Low risk for helminth infection in wastewater-fed rice cultivation in Vietnam

Do Thuy Trang, Wim van der Hoek, Phung Dac Cam, Khuong Thanh Vinh, Nguyen Van Hoa and Anders Dalsgaard

ABSTRACT

This study was done to assess the risk of helminth infection in association with wastewater-fed rice cultivation in an agricultural setting of Nam Dinh city, Vietnam. In a cross sectional survey data were collected for 202 households in a commune where wastewater was used for irrigation and for 201 households in a commune that used river water. Parasitological examination was conducted on single stool samples obtained from 1,088 individuals aged ≥15 years from the households. The irrigation water used in both communes was enumerated for helminth eggs and thermotolerant coliforms. The prevalence of infection with *Ascaris* spp., *Trichuris* spp., and hookworm was 42.2%, 19.9% and 10.5% respectively, with an overall prevalence of infection with any helminth of 53.4%. Surprisingly, the prevalence of infection with *Ascaris* and *Trichuris* was lower among people exposed to wastewater (containing 40–200 helminth eggs/l and 10^4 thermotolerant coliforms/100 ml) compared to people exposed to river water that contained lower worm egg and bacterial numbers. Poor sanitation and hygiene practices and not using protective measures were important independent risk factors for helminth infection. For hookworm infection, no significant difference was observed between the wastewater exposed and unexposed groups. Children living in the wastewater use area had a significantly better nutritional status than those in the area using river water. This suggests a generally higher welfare level of the wastewater use area. In conclusion, this study showed no evidence that rice cultivation with wastewater poses a risk for helminth infection. More detailed studies are needed on the reduction of fecal indicators and helminth eggs in peri-urban wastewater-irrigated rice culture systems and on the relative importance of wastewater irrigation compared to other risk factors for human helminth infection such as poor sanitation and poverty.

Key words | helminth infection, rice cultivation, risk factors, Vietnam, wastewater

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INTRODUCTION

Vietnam is one of the developing countries where wastewater has been used for decades and even centuries by farmers in urban and peri-urban areas as a cheap and reliable source of water and nutrients in agriculture as well as aquaculture systems. Products from wastewater-fed systems are produced at low cost, are generally accepted by the consumers, and seem to contribute significantly to food production and food security in Vietnam (Raschid-Sally *et al.* 2004). Despite the widespread use of wastewater, such practices are often informal and only to a limited degree recognized by the authorities and the public at large.

Being a low-income country, it is unlikely that Vietnam in the near future will be able to provide adequate wastewater treatment facilities for the growing urban population. Irrigation with untreated wastewater can

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therefore pose risks for the health of irrigators, for communities in prolonged contact with the wastewater, to the consumers of produce irrigated with wastewater and through the spread of pathogens in the environment. Epidemiological studies in different countries have established that the highest risk to human health of using wastewater in agriculture is posed by soil-transmitted intestinal helminth infections (Mara & Cairncross 1989; Blumenthal *et al.* 2000). However, little information is available about specific occupational risks for intestinal helminth infections associated with wastewater-irrigated rice culture. Infections with roundworm (*Ascaris*), whipworm (*Trichuris*), and hookworm are widespread in Vietnam (van der Hoek *et al.* 2003). But in addition to the use of wastewater, there are other environmental and human behavioral risk factors for infection with soil-transmitted helminths, of which poor sanitation is probably the most important one. The relative importance of the different risk factors in different wastewater use systems is unknown. This makes it difficult for policy makers and urban planners to make informed decisions about wastewater irrigation.

As part of a larger project on benefits and costs of wastewater use in agriculture, the risk for helminth infection among rice farmers was assessed in a peri-urban agricultural area of Nam Dinh, a medium sized provincial city, representative of many cities in northern Vietnam. The overall aim of this project was to optimize the benefits and reduce the potential negative impacts of wastewater use.

METHODS

Study area

The study was conducted in My Loc district of Nam Dinh province in northern Vietnam. The district was a peri-urban area of Nam Dinh city (population 230,000) and received city raw wastewater through a pumping station located in My Tan commune. Wastewater was used mainly for rice and vegetable cultivation, and partly for fish farming. The district had a mix of farming families exposed to wastewater and those who were not exposed, depending on the proximity of the households to the wastewater pumping station as well as to the water canal system. Two communes in the district, My Tan and My Trung, with different levels of wastewater use, were selected for the study. There was a widespread use of wastewater for irrigation in My Tan commune. Whereas in My Trung, because of the longer distance from the pumping station and the wastewater flow, mainly water from the Red River was used for irrigation and there was no apparent use of wastewater in agricultural and aquacultural activities.

