Intestinal parasitosis and use of untreated wastewater for agriculture in Settat, Morocco

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الإصابة بالطفيليات المعوية واستخدام مياه الفضلات غير المعالجة في الزراعة في سطات بالمغرب

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الخلاصة: قام الباحثون بتقييم اختطار التلوث بطفيليات البراز في السكان، من جراء استخدام مياه الفضلات في الزراعة. واعتمدت الدراسة على ثلاث مجموعات ريفية في المنطقة الشمالية لمدينة سطات بالمغرب: تعرَّضت مجموعتان ريفيتان منها لمياه الفضلات غير المعالجة في الزراعة، أما المجموعة الثالثة فلم تتعرَّض لها. وشملت العينة 333 شخصاً تتراوح أعمارهم بين سن الثلاثين والستين فما فوق: تعرَّض 214 منهم لمياه الفضلات، و119 لم يتعرَّضوا لها. وقام الباحثون بأخذ عيِّنات البراز، وفحصها للكشف عن الطفيليات المعوية. وأظهرت نتائج الدراسة أن معدَّل انتشار الإصابة بالطفيليات المعوية (بمعنى إثبات وجود نوع واحد على الأقل من الطفيليات) في المجموعة التي تعرَّضت لمياه الفضلات غير المعالجة، هو أعلى بكثير منه في المجموعة التي لم تتعرَّض لها، (66.4٪ في مقابل 31.9٪ باختطار نسبي 2.1). كما أن مَنْسَب الاحتشاد الأُسَري كان أعلى في المجموعة التي تعرَّضت لمياه الفضلات غير المعالجة.

ABSTRACT An evaluation was made of the risk of contamination by faecal parasites in the population from the use of wastewater in agriculture. The study was based in 3 rural clusters in the northern region of Settat city, Morocco: 2 rural clusters exposed to untreated wastewater in agriculture and 1 not exposed. The sample comprised 333 people aged 3–60+ years: 214 exposed and 119 non-exposed. Stool samples were collected and examined for intestinal parasites. The results showed that the prevalence of intestinal parasitosis (i.e. evidence of at least 1 type of parasite) in the exposed group was significantly higher that the non-exposed group (66.4% versus 31.9%; relative risk 2.1). The household crowding index was also higher in the exposed group.

Parasitose intestinale et utilisation d'eaux usées non traitées dans l'agriculture à Settat (Maroc)

RÉSUMÉ Une évaluation du risque de contamination de la population par des parasites fécaux du fait de l'utilisation d'eaux usées dans l'agriculture a été réalisée. L'étude portait sur trois grappes (douars) rurales de la région de Settat, au nord du Maroc : 2 exposées à des eaux usées non traitées dans l'agriculture et 1 non exposée. L'échantillon était composé de 333 sujets âgés de 3 à 60 ans et plus dont 214 exposés et 119 non exposés. Des échantillons de selles ont été prélevés et une recherche de parasites intestinaux a été effectuée. Les résultats montrent que la prévalence de la parasitose intestinale (établie par la présence d'au moins un type de parasite) dans le groupe exposé était significativement plus élevée que dans le groupe non exposé (66,4 % contre 31,9 % ; risque relatif : 2,1). L'indice de promiscuité était également plus élevé dans le groupe exposé.

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Introduction

In many areas of the world that are affected by water shortages, untreated wastewater is reused for agricultural purposes. In Morocco, this practice is well established in areas close to large cities. The technique has developed in recent years, due to a number of factors, such as the increasing deficiency in irrigation water, the prevalence of drought, agricultural intensification and the high cost of industrial fertilizers. Other factors are the belief in the nutrient value of the wastewater and social acceptance of this practice [*1,2*]. In the case of Morocco, approximately 70 million m^3 of untreated wastewater are used each year without any sanitary precautions to irrigate an area of more than 7000 hectares. Various crops are irrigated in this way, including animal fodder, vegetables, field crops and fruit trees, even though Moroccan legislation prohibits the use of wastewater to irrigate certain crops, such as vegetables [*3,4*].

