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Prevalence of Intestinal Parasites in the Rural Regions of Kouhdasht, Lorestan Province, Iran, 2008

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Abstract: The major aim of this study was determine of prevalence of intestinal parasites in the rural regions of Kouhdasht, Lorestan Province of Iran. Parasites are living creatures which have to be dependent on other living creatures including plants and animals as their hosts to survive. Those parasites which choose human beings as their hosts are divided into two groups of protozoan and multi-cellular parasites in terms of their appearance. Additionally, they are able to live in various tissues of human beings in terms of the parasite type. However, those which live in the digestive system are called intestinal parasites and have infected a large number of people all over the world. Four hundred and sixty two stool samples were collected from the rural families serviced by the health centers of Kouhdasht using multi-step clustered methods and were examined using direct methods including physiologic serum and Lugol, as well as formalin-ether concentrated method to diagnose all intestinal parasites. In addition, all the stool samples were examined using agar culture method and Harada-Mori to diagnose *Strongyloides stercoralis*. Out of the 462 stool samples collected by concentrated method, 150 ones (32.5%) were infected with intestinal parasites while no positive cases were reported for *Strongyloides stercoralis* larva despite administering various diagnostic methods. The results showed that 32.5%, or one-third, of the studied population were found to be infected with intestinal parasites while the tests were administered only once. Definitely, if the tests and sample collecting methods had been repeated three times, the prevalence rate would have been more than this relatively high one. Therefore, providing actions and facilities on the part of responsible officials and organizations are required to enhance health facilities and to eradicate these troublesome creatures. In addition, in this study significant relationships were found between the prevalence rate of the parasites and the population density as well as regular soap using.

Key words: Intestinal parasites, prevalence, Kouhdasht, Lorestan

INTRODUCTION

Parasites are among the factors of human infections which are divided into two groups of worms and protozoan parasites and can survive in most of human tissues and can cause

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specific diseases and complications in terms of the tissues. A group of these creatures which live in the digestive system are called intestinal parasites and have almost high prevalence and it is estimated that they have infected more than 3 billion people throughout the world (Balcioglu *et al.*, 2007) and in the developed countries the prevalence of the protozoan parasites is higher in comparison to worms (Haque, 1997). Intestinal parasites include nematodes such as *Strongyloides stercoralis* which has infected from 30 to 100 million people (Lim *et al.*, 2004) as well as *Enterobius vermicularis* which has infected millions of people all over the world (Kang *et al.*, 2006). Nevertheless, the prevalence rates differ in various parts of the world and depend on some factors including climate, development, health culture and so on. Moreover, the sensitivity and exactness of diagnostic techniques do not have to be ignored.

In a study conducted in the rural regions of west China, 51.7% of the people were found to be infected with intestinal parasites so that 36.5, 12.7, 3 and 0.08% had been infected with one, two, three and four parasites, respectively (Tang and Lui, 2003).

In another study carried out in Saudi Arabia on schoolchildren in the 6-18 age range, 2233 (22.6%) out of 9881 stool samples analyzed by a simple direct method were positive most of which (13.5%) belonged to *Giardia lamblia* (Abdulaziz *et al.*, 1993). The results of a study performed on people over 2 in 12495 families controlled by the health centers of Iran universities of medical sciences using concentrated formalin-ether method revealed that 19.3% of the people suffered from infections with intestinal parasites and out of this number, 19.7 and 19.1% were males and females, respectively and the most prevalent parasites were *Giardia lamblia* with 10.9%, *Ascaris lumbricoides* 1.5%, *Entamoeba histolytica* 1% and *Enterobius* 0.5% (Sayyari *et al.*, 2005). In a study conducted on patients with gastrointestinal complaints in the health centers of Zahedan from 2004 to 2006, 27.3% of them were infected with more than one intestinal protozoan parasite (Hassing *et al.*, 2009).

