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

S. El Kettani,1 E. Azzouzi,2 K. Boukachabine,3 M. El Yamani,4 A. Maata5 and M. Rajaoui6

1Medical Intern Unit; 5Biomedical Analysis Laboratory, Hospital Hassan II, Settat, Morocco (Correspondence to S. El Kettani: said_elkettani@yahoo.fr).
2Biomedical Analysis Laboratory, Azzouzi, Settat, Morocco.
3Parasitology Laboratory, Faculté des Sciences et Techniques, Settat, Morocco.
4Virology Laboratory, Institut National de la Recherche Agronomique, Settat, Morocco.
6Parasitology Laboratory, Institut National d’Hygiène, Rabat, Morocco.

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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 (étalée 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.

1Medical Intern Unit; 2Biomedical Analysis Laboratory, Hospital Hassan II, Settat, Morocco (Correspondence to S. El Kettani: said_elkettani@yahoo.fr).
3Parasitology Laboratory, Faculté des Sciences et Techniques, Settat, Morocco.
4Virology Laboratory, Institut National de la Recherche Agronomique, Settat, Morocco.
5Biomedical Analysis Laboratory, Azzouzi, Settat, Morocco.
**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 com-
pare the prevalence obtained according to
various characteristics. An association was
considered significant when the P-value
was < 0.05. For the evaluation of the as-
sociation between possible risk factors and
parasitosis, the ratio of prevalence or rela-
tive 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%
males. 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 preva-
ience of polyparasitism was 17.3%. Among
the non-exposed group, this prevalence was
only 2.5%. This difference was also statisti-
cally 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 signifi-
cant difference in the prevalence of intes-
tinal 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 differ-
ence 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 re-
searchers cast some doubt on the reliability
of the responses.

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

Analysis 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
**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>
<thead>
<tr>
<th>Variable</th>
<th>Exposed No.</th>
<th>Non-exposed No.</th>
<th>$\chi^2$-test</th>
<th>P-value</th>
</tr>
</thead>
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<tr>
<td><strong>Sex</strong></td>
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</tr>
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<td>Female</td>
<td>113</td>
<td>60</td>
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<td>0.67</td>
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<tr>
<td>Male</td>
<td>101</td>
<td>59</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Age group (years)</strong></td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3–9</td>
<td>30</td>
<td>15</td>
<td>4.30</td>
<td>0.36</td>
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<tr>
<td>10–14</td>
<td>31</td>
<td>11</td>
<td></td>
<td></td>
</tr>
<tr>
<td>15–29</td>
<td>68</td>
<td>33</td>
<td></td>
<td></td>
</tr>
<tr>
<td>30–59</td>
<td>67</td>
<td>46</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60+ years</td>
<td>18</td>
<td>14</td>
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</tr>
<tr>
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<td></td>
</tr>
<tr>
<td>Educated</td>
<td>109</td>
<td>69</td>
<td>1.52</td>
<td>0.92</td>
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<tr>
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<td>50</td>
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<td><strong>Crowding index</strong></td>
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<td></td>
<td></td>
</tr>
<tr>
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<td>20</td>
<td>4.53</td>
<td>0.10</td>
</tr>
<tr>
<td>$2–2.9$</td>
<td>92</td>
<td>59</td>
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</tr>
<tr>
<td>$\geq 3$</td>
<td>64</td>
<td>40</td>
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<tr>
<td><strong>Toilet in house</strong></td>
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</tr>
<tr>
<td>Yes</td>
<td>171</td>
<td>103</td>
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<tr>
<td>No</td>
<td>43</td>
<td>16</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>214</td>
<td>119</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*aNo. of people in household/no. of rooms.*
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 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