The most common medical risk inherent in this practice is transmission of intestinal parasites, pathogenic or opportunist, from the excrement of humans and animals when contaminated water or food is consumed. [*5,6*]. The daily average amount of excrement of human origin is 1 kg/person [*7*]. Intestinal parasites are the causes of diarrhoeal diseases and malabsorption syndromes. The negative impact on health, particularly that of children, in the form of dehydration, poor nutrition and disorders of growth, can be considerable. The World Health Organization (WHO) has classified the absence of all helminth eggs as one of the standards for agricultural reuse of purified wastewater [*5,6*].

In Settat, domestic and industrial wastewater is drained without any treatment into a creek, Oued Bou Moussa. At the outskirts of the city, the creek crosses the localities of Dladla and Boukallou, where the residents use the water for the irrigation of cereal and fodder crops. The first signs of contamination of groundwater have been detected in these areas [*8*].

An association between crowding within the residence and contamination by intestinal parasites has been noted by several authors [*9–15*].

The present study aimed to evaluate the risk of intestinal parasitosis from the use of wastewater in agriculture in the Settat area and to assess the relationship between the prevalence of intestinal parasites and household crowding.

Methods

Study area

Settat is located in north-western Morocco, about 60 km south of Casablanca. Its climate is arid to semi-arid. Average annual rainfall is around 284 mm with a strong intra- and inter-annual variability (coefficient of variation exceeding 30%). The city of Settat has 120 000 inhabitants. It produces approximately 120 tons of excrement and approximately 9000 m^3 of wastewater per day. This non-treated wastewater is reused to irrigate more than 400 hectares of arable land.

Study population

This cross-sectional study compared a population exposed to agricultural reuse of wastewater with a population not exposed to this. The 2 populations have similar ethnic, demographic and socioeconomic characteristics and also the same culinary and animal husbandry practices. The exposed group was the inhabitants of 2 rural clusters, Dladla and Boukallou, located in north Settat in the district of Sid El Aydi who use the water of Oued Bou Moussa for irrigation and consume the adjacent groundwater. The non-exposed population was the inhabitants of the rural cluster Ouled Afif, located 20 km south-east of Settat, who never use wastewater in irrigation and consume water from different groundwater sources than that of Sid El Aydi.

The study sample comprised 333 people: 214 exposed and 119 non-exposed. A sample of a third of the population in each rural cluster was randomly chosen to be representative by sex and age. The rate of non-response for the exposed and the non-exposed groups were 6.1% and 6.3% respectively. Reasons for non-participation included extended absences from home, refusal to participate, constipation and anxiety about giving stool samples.

All the surveyed individuals lived on houses built on solid foundations. The water used for human consumption in the exposed group came from 6 individual wells and 8 collective wells. For the non-exposed group, the water came from a local distribution network, from a well provided with a motor-driven pump, which fed a castle-tank, regularly treated by chlorination. There was no system of collection of household refuse or of local drainage of wastewater. Only water coming from a distribution network approved by the Office National de l'Eau Potable (National Office of Drinking Water) is considered drinkable. This organization ensures the production, quality control and distribution of water to homes or, in zones difficult to reach, to public fountains.

Data collection

Subjects taking part in the investigation gave informed consent for participation. They were informed about the aims of the study, its methodology, its constraints, the agreement of the Ministry for Health and the confidentiality of the information provided and the right to freely withdraw from the study.

Each subject was given a complete clinical examination and data were collected about socioeconomic and professional status (age, sex, education, number of people in the household), hygiene practices (presence of toilets within the residence and practice of hand-washing before eating), exposure to wastewater (source of water for consumption) and history of previous pathology (such as frequent occurrence of diarrhoea and stomach disorders; data not shown).

Stool samples were collected from each participant on 3 consecutive days into a transparent pot. The parasitological examination of the stools was carried out immediately using direct macroscopic and microscopic examination and after concentration according to the diphasic standard method of Bailenger [*16*]. Subjects were defined as having intestinal parasitosis when examination of the stool revealed the presence of at least 1 type of intestinal parasite, whether adult eggs or larvae in the case of helminths, or vegetative or cystic forms in the case of protozoa. Polyparasitism was the coexistence in the same person of 2 or more types of parasite.

