

Molecular identification of *Entamoeba histolytica* from patients infected with *Helicobacter pylori*

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ABSTRACT

Infection with *Entamoeba histolytica* and *Helicobacter pylori* is considered among the most common and serious infectious diseases, particularly in developing countries. The current study aimed to evaluate the prevalence of *E. histolytica* among patients with *H. pylori* infection and gastrointestinal symptoms in Mosul city and to study the phylogenetic relationships of the isolated strains. A total of 200 tissue biopsies were collected from 100 individuals of both sexes who had abdominal pain accompanied by gastrointestinal symptoms, including 100 gastric biopsies and 100 duodenal biopsies, from August 1, 2024, to March 1, 2025. The samples were examined using histological examination and conventional polymerase chain reaction (PCR), respectively, to identify *E. histolytica* in patients with positive *H. pylori* infection. The current study showed that the prevalence of *H. pylori* infection was 76%. Furthermore, parasite identification test results showed a prevalence of 26.66% for *E. histolytica*. Genetic analysis of *E. histolytica* was conducted, and two new strains were isolated and registered at the National Center for Biotechnology Information (NCBI) under accession numbers PV151461 and PV132085. The phylogenetic tree analysis of the locally isolated *E. histolytica* strains, which were recorded in GenBank, demonstrated high similarity (100%) between the two strains identified in the current study. Strain PV151461 showed high similarity with Iraqi strains from Tikrit and Diwaniya, OL771810 and ON086988, respectively. Isolate PV132085 showed a significant match with the Iraqi isolates obtained from Tikrit.

Keywords: *Entamoeba histolytica*, Gastric biopsies, *Helicobacter pylori*

1 INTRODUCTION

The parasite *Entamoeba histolytica* causes amebiasis, including amoebic dysentery, and is widely distributed worldwide, especially in developing countries with poor health conditions, such as Iraq. According to the World Health Organization, it affects 500 million people and causes 100,000 deaths annually [1–3]. Symptoms do not appear in many patients, and these individuals play an important role in the parasite's epidemiology. Symptoms of amebiasis are evident in 10% of infected individuals and are characterized by diarrhea with blood or mucus in the stool due to ulcers in the intestinal wall.

In severe cases, the amoeba spreads to other tissues and organs, such as the liver, lungs, skin, urinary system, and reproductive system, causing abscesses [4,5]. *Entamoeba histolytica* possesses several mechanisms that enhance its survival in the host during the course of disease, the most important of which is the presence of different genetic patterns within a single parasitic species. These patterns may contribute to rapid growth, efficient stress-response systems, and the stimulation of resistant cellular phases [6,7].

Studies have suggested a possible association between intestinal parasites, particularly *Entamoeba histolytica*, and the bacterium *Helicobacter pylori*. *Helicobacter*

pylori is one of the most common causes of gastritis and gastric cancer and is the main cause of gastric ulcers. Infection with this bacterium is widespread globally and affects approximately 50% of the world's population. Recent studies have recorded high prevalence rates in Iraqi cities, including Mosul [8–10]. *Helicobacter pylori* is transmitted through contaminated food and water and through contact with the saliva and other bodily fluids of infected persons. Infection with *Helicobacter pylori* occurs when the bacterium colonizes the gastric tissues and may contribute to serious gastric diseases [11, 12].

The transmission and infection routes of *Entamoeba histolytica* and *Helicobacter pylori* are similar because both are associated with contaminated water and food, as well as with cultural, social, and economic living standards and poor living conditions. Moreover, *Helicobacter pylori* causes changes in the gastric environment, such as increased gastric acidity or gastric mucosal hyperplasia, which are well-known complications of *H. pylori* infection. These changes may enhance infection with *Entamoeba histolytica* and bacteria and modify disease dynamics through synergistic or antagonistic interactions, leading to changes in disease severity and the host immune response [13, 14]. This study aimed to evaluate the prevalence of *Entamoeba histolytica* among individuals with *Helicobacter pylori* infection and gastrointestinal symptoms in Mosul city, and to construct a phylogenetic tree of the isolated strains.