Study population

Two hundred and twenty households were randomly selected from the official list of each commune. Households that were not involved in agriculture at all were excluded. This resulted in a study population of 202 households in My Tan commune (wastewater irrigation in rice cultivation) and 201 in My Trung commune (river water irrigation in rice cultivation). All females and males from 15 years of age living in these households were included for questionnaire interviews and stool collection for helminth eggs examination. Children were not included in the study as they were rarely engaged in rice cultivation.

Household questionnaire

A cross-sectional survey was conducted from late October until early November 2003. The selected households were surveyed using a household questionnaire administered to the head or the wife of the head of the households. The extent of contact with wastewater was assessed for all individuals ($\geq$15 years old and staying in the study area at the time of the study) on separate individual information sheets. It included the status of any contact with wastewater (yes or no), and in the case of having contact, the seasonal pattern of contact (seasonal or all year around) and average frequency of contact (days per month). This information was obtained either from the person him/herself or from the main respondent of the household in case the person was absent at the time of interview. Other information collected for each individual was age, sex, occupation, educational level, types of agricultural production involved, and the use of protective measures during agricultural work.

At household level information was obtained on household source and availability of drinking water, sanitation (latrine availability, hygienic status and waste handling), excreta use practices in agriculture/horticulture, and hygiene behavior, including hand washing practices. These household variables were then assigned to individuals with the assumption that all members of the same household shared the household characteristics and had similar hygiene practices.

The socioeconomic status (SES) of the households was assessed with a compound indicator that consisted of eight items pertaining to type of house construction, and ownership of a private well, cattle, motorbike, and telephone. Each of the items scored one point and each household could therefore obtain a maximum score of eight. Households with a score below the median were classified as having a low SES and those above the median were classified as having a high SES.

Stool sample collection and enumeration for helminth eggs

Sterilized glass vials containing 3 ml of 10% formalin were distributed to all eligible household members for the collection of stool samples at the end of the interviews. These vials were labeled with study ID numbers corresponding to those recorded in the individual information sheets. Household members were instructed how to collect fecal samples and forward them to the communal medical centers where the study team would pick them up the next day.

Samples were suspended in physiological saline solution and examined for eggs of *Ascaris* spp., *Trichuris* spp., and hookworm, using a standard direct smear method (Than, 1999). For samples that were positive for one or more helminth types, egg counts were expressed as eggs per gram of faeces (EPG).

Children's nutritional status

As part of a program on nutritional improvement, both communes had collected extensive data on child growth, particularly child weight on a routine basis. Such data were recorded monthly for children between 0 and 24 months of age, and every 6 months for those between 25 and 60 months of age. Although children were originally not the subject of the present study we felt that analyses of these secondary data could be important and reflect possible differences in the overall level of development between the two communes. Date of birth, sex, date of measurement, and weight (in kilogram), were collected for all children less than 5 years old in My Tan and My Trung communes that were weighed in October 2003. For children whose data were not available for that month, any value measured in either the closest preceding or subsequent month was used. The weights were then converted into Z-scores of weight-for-age, using the NUTSTAT module of EPI INFO 2000 software. This uses the WHO recommended United States National Center for Health Statistics (NCHS) sex-specific reference values.

Enumeration of helminth eggs and thermotolerant coliforms in irrigation water

Samples of the wastewater (in My Tan commune) and the river water (in My Trung commune) that was used for irrigation were collected for the enumeration of helminth eggs and thermotolerant coliforms in order to assess the level of exposure among rice farmers. Due to logistical reasons, the sampling could not be conducted at the same time as the household surveys and stool collections. However, we managed to do the water sampling at the same time one year later (2004), thereby representing similar seasonal irrigation practices and climatic conditions. A total of four wastewater and river water samples were collected during late October and November 2004. The samples were taken just before the pumping stations that distributed water for irrigation. Wastewater and river water were collected into 1-litre mixed samples (four single water samples taken with 2 hour intervals within one day were mixed into one composite sample to minimize temporal variations in numbers of helminth eggs and thermotolerant coliforms). The single samples were transported to Hanoi where they were mixed and analysed immediately after arrival (approximately 2 hours of transportation). For the enumeration of helminth eggs, the modified Bailanger method was employed (Ayres & Mara 1996). The most probable number (MPN) technique using lactose fermentation and gas production were used as identification and enumeration criteria for thermotolerant coliforms (ISO 1991).