The crowding index was measured by the number of individuals living in the household divided by the number of rooms in the house [*17*].

Analysis

The study subjects were classified into age groups 3–9 years (young children, in education or not, living under the family roof and depending entirely on the parents for food); 10–14 years (older children, in education or not); 15–29 years (adolescents and young adults); 30–59 years (adults) and ≥ 60 years (elderly people) [*9*]. The crowding index was arbitrarily divided into 3 classes: 1–1.9 2–2.9 and ≥ 3.

The chi-squared test was used to compare the prevalence obtained according to various characteristics. An association was considered significant when the *P*-value was < 0.05. For the evaluation of the association between possible risk factors and parasitosis, the ratio of prevalence or relative risk was used. The reference population was the non-exposed group [*18*].

Results

Characteristics of study population

The 214 subjects of the exposed group had a mean age of 28.6 [standard deviation (SD) 19.4] years; 52.8% of them were female and 47.2% male. The 119 subjects of the non-exposed group had a mean age of 31.8 (SD 19.5) years; 50.4% were female and 49.6% male. Table 1 shows that the distribution of the surveyed people by sex, age group, education and crowding index was similar between the 2 groups.

Prevalence of intestinal parasitosis

The prevalence of intestinal parasitosis in the exposed group was 66.4% and in the non-exposed group was 31.9% (Table 2). The difference between the 2 study groups was significant ($P < 0.001$). The relative risk was 2.1.

Among the exposed group, the prevalence of polyparasitism was 17.3%. Among the non-exposed group, this prevalence was only 2.5%. This difference was also statistically significant ($P < 0.001$). The relative risk was 6.9.

The prevalence of parasitosis in the exposed group and the non-exposed groups were 67.3% and 30.0% among females and 65.3% and 33.9% among males respectively (Table 2). The relative risk for was 1.9 males and 2.2 for females. No significant difference in the prevalence of parasitosis was noted between the groups by sex, age group, education or crowding index.

Effects of hygiene practices on prevalence of parasitosis

In the exposed group, there was no significant difference in the prevalence of intestinal parasitosis comparing people using collective wells (n = 174) and those using individual wells (n = 40) (Table 2). The drinking water of the non-exposed group came from a collective water tank.

The prevalence of intestinal parasitosis was very slightly higher in people who had no toilet in their residence, but the difference was not significant. However, it was noted that some people who had toilets reported they did not use them because they had problems of maintenance and hygiene or they reserved them for guests.

The results for hand-washing practices were not included as observations by the researchers cast some doubt on the reliability of the responses.

Parasites identified

The 3 types of amoeba found in stool samples were *Entamoeba coli*, *Endolimax nana* and *Pseudolimax butschlii* (Table 3). All are non-pathogenic amoebae. No cases of *Ent. histolytica* were recorded. The prevalence of *Ent. coli* was highest: 52.3% of the exposed group versus 26.1% of the non-exposed group.

Analaysis for flagellates revealed some cases of *Giardia intestinalis*, which are pathogenic, and *Chilomastix mesnili*. The prevalence of *G. intestinalis* was 11.7% in the exposed group and 2.5% in the non-exposed group.

The helminths identified were mainly *Ascaris lumbricoides* with a prevalence of 4.2% in the exposed group and 0.0% in the non-exposed group. Only 1 case of

Table 1 **Selected sociodemographic characteristics of the 2 study groups (exposed and non-exposed to agricultural reuse of untreated wastewater) in Settat, Morocco**

Variable	Exposed No.	Non-exposed No.	χ^2-test	*P*-value
Sex				
Female	113	60	0.17	0.67
Male	101	59		
Age group (years)				
3–9	30	15	4.30	0.36
10–14	31	11		
15–29	68	33		
30–59	67	46		
60+ years	18	14		
Education				
Educated	109	69	1.52	0.92
Illiterate	105	50		
Crowding index[a]				
1–1.9	58	20	4.53	0.10
2–2.9	92	59		
≥ 3	64	40		
Toilet in house				
Yes	171	103	2.31	0.12
No	43	16		
Total	214	119		

[a]*No. of people in household/no. of rooms.*

Enterobius vermicularis was found in the exposed group.