2 MATERIALS AND METHODS

2.1 Samples collection

Random biopsy samples were collected from 100 patients of both sexes who had abdominal pain accompanied by gastrointestinal symptoms and who visited the endoscopy unit at the General Hospital in Mosul, northern Iraq, from August 1, 2024, to March 1, 2025. Two biopsies were collected from each patient, giving a total of 200 biopsies. The biopsies were obtained using endoscopic forceps, and sterilization precautions were followed. The biopsy samples were divided into two groups: the first group consisted of 100 gastric biopsies, and the second group consisted of 100 duodenal biopsies. The samples were preserved in sterile plastic containers containing 10% neutral-buffered formalin and stored at 20 °C until used for molecular and histopathological identification. The name, age, sex, and use of antibiotics or proton pump inhibitors for at least one week were recorded for each sample.

2.2 Histopathological study

To confirm the presence of *H. pylori*, histological sections were prepared from gastric biopsies. The samples were processed after removal from buffered formalin, and the procedure was performed as follows: washing, clearing and dehydration, impregnation, embedding, trimming, staining, and mounting [15].

2.3 Molecular study

Conventional polymerase chain reaction (PCR) was performed to detect the 18S rRNA gene of *E. histolytica* in tissue samples and the ureA gene of *Helicobacter pylori* in biopsy tissue samples [16, 17].

2.3.1 DNA extraction and conventional polymerase chain reaction (PCR)

DNA from *E. histolytica* was extracted from duodenal samples of 60 patients who tested positive for *H. pylori*. A genomic DNA extraction kit (Addbio, Korea) was used, and the DNA concentration of each patient sample was measured using a Qubit 2.0 fluorometer according to the manufacturer's instructions. The primers were used according to the Microgen (Korea) method, as shown in Table 1. The DNA was amplified in a final reaction mixture volume of 20 µL, as shown in Table 2. The polymerase chain reaction conditions for the amplification cycles of *Helicobacter pylori* and *E. histolytica* DNA are shown in Tables 3 and 4, respectively. The PCR products were separated using 1.5% agarose gel electrophoresis and examined under UV light.

Table 1 The primers used (their sequence and size) in the diagnosis of the *Entamoeba histolytica* and bacteria used in the study.

Microorganism	primer	Sequence (5'-3')	Product size (bp)	Reference
<i>Helicobacter pylori</i>	F	AGGCTTTTAGGGGTGTTAGGGGTTT	294 bp	[16]
	R	AAGTTACTTTCTAACACTAACGC		
<i>Entamoeba</i> genus	F	TAAGATGCACGAGAGCGAAA	770 bp	[17]
	R	GTACAAAGGGCAGGGACGTA		

Table 2 The PCR conditions of the amplification cycles and the number of cycles.

Ingredients	Volume/microliter
Master Mix	10
DNA Template	2
Forward primer10 pmol	1
Reversed Primer10 pmol	1
Nuclease free Water	6
Total volume	20

Table 3 The PCR conditions of the amplification cycles and the number of cycles for *H. pylori*.

Steps	Names	Temperature °C	Time	No. of cycles
1	Initial denaturation	95°C	5 min	1
2	Denaturation	95°C	0.30 sec	40
3	Primer annealing	62°C	0.30 sec	
4	Extension	72°C	0.45 sec	
5	Final extension	72°C	7 min	1

Table 4 The PCR conditions of the amplification cycles and the number of cycles for *Entamoeba* spp.

Steps	Names	Temperature °C	Time	No. of cycles
1	Initial denaturation	95°C	3 min	1
2	Denaturation	94°C	0.40 sec	40
3	Primer annealing	60°C	0.40sec	
4	Extension	72°C	0.40sec	
5	Final extension	72°C	5 min	1

2.3.2 Sequencing analysis

The amplification products (20 µL) from two *E. histolytica* samples isolated from duodenal biopsies were used for sequencing. The sequencing process was performed by NICEM (National Instrumentation Center for Environmental Management) at Seoul National University using a DNA sequencer (3730XL, Applied Biosystems). BioEdit software and BLAST (Basic Local Alignment Search Tool), available on the National Center for Biotechnology Information (NCBI) website, were used for homology analysis [18].

