of water quality'.

Following the principles of the Stockholm Framework which advocates the same level of protection for all water-related exposures (Fewtrell & Bartram 2001), this should be applied *mutatis mutandis* to wastewater use in agriculture.

Therefore, if a DALY loss of 10^{-5} pppy is acceptable (and in countries with high prevalence of ascariasis this level of protection is more than adequate, given the other, more common, environmental transmission routes for ascariasis; Feachem *et al.* 1983), assuming a DALY loss per case of ascariasis of 8.25×10^{-3} (Chan 1997) and, as worst-case scenario, an *Ascaris* disease/infection ratio of 1 (i.e. all those infected with *Ascaris* develop ascariasis), the tolerable *Ascaris* infection risk is given by:

$$\frac{\text{Tolerable DALY loss pppy}}{\text{DALY loss per case of ascariasis}} = \frac{10^{-5}}{8.25 \times 10^{-3}}$$
$$= 1.2 \times 10^{-3}\ \text{pppy}$$

Monte Carlo risk simulations

The quantitative microbial risk analysis–Monte Carlo (QMRA-MC) methodology used to estimate *Ascaris* infection risks as a result of consuming wastewater-irrigated carrots was based on the work of Shuval *et al.* (1997), Haas *et al.* (1999), Mara *et al.* (2007), Benke & Hamilton (2008), and Navarro *et al.* (2009).

In their risk analysis of *Ascaris* infection in children under 15 from the consumption of raw carrots grown in biosolids-amended soil, Navarro *et al.* (2009) found that their *Ascaris* infection data best fitted the β-Poisson dose-response equation:

$$P_I(d) = 1 - [1 + (d/N_{50})(2^{1/\alpha} - 1)]^{-\alpha} \quad (1)$$

where $P_I(d)$ is the risk of infection in an individual who has ingested *d Ascaris* eggs on one occasion; N_{50} is the mean *Ascaris* infective dose; and α is an *Ascaris* 'infectivity constant'. They found the values of N_{50} and α to be 859 and 0.104, respectively (since they were working with epidemiological data on *Ascaris* prevalence, rather than conducting human *Ascaris* dose-challenge studies, the value found for N_{50} is not a measure of the actual median *Ascaris* infective dose, but rather an empirical value arising from their statistical analyses).

The annual risk of infection is given by:

$$P_{I(A)}(d) = 1 - [1 - P_I(d)]^n \quad (2)$$

where $P_{I(A)}(d)$ is the annual risk of infection in an individual resulting from n exposures per year to the single *Ascaris* dose d.

The Benke & Hamilton (2008) method for calculating the annual risk of infection firstly determines an annual risk of infection by doing a Monte Carlo simulation with the number of simulations set equal to the number of days of exposure per year (rounded down to an integral value); it then repeats this any required number of times and determines the resulting 50- and 95-percentile annual *Ascaris* infection risks.

A QMRA-MC computer program was written to determine median *Ascaris* infection risks pppy from the consumption by children under 15 of raw carrots which had been irrigated with wastewaters containing specified numbers of *Ascaris* eggs (the program is available at www.personal.leeds.ac.uk/ ~ cen6ddm/QMRA.html). A series of 10,000-trial QMRA-MC risk simulations was run and the resulting estimates of median *Ascaris* infection risk obtained and the assumptions on which they are based are given in Table 1. This shows that 1 egg per litre results in an *Ascaris* infection risk of $\sim 6 \times 10^{-3}$ pppy and 0.1 egg per litre in a risk of $\sim 6 \times 10^{-4}$ pppy; these risks are higher and lower, respectively, than the tolerable *Ascaris* infection risk of $\sim 10^{-3}$ pppy determined above. This could be taken to confirm the finding of Blumenthal *et al.* (1996) that ≤ 1 egg per litre is not protective of children under 15, and thus reinforce the recommendation of Blumenthal *et al.* (2000) that, when children under 15 are exposed, the guideline value should be ≤ 0.1 egg per litre. However, as noted in the 2006 WHO guidelines, post-treatment health-protection control measures can achieve significant pathogen reductions, so that wastewater treatment does not have to achieve the total pathogen reduction required to protect consumer health.

Implications for wastewater treatment

If it is assumed that, in areas where ascariasis is hyperendemic, untreated wastewater has an *Ascaris* egg concentration of 1,000 per litre, a 4-log unit egg reduction is required to achieve 0.1 egg per litre. Assuming that a 2-log unit reduction occurs through peeling carrots before consumption (WHO 2006), wastewater treatment is required to effect a reduction of only 2 log units from 1,000 to 10 eggs per litre. The Ayres *et al.* (1992) design equation for egg removal in WSP indicates that a 1-day anaerobic pond plus a 5-day facultative pond would reliably produce an effluent with 10 eggs per litre. Such a wastewater treatment requirement is not onerous, nor is it expensive.

Table 1 | Median *Ascaris* infection risks for children under 15 from the consumption of raw wastewater-irrigated carrots estimated by 10,000-trial Monte Carlo simulations*

Number of *Ascaris* eggs per litre of wastewater	Median *Ascaris* infection risk pppy	Notes
100–1,000	0.86	Raw wastewaters in hyperendemic areas
10–100	0.24	Raw wastewaters in endemic areas
1–10	2.9×10^{-2}	Treated wastewaters
1	5.5×10^{-3}	Wastewater quality required to comply with the 1989 and 2006 WHO guidelines
0.1–1	3.0×10^{-3}	Highly treated wastewaters
0.1	5.5×10^{-4}	Wastewater quality recommended by Blumenthal *et al.* (2000)
0.01–0.1	3.0×10^{-4}	Treated wastewaters in non-endemic areas

*Assumptions: 30–50 g raw carrots consumed per child per week (Navarro *et al.* 2009); 3–5 ml wastewater remaining on 100 g carrots after irrigation (Mara *et al.* 2007); $N_{50} = 859 \pm 25\%$ and $\alpha = 0.104 \pm 25\%$; no *Ascaris* die-off between final irrigation and consumption.

CONCLUSIONS

1. For a tolerable DALY loss of $\leq 10^{-5}$ pppy, the corresponding tolerable *Ascaris* infection risk is $\sim 10^{-3}$ pppy. In areas of hyperendemic ascariasis (i.e. where the *Ascaris* count is ~1,000 eggs per litre of raw wastewater) children under 15 who consume $\sim 30 - 50$ g of raw carrots per week can only be protected if the *Ascaris* egg count is reduced by 4 log units; they are not protected by a 3-log unit reduction.
2. A series of WSP comprising only a 1-day anaerobic pond and a 5-day facultative pond can reliably achieve a 2-log unit reduction of *Ascaris* eggs to produce an effluent quality of 10 eggs per litre. Produce peeling prior to consumption achieves the remaining 2-log unit *Ascaris* reduction.

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