<table>
<thead>
<tr>
<th>Variable</th>
<th>Prevalence of intestinal parasitosis</th>
<th>Relative risk of RR</th>
<th>95% CI</th>
<th>Statistics</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Exposed (n = 214)</td>
<td>Non-exposed (n = 119)</td>
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</tr>
<tr>
<td></td>
<td>+ve</td>
<td>−ve</td>
<td>%</td>
<td>+ve</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>76</td>
<td>37</td>
<td>67.3</td>
<td>18</td>
</tr>
<tr>
<td>Male</td>
<td>66</td>
<td>35</td>
<td>65.3</td>
<td>20</td>
</tr>
<tr>
<td>Statistics</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age group (years)</td>
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<td></td>
</tr>
<tr>
<td>3–9</td>
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<td>11</td>
<td>63.3</td>
<td>4</td>
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<td>20</td>
<td>11</td>
<td>64.5</td>
<td>2</td>
</tr>
<tr>
<td>15–29</td>
<td>46</td>
<td>22</td>
<td>67.6</td>
<td>16</td>
</tr>
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<td>30–59</td>
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<td>23</td>
<td>65.7</td>
<td>15</td>
</tr>
<tr>
<td>60+</td>
<td>13</td>
<td>5</td>
<td>72.2</td>
<td>1</td>
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</tr>
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<td>Education</td>
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<td></td>
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<td>66</td>
<td>43</td>
<td>60.6</td>
<td>22</td>
</tr>
<tr>
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<td>29</td>
<td>72.4</td>
<td>16</td>
</tr>
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<tr>
<td>Crowding indexa</td>
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<td></td>
</tr>
<tr>
<td>1–1.9</td>
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<td>25</td>
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<tr>
<td>2–2.9</td>
<td>66</td>
<td>26</td>
<td>71.7</td>
<td>19</td>
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<td>≥ 3</td>
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<td>21</td>
<td>67.2</td>
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<tr>
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<td>No</td>
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<td>14</td>
<td>67.8</td>
<td>5</td>
</tr>
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<td>Statistics</td>
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</tr>
<tr>
<td>Source of water</td>
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<td>(well)</td>
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<tr>
<td>Collective</td>
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<td>57</td>
<td>67.2</td>
<td>–</td>
</tr>
<tr>
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<td>15</td>
<td>62.5</td>
<td>–</td>
</tr>
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</tr>
<tr>
<td>Total</td>
<td>142</td>
<td>72</td>
<td>66.4</td>
<td>38</td>
</tr>
</tbody>
</table>

*aNo. of people in household/no. of rooms.
*bEvidence of at least 1 type of intestinal parasite in faeces.
*n = total number of subjects.
*CI = confidence interval.
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%

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

<table>
<thead>
<tr>
<th>Intestinal parasite</th>
<th>Prevalence of intestinal parasitosisa</th>
<th>Exposed (n = 214)</th>
<th>Non-exposed (n = 119)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rhizopods (amoebae)</td>
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<td></td>
</tr>
<tr>
<td>Entamoeba coli</td>
<td>52.3</td>
<td>26.1</td>
<td></td>
</tr>
<tr>
<td>Endolimax nana</td>
<td>12.1</td>
<td>0.0</td>
<td></td>
</tr>
<tr>
<td>Pseudolimax butschlii</td>
<td>6.5</td>
<td>3.4</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>66.4</td>
<td>29.4</td>
<td></td>
</tr>
<tr>
<td>Flagellates</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Giardia intestinalis</td>
<td>11.7</td>
<td>2.5</td>
<td></td>
</tr>
<tr>
<td>Chilomastix mesnili</td>
<td>1.9</td>
<td>2.5</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>13.6</td>
<td>5.1</td>
<td></td>
</tr>
<tr>
<td>Helminths (nematodes)</td>
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<td></td>
</tr>
<tr>
<td>Ascaris lumbricoides</td>
<td>4.2</td>
<td>0.0</td>
<td></td>
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<tr>
<td>Enterobius vermicularis</td>
<td>0.5</td>
<td>0.0</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>4.7</td>
<td>0.0</td>
<td></td>
</tr>
</tbody>
</table>

aEvidence of at least 1 type of intestinal parasite in faeces.

n = total number of subjects.

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 inter-human 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.

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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