3 RESULTS AND DISCUSSION

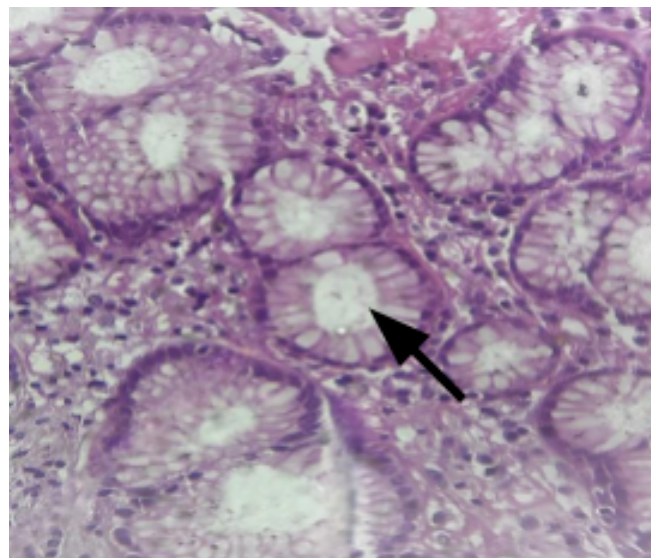
3.1 Identification of *helicobacter pylori* bacteria

3.1.1 Histopathological examination

The results of the histopathological examination for the identification of *H. pylori* in gastric biopsy samples taken from patients with abdominal pain and associated symptoms showed that 76% of the samples were positive for *H. pylori*, whereas 24% were negative, out of a total of 100 samples, as shown in Table 5 and Figure 1.

Table 5 Percentage of gastric biopsy samples with *H. pylori* by using the histopathological method

Number of biopsy samples	Positive H. P		Negative H. P	
	.NO	%	.NO	%
100	76	76	24	24

**Fig. 1** Histological section of stomach lining tissue showing *H. pylori* observed using Hematoxylin stain, 400X magnification.

3.1.2 Identification of *helicobacter pylori* in duodenal biopsy samples using polymerase chain reaction (PCR)

The current study showed that 60% of samples from patients infected with *H. pylori* were positive and 40% were negative, out of a total of 60 samples examined by polymerase chain reaction (PCR), as shown in Table 6. Figure 2 shows the positive results for *H. pylori* using agarose gel electrophoresis, with an amplicon size of 294 bp.

Table 6 Percentage of samples with *H. pylori* in duodenal biopsies samples by using PCR.

Number of biopsy samples	Positive H. P		Negative H. P	
	.NO	%	.NO	%
60	36	60	24	40

The present study found a high prevalence of *H. pylori* infection in duodenal biopsy samples. The high prevalence of *H. pylori* infection may be attributed to its global distribution, particularly in developing countries such as Iraq, where risk factors and favorable environmental conditions contribute to its dissemination [13]. Monitoring the prevalence of *H. pylori* infection in Iraq has revealed increased infection rates across various regions, consistent with the findings of the present study. In a study conducted in Zakho, the prevalence of *H. pylori* was 50% among 100 patients suffering from indigestion [19]. In Babel/Hilla, the infection rate was 73.9% [20], whereas

a rate of 60.7% was reported in a study conducted in Al-Muthanna and Al-Diwaniyah governorates [21].

