ORIGINAL ARTICLE



# Spatial distribution of *Giardia lamblia* infection among general population in Mazandaran Province, north of Iran

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Abstract *Giardia lamblia* is the most prevalent intestinal parasites of humans in Iran and other in the world although information on geographical distribution of giardiasis plays significant role in identifying communities at high risk, little attention has been paid to study human giardiasis using geographical information system. Therefore, the aim of the current study was to determine temporal and spatial patterns of human giardiasis distribution to identify possible high risk areas and seasons in northern Iran. A total of 4788 people referred to health centers in the Mazandaran Province of northern Iran were surveyed January to December 2015. From each person stool sample and questionnaire with socio-demographic data were collected. *Giardia* infection was diagnosed using direct wet mount, formalin ether concentration and trichrome staining. The

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results were analyzed using Moran Local Indicators of spatial association and geographically weighted regression. The overall prevalence of *Giardia* infection was 4.6% (222/4788), and was significantly higher among those aged 5–9 years compared to their older peers (P < 0.0001). Our data showed a significant dependency between the prevalence of *G. lamblia* and age, job, residence, season and height from the sea (P < 0.0001). The results of this study provided a precise and specific spatial and temporal pattern of human giardiasis distribution in the Mazandaran Province, Iran. These evidences should be considered for proper control of disease decisions and strategies.

Keywords  $Giardiasis \cdot Geographical information system \cdot Infectious disease \cdot Iran$ 

# Introduction

Giardia lamblia, is a cosmopolitan protozoan parasite of mammals which causes illness of the host intestinal tract (Sedighi et al. 2015). Giardiasis has variable clinical symptoms ranging from asymptomatic to acute or chronic diarrhea, dehydration, abdominal cramps, nausea, vomiting, weight loss, disaccharide intolerance and malabsorption. In addition, G. lamblia affects growth and cognitive functions of infected children (Jethwa et al. 2015). This parasite transmitted by its cyst form through contaminated food or water (Fecal-Oral). The encysted stage of the organism is relatively resistant to chlorination and ozonolysis can remain viable for several weeks. Humans are considered as the most significant reservoir hosts for human giardiasis (Adam 2001; Al-Mekhlafi et al. 2013), with outbreaks occasionally occurring due to mass exposure to insanitry water and food containing cysts of the parasite and direct contact with infected individuals, particularly in child care centers (Gholami et al. 2016). *Giardiasis is* of particular concern in developing countries with a prevalence ranging from 20 to 60% compared to 2–7% in industrialized countries (Faraji et al. 2015; Yakoob et al. 2005). In Iran, the prevalence of the disease was reported 14.7% (Fakhar et al. 2014). *Giardiasis* can be acquired at any age, it is most prevalent in children living in unhygienic conditions and areas of high population density (Jethwa et al. 2015). Estimated about 200 million people have symptomatic giardiasis across Asia, Africa, and Latin America with 500,000 new reported cases annually, despite current monitoring programs (Al-Mekhlafi et al. 2013; Rayani et al. 2014; Abasian et al. 2013).

A recent study in Tonekabon, west of Mazandaran Province, has shown that *Giardia* protozoa with prevalence 3% was one of the most important pathogens in the protozoan group (Shahdoust et al. 2016).

Due to the notably high degree of infection rates presented above, the collection of a comprehensive dataset and its analyses are necessary to better manage prevention and control strategies for giardiasis. One of these strategies uses geographical information system (GIS) which can prepare geographical patterns in disease distribution and helps develop early warning systems for infectious diseases (Zhou et al. 2009). Currently, GIS has been well documented as an important tool to collect, retrieve, organize, and display public health information (Croner et al. 1996). In addition, it has become a commonly applied platform to visualize and analyze diseases. This powerful tool can run simulative and predictive models required for complex dynamic processes of transmission concerning infectious diseases (Thomas et al. 2002). Therefore, the aim of the current study was to study temporal and spatial patterns of human giardiasis distribution and find possible risk factors in the Mazandaran Province, northern Iran.

## Materials and methods

#### Study area

This study was done in Mazandaran Province, located in south of the Caspian Sea, north of Iran (lat.  $35^{\circ}47''36^{\circ}35''$ , long.  $50^{\circ}34''54^{\circ}10''$ ). This province has 19 cities and populace of around 3,073,943 people. The mean yearly relative humidity is 80%, with rainfall happening in all seasons of the year, and the mean temperature is 18 °C.