Data analysis

Data were entered into a Microsoft Access database and analyzed with STATA 8 (STATA-corporation, College Station, USA). Univariate analysis was done for each helminth species separately as well as for the three helminth parasites combined to explore the impact of exposure to wastewater and other potential risk factors on helminth infections. Factors significantly ($P < 0.05$) associated with helminth infection and those factors that were considered important as shown in previous studies were fitted into a multivariate logistic regression model, providing exposure odds ratios. The stepwise procedure was applied for the logistic regression analysis, and only results from the reduced model were used for interpretation.

Ethical aspects

People participated in the study after informed consent. Ethical clearance for the study was provided by the Medical Ethics Committee of the National Institute of Hygiene and Epidemiology in Hanoi. All individuals with a positive stool sample for intestinal nematodes were informed after the examination and provided with adequate anti-helminth treatment free of charge.

RESULTS

Data from the irrigation water sampling showed that the microbiological quality of the river water was rather consistent during the sampling period, while that of wastewater varied between the four samplings. Nevertheless, as expected, wastewater contained higher concentrations of helminth eggs and thermotolerant coliforms (Table 1). The quality of wastewater (40–200 eggs/litre and 10^4 thermotolerant coliforms/100 ml) exceeded the WHO guidelines for wastewater application in agriculture and aquaculture, which stipulates less than 1 viable nematode egg per litre and less than 1,000 fecal coliforms per 100 ml (Mara & Cairncross 1989). The river water samples had a much lower level of thermotolerant coliforms, within the WHO permissible limit, and helminth eggs could not be found in 3 out of the 4 samples.

A total of 1,139 persons from the 403 households of My Tan (570 persons) and My Trung (569 persons) communes were included in the surveys. Their age ranged from 15 to 94 years old, with a mean age of 40.4 years in the wastewater exposed commune and 42.3 in the unexposed commune. There was no significant difference in the female/male ratio between the two communes. Compliance with stool examination was very high (95.5%) and out of the 1,088 samples that were available for analysis, 53.4% were positive for one or more of the three helminth parasites. Prevalence was 42.2% for *Ascaris* spp., 19.9% for *Trichuris* spp., and 10.5% for hookworm. There was no clear association between prevalence of helminth infection and age. However, it could be noted that for the age group of 60 years and above, the prevalence of infection with *Ascaris* spp. and *Trichuris* spp. decreased but the hookworm infection prevalence slightly increased (Figure 1).

The univariate analyses were performed with the data from the subjects that had provided a stool specimen and results are shown in Table 2. Surprisingly, people who were exposed to wastewater (all living in My Tan commune) had significant lower prevalence of *Ascaris* spp. and *Trichuris* spp. than people in My Trung

Table 1 | Microbiological quality of irrigation water in My Tan and My Trung communes in Nam Dinh province, Vietnam

		Helminth eggs (per 1 l)			Thermotolerant coliforms (per 100 ml)		
Commune	Number of samples	Range	Mean	SD	Range	Mean	SD
My Tan	4	40–200	85	77.2	1.1×10^3–1.5×10^4	5.1×10^3	6.6×10^3
My Trung	4	0–20	5	10	4–2.4×10^2	95	1.03×10^2

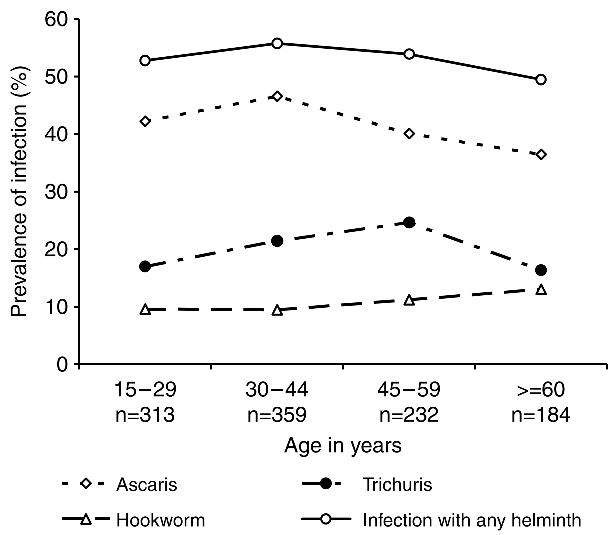

Figure 1 | Prevalence of *Ascaris* spp., *Trichuris* spp., and hookworm infections in My Tan and My Trung communes by age group.

commune and the people in My Tan commune who were not exposed to wastewater. For hookworm infection there was no significant difference between those exposed and not exposed to wastewater.