MATERIALS AND METHODS

This study was conducted from August 1 to December 1, 2008, in order to investigate the prevalence of intestinal parasites in the rural families controlled by the health centers of Kouhdasht in Lorestan province, West Iran. The 462 people were selected using a multi-step clustered sampling method. Then, following the arrangements, some trained health providers filled out questionnaires and distributed single-use coded containers among the families simultaneously and immediately sent the collected samples and the questionnaires to the central library.

Since, the purpose of the present study was to investigate the prevalence rate of all intestinal parasites-accompanied by the study of *Oxiour* prevalence in students using a tape method, the diagnostic methods had to be compatible with the objectives of the study. Consequently, the following five diagnostic methods were applied:

Simple or Direct Method

It uses an isotonic physiologic serum solution (8.5 g of salt in 1000 mL of distilled water) to observe the movement of parasites (Garcia, 2007).

Direct Method With Lugol

It is a combination of iodine and potassium iodide in distilled water that is prepared in a stored way and during the operation. This solution is used to stain parasites quickly particularly protozoan cysts in order to increase their separation from the background and

consequently, to result in faster and more exact diagnosis. Methods 1 and 2 are also called wet-mount methods so that approximately 2 mg of stool is dissolved in one drop of each and examined for 5 to 10 min under microscope with 10× and 40× lenses. Although, these methods are quick and inexpensive, they lack high diagnostic quality and are typically used in moderate to high infections (Garcia, 2007).

Concentrated Formalin-Ether Method

In this method-as the name suggests the combinations-more parasites are centralized since it uses a larger amount of stool sample and eliminates more redundant materials via passing its suspension from two layers of gauze and centrifuges its sediment. In other words, more parasites are concentrated in less volume and when they are fixed in Formalin, their potential to create infection will be decreased or removed so that they can be handled with more confidence. Additionally, ether can create more clarity by dissolving fats around the parasites. This costly and time-consuming method will result in higher diagnostic quality and is an appropriate method for slight infections. The results obtained from these three methods were presented as follows: (1) Qualitative; negative or if positive the names of the parasites were recorded. (2) Quantitative; in positive cases the average number of the parasites for each sample and each method was counted in ten microscopic fields and written as a number in front of the name of the available method in a questionnaire specially prepared for this purpose.

Agar Culture Environment Method

This method with 96% sensitivity is used to diagnose the mobile larva of *Strongyloides stercoralis* which is one of the larva-producing nematodes. In this method, basically, bacteria along with the mobile larva separated from the stool sample are frozen in the agar culture environment setting and then they grow and appear as lines in the setting. However, after preparing the plate of the culture setting using a setting frozen, clear, rich and fertile for all bacteria called Muller-Hinton, a few grams of fresh stool sample was placed in the center of the plate. Then the lid of the plate was closed using a tape and the researchers attempted daily to find the larva or its effects for a week (Garcia, 2007).

Harada-Mori Method

This complementary method to diagnose *Strongyloides stercoralis* is carried out in two ways and in this study the on-slide method was utilized in which about 1 g of fresh stool sample was placed in the middle of cellophane paper tape and in the width of the slide in the plates containing sterilized distilled water obliquely so that the water could reach the sample due to the absorption ability of the study. When larvae were found in the water, a sampler was used to search for the larva microscopically and eventually all the water was centrifuged and the sediment was searched for the larva (Garcia, 2007). To analyze the data obtained from the experiments, the SPSS 15.0 version software was utilized. In order to check for statistical differences, Chi-square test and Fisher's exact test were adopted (Agresti, 2007). The difference between the two groups was considered to be significant when the p-value was lower than 0.05.

RESULTS

Out of the 462 stool samples examined, 150 cases (32.5%) were diagnosed as being infected with intestinal parasites using concentrated formalin-ether method out of whom 143

(95%) and 7(5%) were infected with protozoan and worm parasites respectively, while 13 cases (2.8%) with more than one parasite. However, the diagnosed infected cases with intestinal parasites using Lugol and direct physiologic serum were 118 (25.5%) and 107 (23.2%) cases respectively, but *Strongyloides stercoralis* larva was not found by applied methods including direct Lugol or physiologic serum method, concentrated and agar and Harada-Mori culture methods.