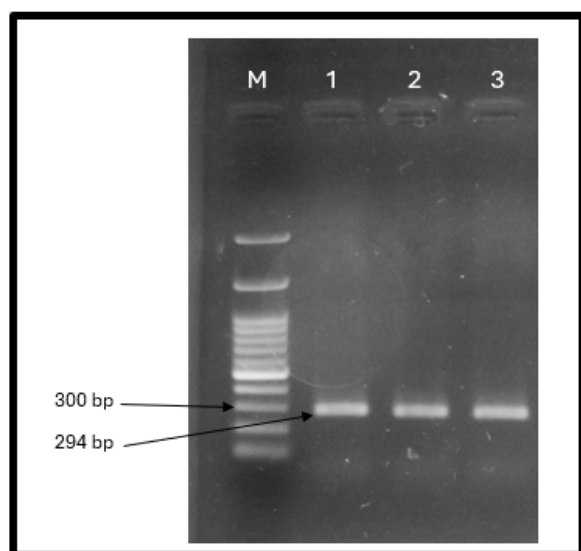


Fig. 2 Agarose gel electrophoresis of PCR product analysis for Urea A gene, using 1.5% agarose, 70V, 60Am for 60 min. (Lanes 1-3: 294 bp *H. pylori* isolates; M: 100bp DNA ladder).

The high prevalence of *H. pylori* infection recorded in the present study may be attributed to risk factors such as consumption of unhealthy foods, poor sanitation, contaminated food and water, poor personal hygiene, low living standards, and poor economic and cultural conditions. Many studies indicate that humans are the primary reservoir for this bacterium [22]. The bacterium is transmitted via fecal–oral and gastric routes [23]. In the present study, several methods were used to identify *H. pylori*, which are characterized by high sensitivity and specificity. Recent studies have shown that combining at least two methods is necessary for accurate identification of *H. pylori* infection due to variation in the sensitivity and specificity of the diagnostic tests used. In Mosul, two methods were used to diagnose *H. pylori* infection, and an infection rate of 58% was recorded among 50 biopsy samples that tested positive for *H. pylori* by molecular testing (PCR), histological examination, and the urease test [9].

The results of identifying *E. histolytica* in duodenal biopsies isolated from patients infected with *H. pylori* using polymerase chain reaction (PCR) showed the presence of *E. histolytica* DNA.

Examination of 60 intestinal biopsies positive for *H. pylori* revealed the presence of *E. histolytica* DNA by

agarose gel electrophoresis, with an amplicon size of 770 bp, as shown in Figure 3.

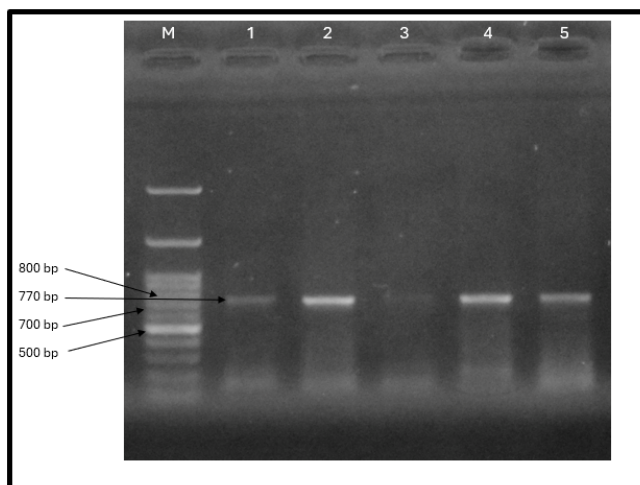


Fig. 3 Agarose gel electrophoresis of PCR amplicon of 18SrRNA gene, using 1.5% agarose, 70V, 60Am, for 60 min., (lanes 1,2,4,5:770 bp *Entamoeba histolytica* isolates; M: 100 bp DNA ladder)

3.1.3 Infection with *E. histolytica* in tissue biopsy samples

The current study recorded *E. histolytica* infection in 26.66% of intestinal biopsy samples, whereas 73.33% were negative for *E. histolytica* among the examined patients, as shown in Table 7.

Table 7 The percentage of infection with *E. histolytica* from tissue biopsy samples of patients infected with *Helicobacter pylori* using polymerase chain reaction (PCR).