Not having a latrine was a strong risk factor for helminth infection (odds ratio [OR] for the three helminth parasites combined was 2.57, 95% CI 1.10–6.70). However, only 31 out of the 1,088 people had no latrine available. Therefore, in the further analyses we used the hygienic status of the latrine, with people not having a latrine regarded as a separate category, although its OR would not be used to interpret the results. People from households that did not apply any treatment on latrine waste after defecation (direct discharge) had an increased risk of infection with *Ascaris* spp. and *Trichuris* spp., such as those who used overhung fishpond latrines or disposed feces directly to open surface drains (OR for combined infection was 1.99, 95% CI 1.37–2.81).

Having only a limited storage capacity for drinking water (less than $2\,m^3$) was a risk factor for helminth infection. Presumably this reflects the amount of water that people had available for personal and household hygiene. Those individuals who practised hand washing after defecation experienced a lower risk of infections with *Ascaris* spp. and *Trichuris* spp.. In particular, hand washing with soap seemed to further reduce the risk of helminth infection. However, only the association between *Ascaris* spp. infection and no handwashing was found statistically significant (Table 2). People using nightsoil, including fresh excreta on their agricultural fields, had a higher risk for helminth infection than those not using nightsoil, but the difference did not reach statistical significance (data not shown). Of the people involved in agricultural work (1,018 people), only 4.4% reported using personal protective measures while doing fieldwork, such as shoes, plastic boots or gloves. These people had a much lower risk of *Ascaris* and *Trichuris* infection than those not using any means of personal protection. Significant differences in the use of overhung fishpond latrines, non-treatment of latrine waste and the lack of protective measures were also observed, with conditions being worse in My Trung than in My Tan.

Socio-economic status (SES) was significantly associated with *Ascaris* spp. and *Trichuris* spp. infections but not with the combined infection of helminths. Households in My Trung were generally of lower SES than in My Tan ($P < 0.05$). The risk for helminth infection was higher in people with lower levels of education (ranging from illiteracy to secondary school) compared to people of higher education (high school or university levels). No significant gender difference in risk of infection was found.

Specific agricultural practices, such as rice and vegetable farming or aquaculture, could not be associated with risk for helminth infection, except for flower planting that gave an increased risk (OR = 1.37, $P = 0.078$, controlling for factor "commune"). However, this could not explain the difference in prevalence of helminth infections between the two communes because flower cultivation was more common among the households in My Tan commune than in My Trung commune. No significant differences between the two communes were found in the status of latrines, use of nightsoil or educational level.

Most risk factors pointed in the same direction for the three helminth parasites. Factors that were significantly associated with any of the helminth species were also significant for single infection with *Ascaris* spp. or *Trichuris* spp.. Unlike for *Ascaris* spp., *Trichuris* spp., and for the three parasites combined, few risk factors for hookworm could be identified. This could be due to the different way of transmission where hookworm depends on the penetration

Table 2 | Risk factors for helminth infections in Nam Dinh province, Vietnam ($N = 1,088$)