Entamoeba coli, which is one of the nonpathogenic protozoa, was in the first rank with 79 positive cases (17.1%) and *Giardia lamblia*, a pathogenic protozoan, with 41 cases (8.9%) was in the second rank, while *Entamoeba histolytica* was reported in only 5 cases (1.1%). Among the worms, the ovum of *Hymenolepis nana* was found in 6 cases (1.3%) and one case was reported for *Oxiour* ovum while the rest of the cases were nonpathogenic protozoa and was less prevalent (Table 1). The prevalence of the studied parasites in all the studied health centers was not constant and a statistically significant difference ($p < 0.001$) was found between them. For instance, the infection rates in Siahcheshmeh and Barekalek centers were 49.1 and 41.2%, respectively while they were found to be 13.8 and 16.9% in Barekheireh and Parian health centers, respectively (Table 2). In addition, there was a significant relationship between the prevalence rate and number of family members ($p < 0.001$) so that the infection rate increased with increasing family members and 9 or more member families with 53.3% had the highest and, in contrary, 2-member families had the lowest infection rate (Table 3).

Although, in this study there was a significant relationship between using soap and amount of infection ($p = 0.04$), the infection was higher in the people who used soap occasionally in comparison to those who always or never used it (Table 4).

Table 1: Frequency and relative frequency of intestinal parasites in the rural areas of Kouhdasht, Lorestan province, Iran

Parasite (s)	Frequency	(%)
<i>Entamoeba coli</i> cyst	79	17.1
<i>Giardia</i> cyst	41	8.9
<i>Giardia lamblia</i> cyst and <i>Entamoeba coli</i> cyst	10	2.2
<i>Hymenolepis nana</i> ovum	6	1.3
<i>Entamoeba histolytica</i> cyst	5	1.1
<i>Iodamoeba butschlii</i> cyst	3	0.6
<i>Giardia</i> cyst and <i>Hymenolepis nana</i> ovum	2	0.4
<i>Giardia</i> cyst and <i>Oxiour</i> ovum	1	0.2
<i>Endolimax nana</i> trophozoite	1	0.2
<i>Endolimax nana</i> cyst	1	0.2
<i>Kilomastix mesinily</i> cyst	1	0.2
Total	150	32.5

Table 2: Frequency and relative frequency of intestinal parasites in terms of the studied health centres

Health centre	All the studied population	Positive cases	% of positive cases to all cases
Barekheireh	29	4	13.8
Parian	83	14	16.9
Komeir	89	27	30.3
Shiravand Gandabeh	95	33	34.7
Siahcheshmeh-Bareanar	26	10	38.5
Barekalek	85	35	41.2
Siahcheshmeh	55	27	49.1
Total	462	150	32.5

Table 3: Prevalence of intestinal parasites in terms of number of family members

No. of family members	Negative (%)	Positive (%)	Total (%)
1-2	12 (85.7)	2 (14.3)	14 (100)
3-4	87 (79.8)	22 (20.2)	109 (100)
5-6	101 (69.7)	44 (30.3)	145 (100)
7-8	89 (62.2)	54 (37.8)	143 (100)
≥9	21 (46.7)	24 (53.3)	45 (100)
Total	310 (68)	146 (32)	456 (100)

Table 4: Relationship between the way of using soap and infected people

Way of using soap	Frequency and percentage of infected people	Frequency and percentage of uninfected people	Total
Always	61 (28.1)	156 (71.9)	217
Occasionally	72 (39.3)	111 (60.7)	183
Never	17 (27.9)	44 (72.1)	61
Total	150 (32.5)	311 (67.5)	461 (100)

In the present study, although no relationships were found between the infected people and quality of washing vegetables with water, detergents and detergents and sterilizing chemicals, the percentages of infection were 36.5, 30.5 and 35%, respectively showing the positive effect of quality of washing on infection decrease. Moreover, no relationships were found between the infected people and digestive signs including stomachache, flatulence, losing appetite and so on. It is noteworthy to mention that although classifying people with regard to their economic situations is difficult, the families in the study were divided into three groups of weak-earning less than 200 thousand tomans (the currency of Iran), good-more than 600 thousand tomans and moderate-between 200 and 600 thousand tomans. The infection rates in the weak and moderate groups were 36.5 and 30.5%, respectively while the rate was 0% in the economically good group, but no difference was found concerning gender ($p = 0.3$).