Number of biopsies examined from patients diagnosed with <i>H. pylori</i>	positive samples for <i>E. histolytica</i>	Negative samples for <i>E. histolytica</i>
	(%) Number	(%) Number
60	16 (26.66)	44 (73.33)

The parasite *Entamoeba histolytica* and the bacterium *Helicobacter pylori* are highly prevalent among patients with gastrointestinal symptoms, and both affect the digestive tract with overlapping pathological manifestations [24,25]. Both organisms inhabit the human host's digestive system, which may exacerbate symptom severity and influence host immune responses, making accurate diagnosis difficult [26,27]. This study was conducted to determine the prevalence of *E. histolytica* in patients diagnosed with *H. pylori*. The results of the current study recorded a high rate of co-infection. The results shown in Table 7 revealed a co-infection rate of 26.66%, with

intestinal biopsy samples positive for *E. histolytica* in patients diagnosed with *H. pylori* using polymerase chain reaction (PCR).

By reviewing recorded rates in Iraq and other countries, as well as the prevalence of *E. histolytica* and *H. pylori* infections, the importance of studying co-infection with these two pathogens becomes evident. This is particularly important for common medically important parasitic species such as *E. histolytica*, which causes diseases or disorders of the digestive system. Therefore, precise diagnostic methods are required to determine the true prevalence of co-infection. In a study conducted by Toma et al. (2023) [28] in Najaf city, the prevalence of *E. histolytica* was 37.28%, the prevalence of *H. pylori* was 45.76%, and the prevalence of infection with both organisms was 16.94% among a total of 350 patients in a study on the role of cytokines in patients infected with *H. pylori* and *E. histolytica*. In Sudan, a study aimed to determine the prevalence of *E. histolytica* infection and its relationship with *H. pylori* found that the prevalence of *E. histolytica* infection was 52.6% among patients with *H. pylori* [29]. In Sharqat, Salah Al-Deen Governorate, the prevalence of infection with *E. histolytica* and *H. pylori* was 29.03% across 409 samples, as determined by smear and concentration methods. The results of these studies are consistent with the current study's findings on the prevalence of *E. histolytica* infection among patients diagnosed with *H. pylori* in Mosul city [30].

The high infection rates of *E. histolytica* and *H. pylori* recorded in the present study may be attributed to the increased risk of *E. histolytica* infection in patients diagnosed with *H. pylori*. This may result from changes

in the gastric environment induced by *H. pylori*, including increased gastric acidity via urease activity and gastric mucosal hyperplasia [18]. In addition, infection with *E. histolytica* may stimulate host immune responses similar to those induced by *H. pylori*. As a result of their coexistence, both organisms may contribute to robust Th1 cell polarization, thereby stimulating the secretion of pro-inflammatory cytokines such as IL-8. This may lead to significant exacerbation of gastrointestinal mucosal damage and an increased risk of peptic ulcer disease [31, 32]. These mechanisms may enhance the synergistic relationship between *E. histolytica* and *H. pylori*, as reflected in altered clinical symptoms for each organism and increased gastrointestinal symptom severity [33]. The presence of tissue-invasive amoebae in patients infected with *H. pylori* may stimulate the emergence of novel genetic patterns within a single parasitic species, leading to rapid growth and the evolution of highly efficient stress-response systems [6, 7].

3.1.4 Gene sequencing

Table 8 shows the similarity of the *E. histolytica* isolates identified in the current study with global isolates. The BLAST analysis and GenBank database results showed 100% similarity between the two newly identified isolates in the current study. These isolates were registered in GenBank under accession number PV151461.1. The similarity percentages were also close to those of several isolates from Tikrit and Diwaniyah, with varying degrees of similarity, as shown in Table 8.

Table 8 Shows the approximate similarity between the *E. histolytica* parasite identified in the current study and registered in the gene bank with the serial Number: (PV151461.1).

