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Estimation of the Parasitic Infection Prevalence in Children With *Helicobacter pylori* Infection in Ilam City (2012-2013)

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Background: *Helicobacter pylori* is a common cause of chronic infection in human beings. The infection has universal prevalence and contracts all age groups. Probably, these bacteria are the cause of the most common chronic bacterial infection in man and have infected approximately half of the world population. The urease of these bacteria degrades the urea in stomach's mucosa to ammoniac which results pH increment of the stomach lumen. This may allow the pathogenic intestinal protozoa to take the opportunity to cross through stomach's decreased pH situation and cause the disease.
Objectives: The current study aimed to evaluate the prevalence of parasitic infections (such as giardiasis) in children with *Helicobacter pylori* infection in Ilam city.
Patients and Methods: Following the sample collection during 12 months from children in Ilam (Ilam, Iran), *Helicobacter pylori* infection was determined based on stool antigen analysis (HPSA) by enzyme-linked immunosorbent assay (ELISA) method in children who had recurrent abdominal pain. Stool specimens were examined by the direct examination and spontaneous sedimentation method to detect both trophozoite and cyst of parasites.
Results: In this study 37 children with *H. pylori* infection were evaluated, and the patients with positive results for *Giardia lamblia*, and *Entamoebahistolytica/dispar* were found 29.7%, and 10.8% respectively.
Conclusions: The results of the current study suggest that *H. pylori* infection may provide favorable conditions for Giardiasis infection, but this presumption needs to be investigated further with more samples.

Keywords: Helicobacter pylori; Infection; Parasitic Diseases

1. Background

Helicobacter pylori causes the most common infection in human beings. The infection is widespread worldwide and affects all age groups. On the other hand, it is estimated that 50% of the world population are contaminated with *H. pylori*. In developed countries like United States, it is unusual to see infections in children, and usually adults are affected by *H. pylori*. But most of the children in developing countries are contaminated before 10, and 10% of people are contaminated before the age of 50 (1, 2). It has been reported that the prevalence of *H. pylori* in Iranian children is approximately 82-92% (3).

As one of its particular characteristics, *Helicobacter pylori* has a large amount of urease enzyme (six percent of total proteins produced by *H. pylori*) with intense activity. This enzyme degrades plasma urea secreted through stomach wall to ammonium ion that protects bacteria from the destructive effect of stomach acid by neutralization. There is a distinction between urease produced by *H. pylori* and other bacteria. Urease of *H. pylori* has two subunits with approximately 33 and 66 kDa molecular weight in contrast to other bacteria which have three subunits. UreA, and UreB code two subunits of urease made by *H. pylori* (1, 2). The pH increment of stomach's lumen facilitates passing through stomach's acid environment easily for some protozoa (4). *Giardia spp.* is a flagellated protozoan observed on the mucosa of duodenum, first section of jejunum, ileum, rarely on stomach's wall, and colon (4). The prevalence of Giardia *spp.* in the world and industrialized societies is 20-60% and 2-7%, respectively (5). Of course the prevalence rate varies in different countries, and depends on the hygiene level.

On the other hand, the studies have shown that un-

Implication for health policy/practice/research/medical education:
The current study is an original work and aimed to evaluate the relevance between *H. pylori* and parasitic infections.

der unsuitable situations, primarily when the acidity of stomach is reduced because of *H. pylori* infection, development of giardiasis related gastritis will be probable. Reduction of stomach's acidity due to urease of *H. pylori* is the risk factor for infection by *Giardia spp.* (6).

2. Objectives

The current study aimed to evaluate the prevalence of parasitic infections (giardiasis) in children with *Helicobacter pylori* infection in Ilam city.

3. Patients and Methods

This is a descriptive study. We followed all children who referred to laboratory with abdominal pain during 2012-2013. Children who were positive for *H. pylori* antigen in stool samples examined by enzyme-linked immunosorbent assay (ELISA) method entered the study. Sensitivity and specificity of this method are 83% and 92%, respectively. Stool specimens were examined via light microscope with 40× magnification power as direct examination of the parasite trophozoites. Also in sedimentation method fifty grams of feces was mixed with approximately 100 mL of tap water and sieved through 2 mL mesh sieve. Afterward, it was washed with 50 mL water and then pressed with a spatula to recover the water as much as possible. After 40 min, the supernatant was decanted to remain 50 mL of that. The beaker was refilled with tap water to final volume of 200 mL and then the suspension was allowed standing. After 40 min, the supernatant was decanted to save 30 mL, and 1 mL of that was examined (in approximately 200 µL aliquots) via light microscope with 100X magnification power to survey the presence of the parasite ova. Ziehl-Neelsen staining was performed to look for acid-fast protozoa (*Isospora belli*, *Cyclosporacayetanensis*, and *Cryptosporidium parvum*).

4. Results

In this study, 37 children out of total 68 children with abdominal pain were infected with *H. pylori*. Children without *H. pylori* infection were not included in the study. In the direct examination, trophozoite and cyst of *Giardia lamblia* were observed in 11 (29.7%) and 7 (18.9%) subjects, respectively. Nevertheless, we only evaluated children who had excreted trophozoite of *Giardia lamblia*, because the presence of trophozoite is the sign of acute Giardiasis. Cyst of *Entamoeba histolytica/dispar* was observed in 4 (10.8%) of 37 subjects. In sedimentation method, ova of parasites were never observed. At last, in Ziehl-Neelsen staining, acid-fast parasites were never observed in patients infected with *H. pylori*.