Variables	N	*Ascaris* spp.		*Trichuris* spp.		Hookworm	
		OR[a]	95% CI	OR	95% CI	OR	95% CI
Wastewater use							
Yes	495	0.42	0.32–0.54	0.45	0.32–0.63	0.93	0.61–1.40
No	593	1		1		1	
Latrine availability*							
No	31	3.47	1.52–9.63	2.62	1.14–5.79	0.91	0.17–3.03
Yes	1,057	1		1		1	
Latrine status*							
Unhygienic	581	1.56	1.21–1.99	1.20	0.88–1.63	1.00	0.67–1.48
No latrine	31	4.44	2.00–9.86	2.91	1.36–6.21	0.91	0.27–3.11
Hygienic	476	1		1		1	
Use of overhung latrines and direct discharge to drain*[b]							
Yes	154	1.90	1.33–2.72	1.45	0.95–2.19	0.98	0.52–1.74
No	928	1		1		1	
Treatment of latrine waste*							
No	188	2.00	1.44–2.79	1.48	1.00–2.17	1.02	0.58–1.72
Yes	900	1		1		1	
Hand washing*							
No	347	3.97	1.11–14.19	2.13	0.47–9.58	2.01	0.26–15.62
Yes without soap	725	2.89	0.82–10.23	1.59	0.36–7.07	1.65	0.22–12.70
Yes with soap	16	1					
Use of protective measures							
No	973	1.69	1.10–2.63	2.08	1.13–4.12	1.12	0.58–2.39
Yes	115	1		1		1	
Availability of drinking water*							
$\leq 2\,m^3$	153	1.71	1.19–2.45	1.40	0.91–2.11	0.997	0.53–1.77
$> 2\,m^3$	935	1		1		1	
Level of education							
Low	177	1.48	1.04–2.10	1.06	0.70–1.64	1.21	0.69–2.26
High	911	1		1		1	
Socioeconomic status*							
Low	548	1.31	1.02–1.69	1.51	1.10–2.06	1.02	0.68–1.54
High	540	1		1		1	
Gender							
Female	608	1.05	0.82–1.35	1.02	0.75–1.39	1.4	0.92–2.15
Male	480	1		1		1	

*Variables at household level that were assigned to all individuals in the household.
[a]OR = odds ratio.
[b]Excluding people using latrine of neighbors ($N = 1,082$).

of human skin (usually the feet) by larvae, which develop from eggs passed in human stools into the environment. However, the much lower prevalence of hookworm compared with *Ascaris* could in itself explain why most risk factors did not reach statistical significance.

Because of a similar oral transmission pattern, the infections with *Ascaris* spp. and *Trichuris* spp. were combined as the outcome variable in the multivariate analysis (Table 3). Exposure to wastewater remained a significant protective factor, even when controlling for confounding by other variables. This indicates that other differences between the two communes than the ones considered in the study had impact on the prevalence of helminth infections. Poor standard of hygiene and sanitation, not using protective measures, and lack of access to large quantities of water, were independent risk factors in the multivariate analysis (Table 3). Because not every subject did participate in agricultural production, a separate logistic regression model was run, including only people doing farm work ($N = 1,018$), to assess the effect of not using protective measures. However similar results were obtained with only slight changes in the odds ratios (data not shown). Variables that were significant in this model also stayed significantly in the other. Adding interaction terms of covariates did not improve the fit of the multivariate model (data not shown).

The helminth egg counts were low for all helminths (Table 4). Except for a few infections with *Ascaris* spp. that would be classified as "moderate", all other infections would be classified as "light" according to WHO criteria (2002).

There were clear differences in nutritional status between the two communes. The mean Z-score of weight-for-age for children in My Tan was significantly ($P < 0.01$) higher than that of children in My Trung. The percentage of children underweight (<-2SD from the median of the NCHS reference) was 20.7% in My Tan and 27.8% in My Trung (Chi squared test = 5.45, $P = 0.02$). Children living

Table 3 | Assessment of risk factors for helminth infections (*Ascaris* spp. and *Trichuris* spp. combined) using multivariate logistic regression ($N = 1,088$)

	***Ascaris* spp. and *Trichuris* spp.**			
Variables	**OR***	***P*-value**	**OR****	***P*-value**
Wastewater use	0.40 (9.32–0.52)	0.000	0.36 (0.28–0.47)	0.000
Hygienic status of latrine	1.49 (1.17–1.89)	0.001	1.31 (1.00–1.71)	0.051
Nightsoil use	1.27 (0.91–1.77)	0.166	1.17 (0.81–1.69)	0.410
Use of fresh excreta	1.31 (1.00–1.71)	0.046	n/a	
Treatment of latrine waste	1.97 (1.43–2.73)	0.000	n/a	
Hand washing	1.34 (1.04–1.73)	0.026	1.48 (1.33–3.04)	0.005
Use of protective measures[a]	1.78 (1.20–2.66)	0.004	2.01 (1.34–3.09)	0.001
Availability of drinking water	1.71 (1.20–2.43)	0.003	1.45 (0.98–2.14)	0.060
Level of education	1.49 (1.08–2.06)	0.016	1.35 (0.96–1.91)	0.084
Socioeconomic status	1.29 (1.02–1.64)	0.034	1.05 (0.81–1.36)	0.704

*Crude OR.
**From the reduced model.
n/a: the variable was dropped out during the stepwise multivariate analysis so there was no OR obtained in the reduced model.
[a]People who did not do agricultural work were included in the group that used protective measures since it was assumed that they would probably do so when performing some fieldwork.