	Isolate	Accession & Version	City/ Country	Total Score	Compatibility %	Acc. Length
1	<i>Entamoeba histolytica</i> isolate MR-ENT-63 small subunit ribosomal RNA gene, partial sequence	PV151461.1	Mosul, Iraq	1461	100	791

Continued on next page

	Isolate	Accession & Version	City/ Country	Total Score	Compatibility %	Acc. Length
2	<i>Entamoeba histolytica</i> isolate H211110-028_M01_27_E1.ab1770 small subunit ribosomal RNA gene, partial sequence	OL771810.1	Tikrit, Iraq	1253	96.06	770
3	<i>Entamoeba histolytica</i> isolate H211110-028_C01_21_E1.ab11225 small subunit ribosomal RNA gene, partial sequence	OL771814.1	Tikrit, Iraq	846	89.8	1225
4	<i>Entamoeba histolytica</i> isolate H210824-030_A15_1_E1.ab1658 small subunit ribosomal RNA gene, partial sequence	OL771809.1	Tikrit, Iraq	821	93.78	658
5	<i>Entamoeba histolytica</i> isolate H210824-030_I02_16_E1.ab1660 small subunit ribosomal RNA gene, partial sequence	OL771806.1	Tikrit, Iraq	821	93.78	660
6	<i>Entamoeba histolytica</i> isolate H210824-030_K02_18_E1.ab1515 small subunit ribosomal RNA gene, partial sequence	OL771805.1	Tikrit, Iraq	811	95.25	514
7	<i>Entamoeba histolytica</i> isolate H211110-028_A01_20_E1.ab11452 small subunit ribosomal RNA gene, partial sequence	OL771815.1	Tikrit, Iraq	750	92.88	1452
8	<i>Entamoeba histolytica</i> strain EH-2 small subunit ribosomal RNA gene, partial sequence	ON086988.1	Diwanyiah, Iraq	741	93.27	614
9	<i>Entamoeba histolytica</i> isolate MR-ENT-18 small subunit ribosomal RNA gene, partial sequence	PV132085.1	Mosul, Iraq	695	95.32	468
10	<i>Entamoeba histolytica</i> strain EH-1 small subunit ribosomal RNA gene, partial sequence	ON086987.1	Diwanyiah, Iraq	584	91.92	582

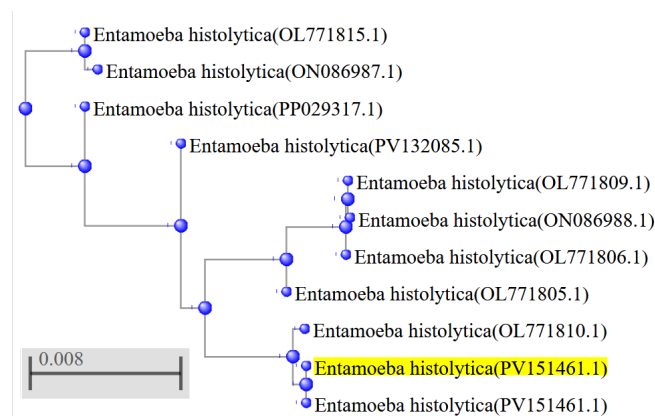


Fig. 4 Shows the evolutionary tree that demonstrates the relationship between the isolate identified with the current study with the serial number of (PV151461.1).

For the second isolate, which was registered in the international GenBank database under accession

number PV132085.1, the analysis showed that it matched several isolates identified in Tikrit, Iraq, as shown in Table 9.

The purpose of studying the genetic sequencing of parasites is to confirm parasite identification, map the parasite's genetic structure, understand the origin and evolution of infection, and identify the source of infection. Based on the nucleotide sequence results and phylogenetic tree analysis of locally isolated *E. histolytica* strains registered in GenBank, a 100% similarity was observed between the two strains identified in the current study. Isolate PV151461 showed high similarity with Iraqi isolates from Tikrit and Diwaniyah, OL771810 and ON086988, respectively. Isolate PV132085 showed a high degree of similarity with Iraqi isolates from Tikrit, as shown in Tables 8 and 9.