5. Discussion

H. pylori is the main cause of chronic stomach inflammation, peptic ulcer, duodenal ulcer, non-ulcerous dyspepsia, gastric cancer, and gastric mucosa associated lymphoid tissue lymphoma (7, 8). Urease of the bacteria can convert the urea of stomach wall to ammoniac. This would result the increment of stomach environment's pH (1, 9). Acidity of stomach is an innate immune system barrier against pathogens, therefore the diminished acidity will allow pathogens to break this barrier and go across it. Giardiasis, a parasitic infection of small intestine in most of the vertebrates and human beings, which has global prevalence, is the result of contamination by a flagellated protozoan called *Giardia lamblia* (10, 11). *Giardia lamblia* can be transmitted from affected person to others. Its transmission can occur through ingestion of contaminated water and food. It is the main cause of diarrhea in children, passengers, and homosexuals (12). Also, many factors like population density, weather situation, economic condition, and hygiene level would facilitate the situation for contamination by *Giardia lamblia* (10, 14). The prevalence of *Giardia spp.* in the world and the industrialized societies is 20-60% and 2-7%, respectively (5). Of course the prevalence rate varies in different countries, and depends on the hygiene level. On one hand according to a local epidemiological study on the prevalence of enteric parasites done in 2013, prevalence of *G. lamblia* in children was 11.7% in Ilam (15). On the other hand, the current study demonstrated that prevalence of *G. lamblia* in the *H. pylori* infected children of Ilam was approximately 50% (trophozoite and cyst). Considering these two facts it is not vague that *H. pylori* infection has affected the rate of parasitic infection in *H. pylori* infected children. The importance of polymicrobial infections has gained tremendous impact in recent years, and some synergistic infections have been identified (15). In synergistic polymicrobial infections, one microbe creates a favorable environment in order for another one to more easily colonize a specific niche of their common host (16). *H. pylori* has been linked to co-infections earlier, e.g. the fluke *Schistosoma japonicum* is associated with an alteration in the antibody response to *H. pylori* during co-infections (17). Another interesting example is co-infections of *H. pylori* and *Salmonella typhimurium* in mice (18). In another

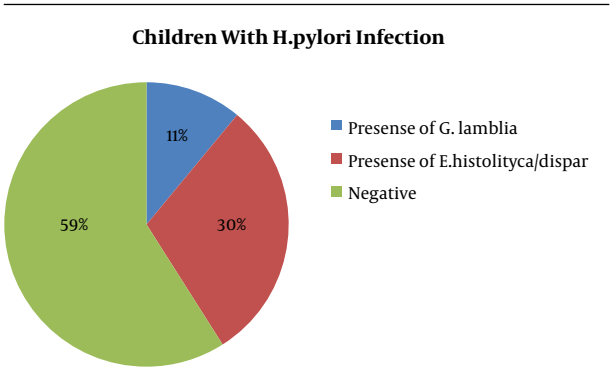

Figure 1. Proportion of Parasitic Infections in Patients Infected With *H. pylori*.

study, Maria PD et al. did not find an association between *H. pylori* and pediatric asthma (19). Johan A et al. found a significantly higher frequency of *Giardia spp*. infection in cases where infected children also harbored the bacterial pathogen *H. pylori* (20).

Moreira ED Jr et al. found an association between *H. pylori* infection and the presence of *G. lamblia* in feces (21). Isaeva G et al. showed that 100% of H. pylori-infection combined with iardiasis (9). Abou El-Hoda MM et al. found a significant increase in urease activity in the group with combined infection (Giardiasis and *H.pylori*) compared to the group infected with *G. lamblia* alone (22). The large amount of co-infections in our study is possibly due to an elevated risk of *Giardia spp.* colonization upon the presence of *H. pylori* in human patients or, alternatively, *H. pylori* colonization may be facilitated by a previous establishment of *Giardia spp.*

In this study, parasitic contamination was surveyed in children contaminated by *H. pylori*. Rate of parasitic contamination in *H. pylori* infected children was remarkable (active infection and carriers of *Giardia spp*. approximately were 50%). According to the Simultaneous study, the prevalence of *G. lamblia* in the children was 11.7% in Ilam (15), therefore there was a significant correlation between the contamination to *Giardia lamblia* and *H. pylori* infections in children.

Considering all the facts, it is obvious that acidity of stomach can be important. Also, pH increment would be concentrated on as a risk factor for contraction to parasites, especially those that can transmit through digestive tract like *Giardia lamblia*. More studies are suggested to get vigorous results, and validate the relevance between *H. pylori* infection and contamination to parasites.

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Authors' Contribution

Hossein Kazemian Loke developed the original idea and the protocol, abstracted and analyzed data, wrote the manuscript, and is guarantor. Financial and material supports for the research and work were applied by Dr. Sadeghifard.

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There is no conflict of interest to declare.

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