### Samples collection

This cross-sectional study was performed on 4788 people referring to health centers of Mazandaran Province from January to December 2015. Stool specimens were collected after receiving consent from the individuals and filling out the questionnaire. The specimens were preserved in polyvinyl alcohol (PVA) and carried to the Parasitology Laboratory of Mazandaran University of Medical Sciences.

### **Stool examination**

All specimens were examined using normal saline (0.85% NaCl) for observed of trophozoites and Lugol's iodine staining for detect of the *Giardia* cysts under the light microscope by  $40 \times$  objective magnifications. Then, Formalin-ether concentration technique and trichrome staining were done for all samples (Fig. 1).

#### **Geographical data**

In our study, data related to height (three groups: less than 500, 500–1000 m and above 1000 m) and distance from the sea (three groups: less than 10,000, 10,000–20,000 m and more than 20,000 m) were obtained from Google Earth software (version 16). Ecological data (such as temperature, rain, humidity, height and distance from the sea) were collected from the Mazandaran Metrological Organization.

#### Statistical analysis procedure

The obtained results were analyzed by the SPSS 16.0 and using Moran Local Indicators of Spatial Association to determine the distribution of human giardiasis in different areas of the province. Furthermore, geographically weighted regression (GWR) was used to study the geographical relationship of prevalence of human giardiasis and associated important variables including precipitation and temperature.



Fig. 1 Cyst from of Giardia stained with trichrome  $(\times 100)$ 

#### Results

The overall prevalence of giardiasis was 4.6% (222/4788). 53.9% (2579) were male and 46.1% (2209) female. The mean of ages was  $32.39 \pm 17.75$  years (range 1–77 years old).

A significant association was found between the prevalence of *G. lamblia* with age, season, job, residence and education (P < 0.05) (Table 1).

Our results showed that the prevalence rates of *G. lamblia* were as follow: < 500 m, 3.5% (61/1710); 500–1000 m, 5.1% (152/2954); and > 1000 m, 7.2% (9/124). Among metrological risk factors, only height 500–1000 m prevalence of *G. lamblia* showed a significant difference according to the height from the sea (P < 0.05). According to mapping prevalence of *G. lamblia* indicated that Amol (9.3%) and Galugah districts (1.8%) had the highest and lowest prevalence rate in Mazandaran Province (Fig. 2).

### Discussion

*Giardia lamblia* is a major human pathogen with a worldwide distribution. Prevalence of infection is higher in countries that lack proper sanitation and hygienic conditions (Hakim et al. 2011). Although *Giardia* was added to the 'WHO Neglected Diseases Initiative' in 2004, it continues to be a common and problematic human parasite (Savioli et al. 2006).

In the present study, the prevalence of *G. lamblia* infection among the general population in the Mazandaran Province of Iran was 4.6%. The prevalence of *G. lamblia* is high in some countries such as Amman, Jordan (78%), Western Nepal (73.4%), Malaysia (44.1%), Latin America (24%) (Shakkoury and Wandy 2005; Easow et al. 2005; Noor Azian et al. 2007; Reinthaler et al. 1998). Different studies in Iran have shown infection rates of 1.4–39.5%. It appears that changes in the prevalence of *G. lamblia* infection should be due to different factors including seasons of sampling, kind of consumed water, different laboratory tests, climate and environmental factors.

In this study, the most prevalence rate of *G. lamblia* was found among individuals aged 5–9 years (9.3%), and this is in agreement with previous studies in India, Malaysia and Iran (Jethwa et al. 2015; Choy et al. 2014; Arani et al. 2008). This could be attributed to the higher exposure of young children to a lot of infection sources, which could be due to having lower standards of personal hygiene when compared to old children and adults. In this study prevalence of giardiasis in males were 4.7% and in females 4.5%. The results were similar to study of Al-Saeed 2006 in Iraq (Al Saeed and Issa 2006). Another study carried out in Palestine and Mexico reported a higher rate (24%) of giardiasis among males than in females (Al-Zain and Al-Hindi 2005; Quihui et al. 2006). This high rate of prevalence in male that may be they are more exposure to sources of parasites or exposed to unhygienic conditions in fields during outdoor activity while females may be remain indoors (Khoshnood et al. 2015; Wani et al. 2010). Residential areas play an important role for prevalence of giardiasis. In the present study, the relationship between the prevalence of infection and the residential area was statistically significant and similar to a study in South Khorasan (Taheri et al. 2011; Mahni et al. 2016; Tork et al. 2016; Rahimi-Esboei et al. 2013). The prevalence of infection in this work was higher in rural areas (6.4%) rather that urban region (3.0%), which can be explained by low levels of sanitation knowledge, lack of healthy drinking water reservoir in some villages, more contact with soil, unpleasant environmental conditions and large numbers of family members (Rahimi-Esboei et al. 2013).