Table 4 | Intensities of infections with *Ascaris* spp., *Trichuris* spp. and hookworm in My Tan and My Trung communes

	Infected people		Whole population		Range	
Parasite	**n**	**EPG[a] geometric mean (95% CI)**	**N**	**EPG geometric mean (95% CI)**	**Minimum**	**Maximum**
Ascaris spp.	459	345.9 (312.4–382.9)	1,088	10.8 (8.87–13.15)	0	11,000
Trichuris spp.	217	78.7 (72.6–85.4)	1,088	1.39 (1.16–1.66)	0	600
Hookworm	114	86.0 (75.1–98.4)	1,088	0.60 (0.38–0.73)	0	800

[a]Egg count per gram.

in the non-wastewater site had an increased risk of being underweight (OR = 1.47, 95% CI 1.05–2.06).

DISCUSSION

Contrary to what was expected, people who were exposed to wastewater had a lower prevalence of helminth infection than those who were not exposed. The study could therefore not find evidence that rice cultivation with urban wastewater posed a risk for intestinal helminth infection in agricultural workers. Despite taking account of differences in socio-economic status and sanitary practices, the variables included in the study could not fully explain the difference in infection prevalence between the exposed and non-exposed. The most probable explanation for the lower prevalence of helminth infection in My Tan commune would still be the overall higher welfare level as compared with My Trung. This was reflected in the better child nutritional status in the exposed commune. Nutritional status is a well-known compound indicator representing effects of access to food, standards of living, womens' educational level, access to water supply and sanitation, and burden of infectious diseases. These aspects might have been insufficiently captured in the variables that we used. This also bears on possible flaws in the design of the study. The study was designed as a cross-sectional study with analysis at the individual level. However, one could criticize the approach as being a comparison between an exposed and an unexposed community. Such a "one to one comparison" is a major methodological problem in many studies on environmental sanitation, as pointed out by Blum & Feachem (1983). Our interest was not the difference between the two communities but the relative importance of different risk factors throughout the entire study population. We felt that the chosen design was justifiable because the communities were located very close to each other and were similar in many aspects. Another limitation of the study is that some of the variables pertained to individuals while other variables pertained to all members in a household. It is well known that helminth infections tend to aggregate in certain individuals within households, in certain households, and in certain communities. We did not account for this aggregation. Especially aggregation of heavy infections is extremely relevant for targeting control measures but we feel that this phenomenon of aggregation did not affect the validity of the individual-level risk assessments.

Our analyses showed that most of the people infected with *Ascaris* spp. and *Trichuris* spp. came from lower socioeconomic groups and households with inadequate sanitary conditions and poor hygiene practices. This is in line with findings from previous studies such as by Kightlinger *et al.* (1995), Cifuentes (1998), and Al-Shammari *et al.* (2001).

Another possible explanation of the counterintuitive findings of the present study is that the concentration of helminth eggs in the irrigation water might have been too low to cause an increase in human helminth infection prevalence. Although egg counts in wastewater samples exceeded the WHO guideline level, the samples were collected just before the wastewater entered the pumping station, from where the wastewater was distributed to local irrigation canals and then to the paddy fields. After the pumping station, the helminth eggs in wastewater would have been diluted because of natural sedimentation processes along the wastewater flow, together with the die-off of eggs because they do not have optimal conditions for their survival and development, which is a moist, warm and shady environment (Paniker 2002). This could have resulted in a low concentration of helminth eggs in the water in the fields, to which the farmers were actually exposed. The quality of the wastewater in My Tan commune was to some extent similar to or better than that studied elsewhere, such as in the Mezquital Valley (Mexico) with concentrations of 10^8 thermotolerant coliforms/100 ml and 90–150 helminth eggs/litre (Cifuentes 1998) or in Calcutta with 200–2130 helminth eggs/litre (Ayres *et al.* 1992). Furthermore, it could be questioned whether farmers using wastewater in wet rice cultivation would be exposed to any particular risk for *Ascaris* spp. and *Trichuris* spp. infections, since such farmers must have an oral intake of the eggs present in the wastewater to acquire the infection. This is different from wastewater irrigated crops, including crops consumed raw, where an oral intake of eggs will mainly occur through consumption of contaminated produce.