DISCUSSION

This study aimed to investigate the prevalence of intestinal parasites; therefore, all feasible diagnostic methods were used to achieve the goals. In a concurrent study carried out on elementary schoolchildren of the city, in a step the infection to *Oxiour* was found to be 32.8% using the tape method, but despite using direct methods including physiologic serum and Lugol, formalin-ether concentrated method, agar culture method with sensitivity more than 96% (Sato *et al.*, 1995) and Haradi-Mori method, no *Strongyloides stercoralis* larvae were found. On the other hand, the fact that no cases of this parasite larva were reported in Lorestan province in the last five years can confirm the results of the present study and the reasons can be attributed to using roofed lavatories due to cultural reasons, regular shoe using and also because the parasite is not indigenous to the province. Moreover, in a study in Iran only 2 out of 781 HIV positive patients were infected with the parasite larvae despite the fact that HIV positive patients are vulnerable and can be the host of the parasite due to the immunodeficiency of their bodies and the infection power of the parasite (Meamar *et al.*, 2007; Marchi and Cantos, 2003).

In this study a multi-step clustered sample collecting method was used and out of 462 people who were randomly chosen using the list of families controlled by the health centers of Kouhdasht in Loresatn province, west Iran, 150 cases (32.5%), or one-third of the cases, were infected with the parasite. Although, the infection rates varied in different health centers from 13.8 to 49.1% and significant differences were found among the results, on the whole, it is a high infection rate since in a study done on schoolchildren's stool samples in Thailand using the formalin-ether method, 242 (12.6%) cases out of 1920 samples were infected with intestinal parasites (Warunee *et al.*, 2007). Also, in another study in Tehran 10.7% of the people were infected with the parasite (Shojaei *et al.*, 2008) and the protozoan parasites had infected 25% of the population in Mazandaran province in Iran (Kia *et al.*, 2008). However, the infection rate reported in the present study is lower in comparison with the results from special population groups; for instance, a study on food vendors and catering staff in Shiraz showing the infection of 59.4% with the parasites using concentrated formalin-ether method (Neghab *et al.*, 2006).

Table 4 shows a significant relationship between the infection and soap using way ($p = 0.04$) but in this study, contrary to what is common, the infection rate in those who used soap occasionally was higher than those who used it regularly or those who never used it and it may be attributed to the fact that not using soap in societies where using soap is considered as important arouses sensitivity while a study in Uzbekistan showed that re-infection in those for whom the incentives had been effective was 30% less those who had used medicine (Gungoren *et al.*, 2007).

Although, no relationship was observed between the infection and the quality of washing vegetables, the infection rates for washing vegetables with water alone that shows indifference to health issues, washing them with detergents and washing them with detergents and sterilizing chemicals that shows suitable hygienic behavior were 36.5, 30.5 and 25%, respectively, indicating the positive effect of washing quality on infection rate. It is noteworthy to explain that consuming raw vegetables has a vital role in parasitic infections so that in a study in Ardabil 71% of the garden vegetables and 50% of the store vegetables were infected (Daryani *et al.*, 2008).

Although, the researchers were satisfied with the planning, procedure and results of the present study, there were some disturbances due to particular customs and traditions concerning sample collecting and also breaking promises on the part of other researchers which led to ignorance of the society about the results of previous studies. It is hoped that the results of the present study will be beneficial to responsible health officials as well as infected people needing relevant information.