Discussion

The use of urban wastewater in agriculture is a common practice, not only in arid and seasonally arid zones but also in countries that are not water-short. The reasons for this are diverse and depend on the situation and local context. For instance, in some countries, wastewater is used for its water value even in its untreated form, and as a source of plant nutrients. In others, it is used because of the absence of an alternative unpolluted source of water. The added benefit of its fertilizer value is incidental. The reasons for wastewater use, the diverse conditions under which it is used and its impact are still not clearly understood and require further research before ways to improve the situation can be suggested. Concern for human health and the environment are the most important constraints in the reuse of wastewater. While the risks do need to be carefully considered, the importance of this practice for the livelihoods of countless small-holders must also be taken into account.

Table 2 **Prevalence of intestinal parasitosis in the 2 study groups (exposed and non-exposed to agricultural reuse of untreated wastewater) according to sociodemographic characteristics and source of water consumed, Settat, Morocco**

Variable	Prevalence of intestinal parasitosis[b] Exposed (*n* = 214) +ve	–ve	%	Non-exposed (*n* = 119) +ve	–ve	%	Relative risk (RR)	95% CI of RR	Statistics
Sex									
Female	76	37	67.3	18	42	30.0	2.24	1.49–3.37	$\chi^2 = 21.8$; ***P*** < 0.001
Male	66	35	65.3	20	39	33.9	1.93	1.31–2.83	$\chi^2 = 14.7$; ***P*** < 0.001
Statistics	$\chi^2 = 0.09$; ***P*** = 0.76			$\chi^2 = 0.21$; ***P*** = 0.64					
Age group (years)									
3–9	19	11	63.3	4	11	26.7	2.38	0.98–5.74	$\chi^2 = 5.26$; ***P*** = 0.02
10–14	20	11	64.5	2	9	18.2	3.55	0.99–12.7	$\chi^2 = 6.82$; ***P*** = 0.009
15–29	46	22	67.6	16	17	48.5	1.40	0.95–2.06	$\chi^2 = 3.41$; ***P*** = 0.06
30–59	44	23	65.7	15	31	32.6	2.01	1.28–3.16	$\chi^2 = 11.84$; ***P*** = 0.0005
60+	13	5	72.2	1	13	7.1	10.11	1.50–68.3	$\chi^2 = 13.13$; ***P*** = 0.0002
Statistics	$\chi^2 = 0.51$; ***P*** = 0.97			$\chi^2 = 9.28$; ***P*** = 0.05					
Education									
Educated	66	43	60.6	22	47	31.9	1.90	1.30–2.77	$\chi^2 = 13.81$; ***P*** < 0.0003
Illiterate	76	29	72.4	16	34	32.0	2.26	1.48–3.45	$\chi^2 = 22.75$; ***P*** < 0.0001
Statistics	$\chi^2 = 3.34$; ***P*** = 0.06			$\chi^2 = 0.00$; ***P*** = 0.98					
Crowding index[a]									
1–1.9	33	25	56.9	7	13	35.0	1.63	0.86–3.08	$\chi^2 = 2.85$; ***P*** = 0.09
2–2.9	66	26	71.7	19	40	32.2	2.23	1.51–3.30	$\chi^2 = 22.69$; ***P*** < 0.0001
≥ 3	43	21	67.2	12	28	30.0	2.24	1.35–3.71	$\chi^2 = 13.53$; ***P*** < 0.001
Statistics	$\chi^2 = 3.54$; ***P*** = 0.17			$\chi^2 = 0.16$; ***P*** = 0.92					
Toilet in house									
Yes	116	55	67.4	33	70	31.3	2.12	1.57–2.86	$\chi^2 = 33.08$; ***P*** < 0.0001
No	29	14	67.8	5	11	32.0	2.16	1.01–4.60	$\chi^2 = 6.15$; ***P*** = 0.013
Statistics	$\chi^2 = 0.00$; ***P*** = 0.96			$\chi^2 = 0.00$; ***P*** = 0.95					
Source of water (well)									
Collective	117	57	67.2			–	–		
Individual	25	15	62.5			–	–		
Statistics	$\chi^2 = 0.00$; ***P*** = 0.56								
Total	142	72	66.4	38	81	31.9	2.08	1.57–2.75	$\chi^2 = 36.38$; ***P*** < 0.0001