A significant association was observed between the educational level of people referred to health centers and parasitic infection, as infection rates in illiterate people (6.2%) was greater than those of educated people (1.9%) which shows close agreement with finding of other studies (Heidari and Rokni 2003; Al-Megrin 2015).

Our study shows a seasonal pattern with infection rates peaking in the summer, which is in agreement with other studies (Martin et al. 2003; Yoder et al. 2010; Robertson and Gjerde 2001). The high number of infection cases in this season is probably due to more consumption of contaminated vegetables and fruits which leads to higher number of patients referring to laboratory diagnosis. Additionally, the moist and warm climate of Mazandaran in the summer provides proper conditions for survival of the cyst.

In our study, there was a significant relationship between job and rate of infection the prevalence rate of giardiasis among farmers (7.1%) was more than other jobs. This increase in the prevalence of these can be more exposure to soil or contaminated water (Feng and Xiao 2011). In this work, GWR was used to study geographical relationship of the prevalence of human giardiasis in relation with some important variables including precipitation, temperature and livestock. The results show that 61 and 68% of prevalence of human giardiasis are contributed to rainfall and contact to domastic animals, respectively. This fact highlights the great impact of these two major variables.

It is noteworthy to mention that Mazandaran Province annually attracts a great number of tourists, immigrants and refugees owing to its high potential regarding of holidays, job-seeking and financial activities. Travelers' diarrhea is considered to be a great threat for tourists and *G. lamblia* is

Table 1 Frequency of G. lamblia in Mazandaran Province by demographic data and risk factors