Our findings of lower prevalence of helminth infections in a wastewater dependent area are inconsistent with other studies of the effect of wastewater on enteric infections. In a study in Mexico, Blumenthal *et al.* (2001) reported that the risk of infection with *A. lumbricoides* in those aged >5 years when directly exposed to untreated wastewater (6×10^7 thermotolerant coliforms/100 ml and 90–135 helminth eggs/litre) was 13.5 times higher than for those who were not exposed. However, the prevalence of *Ascaris* spp. infection in adults above 15 years of age in the exposed group (4.3%) was much lower than what was found in the present study in Vietnam. Living in a wastewater using area was also found to be associated with ascariasis, but not with trichuriasis, in children in Morocco (Habbari *et al.* 2000). That study also found low intensities of the helminth infections in wastewater as well as non-wastewater areas.

The prevalence of infection for each of the three helminths in the present study was lower than the previously reported national estimates, which were 44.4% for infection with *Ascaris* spp., 23.1% for *Trichuris* spp. and 28.6% for hookworm (van der Hoek *et al.* 2003). That nationwide review did not have data for Nam Dinh province, but comparison with recent studies on the prevalence of nematode infections conducted in the neighboring areas of Nam Dinh province, such as Ha Nam province (Needham *et al.* 1998) or Hoa Binh province (Verle *et al.* 2003), also showed lower infection prevalence. Although these studies mainly focused on the prevalence and intensities of helminth infections, but not risk factors for the infections (except age and sex), it remains unexplainable to us why the prevalence in Nam Dinh was so low, particularly the prevalence of hookworm infection, while about 90% of the farmers reported not wearing any protective measures when working in the fields. It could be speculated that hookworm larvae in an aquatic wastewater irrigated rice culture environment may be less able to penetrate the skin of farmers compared to a moist soil environment. This warrants further studies.

The changes in prevalence patterns of the three helminths in relation to age reflected their epidemiological characteristics. The decrease in the prevalence of *Ascaris* spp. and *Trichuris* spp. infections in the group of people ≥ 60 years old is likely to correspond with the epidemiology of these two intestinal nematodes. Peak prevalence and intensity of infections of both *Ascaris* spp. and *Trichuris* spp. often occur in school-aged children and decline with age (Bundy *et al.* 1987). The similar patterns of infections of these two species were presumably reflecting their same mode of infection through the fecal-oral route. Although in our study the peak of *Trichuris* spp. infection was with the 45–59 age class, a marked decline in the prevalence of both species was observed in the oldest age group. Bundy *et al.* (1987) also noted that often in the adult groups, the prevalence of *Trichuris* maintained a relatively constant value throughout adulthood, while that of *Ascaris* declined when people got older, and there were sometimes indications of age-dependent increase in the younger age groups of adulthood for both species. For hookworm, we observed an increase in the prevalence of infection in the age group of 60 and above. It is because the infection with hookworm is largely independent of the other two helminths, where infection occurs via the skin penetration by the larvae. Our finding was also inline with previous studies (Needham *et al.* 1998; Verle *et al.* 2003) where the prevalence of hookworm infection was found to increase with age. This may be explained by the fact that adults spend more time in the muddy rice fields and the hookworm larvae therefore accumulate over time to increase the worm load in the human body.

Nightsoil use was a household variable. Because 85% of households used nightsoil, this household-level variable did not provide an important exposure contrast within the study population. Nightsoil use is an established risk factor for hookworm infection (Humphries *et al.* 1997) and an individual-level variable might have shown a different picture. However, 74% (684/926) of individuals in the study population came from the households that used nightsoil after a composting process. About 71% of these 684 individuals lived in households with compost durations greater than 3 months. The length of composting could have allowed for the degeneration of helminths eggs, thereby reducing the risk of helminth infections from exposure to nightsoil use.

CONCLUSION

The counterintuitive finding of lower risk for helminth infection among people exposed to wastewater means that no clear recommendations relevant to health policy and urban planning can be given based on this study. It does however emphasize the need for more detailed studies on the reduction of fecal indicators and helminth eggs in peri-urban wastewater-irrigated rice culture systems.

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