[a]*No. of people in household/no. of rooms.*
[b]*Evidence of at least 1 type of intestinal parasite in faeces.*
n ***= total number of subjects.***
CI = confidence interval.

To assess the impact of the use of wastewater on human health, we determined the prevalence of intestinal parasitosis in a population exposed to wastewater used in agriculture and a control population who were not exposed. The results showed that the parasitic load was 66.4% within the exposed group but 31.9% in the non-

Table 3 **Prevalence of intestinal parasitosis in the 2 study groups (exposed and non-exposed to agricultural reuse of untreated wastewater), Settat, Morocco**

Intestinal parasite	Prevalence of intestinal parasitosis[a] Exposed (*n* = 214)	Non-exposed (*n* = 119)
Rhizopods (amoebae)		
Entamoeba coli	52.3	26.1
Endolimax nana	12.1	0.0
Pseudolimax butschlii	6.5	3.4
Total	66.4	29.4
Flagellates		
Giardia intestinalis	11.7	2.5
Chilomastix mesnili	1.9	2.5
Total	13.6	5.1
Helminths (nematodes)		
Ascaris lumbricoides	4.2	0.0
Enterobius vermicularis	0.5	0.0
Total	4.7	0.0

[a]***Evidence of at least 1 type of intestinal parasite in faeces.***
n ***= total number of subjects.***

exposed group. The prevalence of intestinal parasitosis in the non-exposed group was lower than that in a 1996 study undertaken in Ouarzazate with 221 individuals, 112 exposed to wastewater purified after treatment by oxidation ponds and 109 with conventional water. It showed that the prevalence of intestinal parasites was similar in the 2 groups with 64.6% for the reference group and 56.7% for the exposed group ($P < 0.2$) [*16*]. Our findings were also similar to those observed in other studies in rural environments in Morocco (Figure 1). But the prevalence observed within the exposed group in Settat was higher than those reported in urban environments [*13,19–21*].

The prevalence of intestinal parasitosis in the exposed group among children aged 3–9 years and 10–14 years were 60.0% and 62.5% respectively, whereas in the non-exposed group the rates were 26.7% and 36.4% respectively. Similar prevalence rates ranging between 69.9% and 87.9%

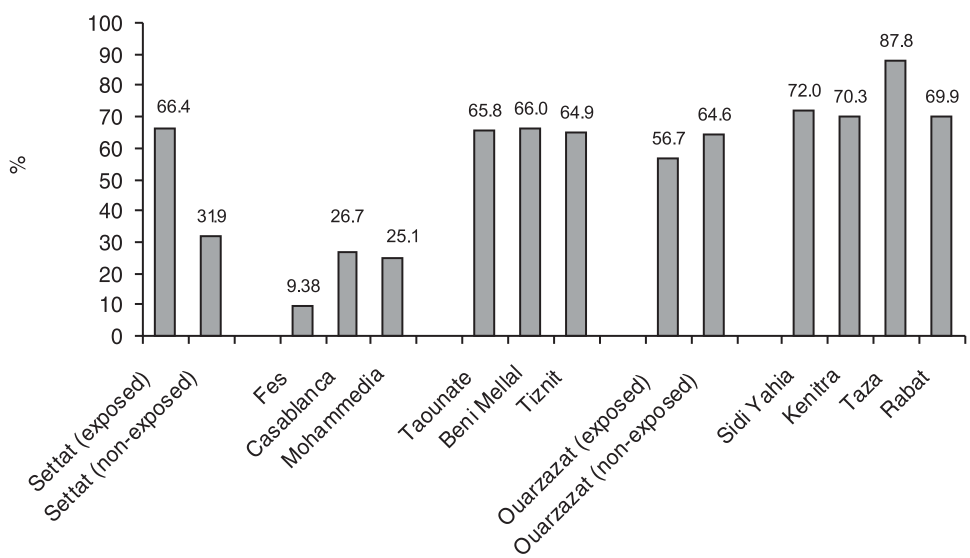

Figure 1 **Prevalence of intestinal parasitosis in the present study in Settat compared with studies in other areas of Morocco; sources: [*9–15,17*]**