Age	Risk factors	No. specimens examined	No. positive (%)	OR (95% CI)	P value
< 564825 (3.8%)1 $-$ 5-982878 (9.4%)0.3 (0.2-0.6)<0.001	Age				
5-982878 (9.4%)0.3 (0.2-0.6)<0.000110-147.2236 (4.9%)0.7 (0.4-1.3)0.315-2498831 (3.1%)1.2 (0.6-2.1)0.425-3991329 (3.1%)1.2 (0.6-2.1)0.425-406892.3 (3.3%)1.1 (0.6-2.1)0.6Sex	< 5	648	25 (3.8%)	1	_
10-1472236 (4.9%)0.7 (0.4-1.3)0.315-2498831 (3.1%)1.2 (0.6-2.1)0.4≥ 4068923 (3.3%)1.1 (0.6-2.1)0.6≥ 4068923 (3.3%)1.1 (0.6-2.1)0.6Ser </td <td>5–9</td> <td>828</td> <td>78 (9.4%)</td> <td>0.3 (0.2–0.6)</td> <td>&lt; 0.0001</td>	5–9	828	78 (9.4%)	0.3 (0.2–0.6)	< 0.0001
15-24     988     31 (3.1%)     12 (0.6-2.1)     0.4       25-39     913     29 (3.1%)     1.2 (0.6-2.1)     0.6       Scv     23 (3.3%)     1.1 (0.6-2.1)     0.6       Scv     122 (4.7%)     1     0.6       Residence     209     100 (4.5%)     1.04 (0.7-1.3)     0.7       Residence     1     1     0.6     0.001       Consumed water     213     146 (6.4%)     1       Tab water     3984     186 (4.7%)     1     1       Shaft water     308     9 (2.9%)     1.6 (0.8-3.6)     0.1       Mineral water     308     9 (2.9%)     1.6 (0.8-3.6)     0.1       Job     1     1     -     -       Student     1587     71 (4.4)     1     -       Private business     1085     52 (4.7)     1.09 (0.7-1.5)     0.6       Housewife     1139     43 (3.7)     1.2 (0.8-1.7)     0.3       Government employee     348     11 (3.1)     1.7 (0.9-3.7)     0.7       Agriculture     629     45 (7.1)     0.06 (0.04-0.1)     0.001       Education     1     -     -     -       University     722     14 (1.4%)     1     -       Private	10–14	722	36 (4.9%)	0.7 (0.4–1.3)	0.3
25-39       913       29 (3.1%)       1.2 (0.6-2.1)       0.4 $\geq$ 40       689       23 (3.3%)       1.1 (0.6-2.1)       0.6         Ser	15–24	988	31 (3.1%)	1.2 (0.6–2.1)	0.4
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Sex         Sex           Male         2579         122 (4.7%)           Fenale         2009         100 (4.5%)         1.04 (0.7-1.3)         0.7           Residence         Itele (6.4%)         Itele (6.4%)         Itele (6.4%)         Itele (6.4%)         (0.001           Consumed water         Itele (6.4%)         1         (0.101	$\geq 40$	689	23 (3.3%)	1.1 (0.6–2.1)	0.6
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Fenale         2209         100 (4.5%)         1.04 (0.7-1.3)         0.7           Residence	Male	2579	122 (4.7%)		
Residence         Rural         2273         146 (6.4%)           Urban         2015         76 (6.30%)         2.2 (1.6 - 2.9)         < 0.001	Female	2209	100 (4.5%)	1.04 (0.7–1.3)	0.7
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Urban251576 (3.0%)2.2 (1.6 - 2.9)< 0.0001Consumed waterTT	Rural	2273	146 (6.4%)		
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Tab water $3984$ $186 (4.7\%)$ 1Shaft water $308$ $9 (2.9\%)$ $1.6 (0.8-3.6)$ $0.1$ Mineral water $496$ $27 (5.4\%)$ $0.8 (5-1.3)$ $0.4$ Job $Job$ $Job$ $Job$ $I$ $-$ Student $1587$ $71 (4.4)$ $1$ $-$ Private business $1085$ $52 (4.7)$ $1.09 (0.7-1.5)$ $0.6$ Housewife $1139$ $43 (3.7)$ $1.2 (0.8-1.7)$ $0.3$ Government employee $348$ $11 (3.1)$ $1.7 (0.9-3.7)$ $0.07$ Agriculture $629$ $45 (7.1)$ $0.06 (0.04-0.1)$ $< 0.0001$ Education $I$ $ -$ Primary $41477$ $85 (5.8\%)$ $0.6 (4-1.1)$ $0.7$ High school $1646$ $65 (3.9\%)$ $0.5 (0.3-0.8)$ $0.01$ University $722$ $14 (1.9\%)$ $0.8 (0.5-1.3)$ $< 0.0001$ Cottact to domestic animals $  -$ Yes $2427$ $21 (4.9)$ $1.2 (0.9-1.5)$ $0.1$ No $2361$ $0.12$ $ -$ Season $   -$ Winter $1167$ $39 (3.3\%)$ $1$ $-$ Spring $1166$ $59 (4.9\%)$ $0.6 (0.4-1.1)$ $0.06$ Summer $1256$ $76 (6.1\%)$ $0.5 (0.3-0.8)$ $0.002$ Autunn $106$ $46 (4.2\%)$ $0.8 (0.6-1.2)$ $0.5$ No $3702$ $176 (4.8\%)$ $ -$	Consumed water				
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Job         Student       1587       71 (4.4)       1       -         Private business       1085       52 (4.7)       1.09 (0.7-1.5)       0.6         Housewife       1139       43 (3.7)       1.2 (0.8-1.7)       0.3         Government employee       348       11 (3.1)       1.7 (0.9-3.7)       0.07         Agriculture       629       457 (7.1)       0.06 (0.04-0.1)       <001	Mineral water	496	27 (5.4%)	0.8 (.5–1.3)	0.4
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Government employee         348         11 (3.1)         1.7 (0.9-3.7)         0.07           Agriculture         629         45 (7.1)         0.06 (0.04-0.1)         < 0.0001	Housewife	1139	43 (3.7)	1.2 (0.8–1.7)	0.3