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REFERENCES

- Abdulaziz, M., L. Al-Sekait, A. Al-Ansary and F. Al-Zamel, 1993. Prevalence of pathogenic intestinal parasite among Saudi rural schoolchildren. *Eur. J. Public Health*, 3: 232-236.
- Agresti, A., 2007. *An Introduction to Categorical Data Analysis*. Wiley-Interscience, USA.
- Balcioglu, C., Ö. Kurt, M.E. Limoncu, G. Dinç and M. Gumuş, 2007. Rural life, lower socioeconomic status parasitic infections. *Parasitol. Int.*, 56: 129-133.
- Daryani, A., G.H. Eftehad, M. Sharif, L. Ghorbani and H. Ziaei, 2008. Prevalence of intestinal parasites in vegetables consumed in Ardabil, Iran. *Food Control*, 19: 790-794.
- Garcia, L.S., 2007. *Diagnostic Medical Parasitology*. 5th Edn., ASM Press, Washington DC. USA.
- Gungoren, B., R. Latipov, G. Regallet and E. Musabaev, 2007. Effect of hygiene promotion on the risk of reinfection rate of intestinal parasites in children in rural uzbekistan. *Trans. R. Soc. Trop. Hyg.*, 101: 564-569.
- Haque, R., 2007. Human intestinal parasites. *J. Health Population Nutr.*, 25: 387-391.
- Hassing, R.J., P.J. Wismans, R. Koelewijn, J.J. van Hellemond and P.J. van Genderen, 2009. Comment on: Frequency of enteric protozoan parasites among patients with gastrointestinal complaints in medical centers of Zahedan, Iran. *Trans. R. Soc. Trop. Med. Hyg.*, (In Press). 10.1016/j.trstmh.2009.02.008
- Kang, S., H.K. Jeon, K.S. Eom and P. Joong-Ki, 2006. Egg positive rate of enterobius vermicularis among preschool children cheongju korea. *Korean J. Parasitol.*, 44: 247-249.

- Kia, E.B., M. Hosseini, M.R. Nilforoushan, A.R. Meamar and M. Rezaeian, 2008. Study of intestinal protozoa parasites in rural in habitant of Mazandaran Province. *Iran. J. Parasitol.*, 3: 21-25.
- Lim, S., K. Katz, S. Krajden, M. Fuksa, J.S. Keystone and K.C. Kain, 2004. Complicated fatal *Strongyloides* infection in Canadians: Risk factors, diagnosis management. *CMAJ*, 171: 5-5.
- Marchi, B.J. and G.A. Cantos, 2003. Evaluation of techniques for the diagnosis of *Strongyloides stercoralis* in human immunodeficiency virus (HIV) positive and HIV negative individuals in the city of Itaja, Brazil. *J. Infect. Dis.*, 7: 402-408.
- Meamar, A.R., M. Rezaian, M. Mohraz, R. Hadighi and E.B. Kia, 2007. *Strongyloides stercoralis* hyper-infection syndrome in HIV+/AIDS patients in Iran. *Parasitol. Res.*, 101: 663-665.
- Neghab, M., S. Moosavi and M.D. Moemenbellah-Fard, 2006. Prevalence of intestinal parasitic infections among catering staff of students canteens at Shiraz, Southern Iran. *Pak. J. Biol. Sci.*, 9: 2699-2703.
- Sato, Y., J. Kobayashi, H. Toma and Y. Shiroma, 1995. Efficacy of stool examination for detection of *strongyloides* infection. *Am. J. Trop. Med. Hyg.*, 53: 248-250.
- Sayyari, A.A., F. Imanzadeh, S.A. Bagheri Yazdi, H. Karami and M. Yaghoobi, 2005. Prevalence of intestinal parasitic infections in the islamic republic of Iran. *East Mediterr Health J.*, 11: 377-383.
- Shojaei, A., R. Alaghehban and L. Akhlaghi, 2008. Prevalence of intestinal parasites in a population in South of Tehran, Iran. *Rev. Inst. Med. Trop.*, 50: 145-149.
- Tang, N. and N.J. Lui, 2003. A cross-sectional study of intestinal parasitic infections in a rural district of West China. *Can. J. Infect. Dis.*, 14: 159-162.
- Warunee, N., L. Choomanee, P. Sataporn, Y. Rapeeporn and W. Nuttapon, 2007. Intestinal parasitic infections among school children in Thailand. *Trop. Biomed.*, 24: 83-88.