were found in other areas of Morocco among children of preparatory school age (6–8 years) [*12,14,15*] or primary school age (6–15 years) [*10–12,22*]. Thus, in our study, as in the national survey of rural areas, age did not have a clear effect on the prevalence of intestinal parasites [*9*], whereas the study in Ouarzazate noted that age was the best predictive factor for intestinal parasites [*17*]. The risk of parasitic infestation was higher among older participants [*17*].

In addition, sex did not influence the prevalence of the intestinal parasitosis. Some studies have noted a higher prevalence among females [*9,10,19,21*] and others have observed a small increase in the prevalence among male subjects [*9,13–15*] or an equality between the sexes [*12*]. But no study has observed a significant difference.

Household crowding is an important factor in parasitosis. A national survey showed that the prevalence of intestinal parasites was higher in rural environments of the studied provinces in houses with more than 5 persons [*9*]. Several studies on childhood behaviour noted the effect of crowding on the rate of parasite infestation [*10,12,14,15*]. Several studies, as well as ours, reported a higher frequency of intestinal parasitosis in adults and children living in residences without toilets [*9,10,12,14,15*].

Ent. coli, *End. nana* and *Pseu. butchlii* were the 3 amoebae found within the stool samples of the study groups. These are frequently found in the climate of Morocco. They have no or limited pathogenic power. Their presence in the stool, however, is proof that the carrier consumed food contaminated by faeces [*23*].

The prevalence of *G. intestinalis*, a flagellate pathogen of the digestive tract of man, was 11.7% in the exposed group compared with 2.5% in the non-exposed group. This parasite is particularly widespread in hot areas and is related to faecal contamination. The cysts, which remain alive in the outside environment for a long time, ensure interhuman contamination [*24*]. *Chilomastix mesnili*, another flagellate intestinal parasite, is rarely or never pathogenic [*23*]. Its prevalence was similar in the exposed group and the non-exposed group.

Polyparasitism was observed in 17.3% of cases among the exposed group and 2.5% in non-exposed groups ($P < 0.001$). The Ouarzazate investigation did not show a significant difference concerning polyparasitism between the people who used purified wastewater (18.3%) and the reference group (13.1%) ($P = 0.31$) [*16*]. Polyparasitism was also observed in studies of children at school with prevalence between 28.6% and 79.0% [*10,12,14,15*]. Whereas in urban studies devoted to a broader population [*13,19,21*], the rate of polyparasitism varied only between 1.5% and 9.1% [*13,19*]. At the rural level, the national survey of 1994 showed different rates of multiparasitism, 25.1%, 49.7% and 58.3% in Beni Mellal, in Tanouante and in Tiznit respectively [*9*].

Although our study found around 2 out of 3 people were infested with parasites in the exposed group compared with only 1 person out of 3 in the non-exposed group, a direct effect of wastewater on intestinal parasite transmission could not be established because our study did not include a parasitological examination of the wastewater. Therefore we were unable to exclude other sources of contamination in the study population. However, the study provides some evidence to implicate wastewater in the transmission of parasites, either by direct contact or via irrigated food crops, which merits further investigation.

Conclusion

In Settat, the prevalence of intestinal parasitosis was significantly higher among people using untreated wastewater in agriculture. The relative risk was 2.1. The use of untreated wastewater in agriculture as a source of intestinal parasite contamination could be responsible for a degradation of the health of this population. Preventive measures have been taken. These consist of sensitizing the population to aspects of faecal hygiene, nutrition of children and the health impacts of wastewater.

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