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Education         Illiterate       943       58 (6.2%)       1       -         Primary       1477       85 (5.8%)       0.6 (4-1.1)       0.7         High school       1646       65 (3.9%)       0.5 (0.3-0.8)       0.01         University       722       14 (1.9%)       0.8 (0.5-1.3)       < 0.0001	Agriculture	629	45 (7.1)	0.06 (0.04-0.1)	< 0.0001
Illiterate       943       58 (6.2%)       1       -         Primary       1477       85 (5.8%)       0.6 (4-1.1)       0.7         High school       1646       65 (3.9%)       0.5 (0.3-0.8)       0.01         University       722       14 (1.9%)       0.8 (0.5-1.3)       < 0.001	Education				
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University72214 (1.9%)0.8 (0.5–1.3)< 0.001Contact to domestic animalsYes2427121 (4.9)1.2 (0.9–1.5)0.1No2361101 (4.2)SeasonWinter116739 (3.3%)1-Spring119659 (4.9%)0.6 (0.4–1.1)0.06Summer125676 (6.1%)0.5 (0.3–0.8)0.002Autumn116948 (4.1%)0.8 (0.5–1.320.4Anti parasitic drug consumptionYes108646 (4.2%)0.8 (0.6–1.2)0.5No3702176 (4.8%)176 (4.8%)0.8 (0.6–1.2)0.5	High school	1646	65 (3.9%)	0.5 (0.3–0.8)	0.01
Contact to domestic animalsYes2427121 (4.9)1.2 (0.9–1.5)0.1No2361101 (4.2)Season1-Winter116739 (3.3%)1-Spring119659 (4.9%)0.6 (0.4–1.1)0.06Summer125676 (6.1%)0.5 (0.3–0.8)0.002Autumn116948 (4.1%)0.8 (0.5–1.320.4Anti parasitic drug consumptionYes108646 (4.2%)0.8 (0.6–1.2)0.5No3702176 (4.8%)176 (4.8%)1000001000000000000000000000000000000000000	University	722	14 (1.9%)	0.8 (0.5–1.3)	< 0.0001
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No2361101 (4.2)Season	Yes	2427	121 (4.9)	1.2 (0.9–1.5)	0.1
Season           Winter         1167         39 (3.3%)         1         –           Spring         1196         59 (4.9%)         0.6 (0.4–1.1)         0.06           Summer         1256         76 (6.1%)         0.5 (0.3–0.8)         0.002           Autumn         1169         48 (4.1%)         0.8 (0.5–1.32         0.4           Anti parasitic drug consumption	No	2361	101 (4.2)		
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Summer125676 (6.1%)0.5 (0.3–0.8)0.002Autumn116948 (4.1%)0.8 (0.5–1.320.4Anti parasitic drug consumption </td <td>Spring</td> <td>1196</td> <td>59 (4.9%)</td> <td>0.6 (0.4–1.1)</td> <td>0.06</td>	Spring	1196	59 (4.9%)	0.6 (0.4–1.1)	0.06
Autumn116948 (4.1%)0.8 (0.5–1.320.4Anti parasitic drug consumptionYes108646 (4.2%)0.8 (0.6–1.2)0.5No3702176 (4.8%)	Summer	1256	76 (6.1%)	0.5 (0.3–0.8)	0.002
Anti parasitic drug consumption         46 (4.2%)         0.8 (0.6–1.2)         0.5           Yes         3702         176 (4.8%)         0.5         0.5	Autumn	1169	48 (4.1%)	0.8 (0.5–1.32	0.4
Yes         1086         46 (4.2%)         0.8 (0.6–1.2)         0.5           No         3702         176 (4.8%)         0.5	Anti parasitic drug consumpt	tion			
No 3702 176 (4.8%)	Yes	1086	46 (4.2%)	0.8 (0.6–1.2)	0.5
	No	3702	176 (4.8%)		

considered as a one of major causative agents of diarrhea (Feng and Xiao 2011). Therefore, there is a danger of acquiring *Giardia* infection for tourists in these areas. Considering to these facts, a regular monitoring program should be undertaken to decrease the prevalence of giardiasis in this province.

# Conclusions

This for the first time this study identified and introduced a vital spatial and temporal pattern of human giardiasis distribution in the Mazandaran Province of northern Iran. These visual and statistical evidences for spatial clustering



Fig. 2 Geographic distribution by GIS-based map of estimated prevalence rates of giardiasis in Mazandaran Province, north of Iran

and seasonal patterns should be considered for proper disease control decisions and strategies. In addition, there is a decreasing temporal trend in the distribution of giardiasis rates and Amol is major hot spot identified area. The results of this study also provided a precise and specific spatial and temporal pattern of human giardiasis distribution in the northern of Iran. These evidences should be considered for proper to control of disease decisions and strategies.

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#### Compliance with ethical standards

**Conflict of interest** All authors declare that they have no conflict of interest.

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