Table 9 Shows the approximate similarity between the *E. histolytica* parasite identified in the current study and registered in the gene bank with the serial Number: (PV132085.1).

	Isolate	Accession & Version	City/ Country	Total Score	Compatibility %	Acc. Length
1	<i>Entamoeba histolytica</i> isolate MR-ENT-18 small subunit ribosomal RNA gene, partial sequence	PV132085.1	Mosul, Iraq	865	100	468
2	<i>Entamoeba histolytica</i> isolate H211110-028_M01_27_E1.ab1770 small subunit ribosomal RNA gene, partial sequence	OL771810.1	Tikrit, Iraq	791	99.54	770
3	<i>Entamoeba histolytica</i> isolate H211110-028_C01_21_E1.ab11225 small subunit ribosomal RNA gene, partial sequence	OL771814.1	Tikrit, Iraq	782	99.31	1225
4	<i>Entamoeba histolytica</i> isolate H211110-028_A01_20_E1.ab11452 small subunit ribosomal RNA gene, partial sequence	OL771815.1	Tikrit, Iraq	769	99.3	1452

Continued on next page

	Isolate	Accession & Version	City/ Country	Total Score	Compatibility %	Acc. Length
5	<i>Entamoeba histolytica</i> isolate H210824-030_K02_18_E1.ab1515 small subunit ribosomal RNA gene, partial sequence	OL771805.1	Tikrit, Iraq	769	99.53	514

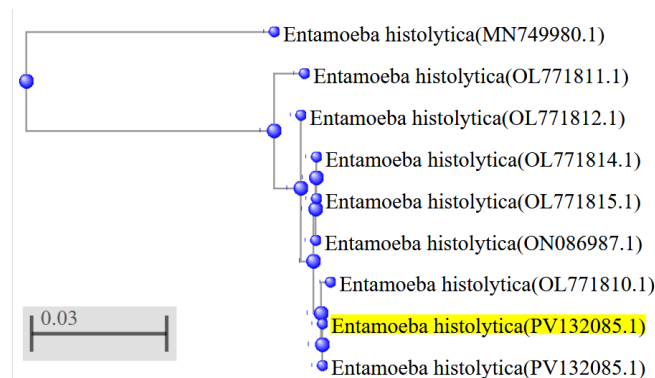


Fig. 5 Shows the evolutionary tree that demonstrates the relationship between the isolate identified with the current study with the serial number of (PV132085.1).

Although genetic evolutionary analysis has not been clearly addressed in several studies, especially in cases of co-infection with other organisms such as *H. pylori*, it is of great importance for understanding the evolutionary relationships among organisms and their transmission patterns. Through such studies, the relationships among these strains can be determined, and the source of infection and parasite virulence may be identified. This ancient protozoan is a clear example of how parasites have evolved with humans by exploiting multiple mechanisms to evade immune responses, interacting with both non-pathogenic and pathogenic gut microbes, such as *H. pylori*, to meet their nutritional and protective needs, and using host resources for growth, division, and encystation. In addition, the tissue-lysing amoeba possesses multiple mechanisms that enhance its survival in the host during the course of disease, the most important of which is the presence of different genetic patterns within a single parasitic species. These patterns may contribute to rapid

growth, highly efficient stress-response systems, and the stimulation of resistant cellular phases [8, 9].

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Fig. 6 *Entamoeba histolytica* isolate MR-ENT-63 small subunit ribosomal RNA gene, partial sequence GeneBank: PV151461.1.

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Fig. 7 *Entamoeba histolytica* isolate MR-ENT-18 small subunit ribosomal RNA gene, partial sequence GeneBank: PV132085.1

4 CONCLUSION

The study recorded two new *E. histolytica* strains, named after the researchers, in the National Center for Biotechnology Information (NCBI) database. These isolates were obtained from patients infected with *H. pylori*. These new strains may be more virulent due to their association with *H. pylori* infection, and this virulence may contribute to the exacerbation of gastrointestinal symptoms and the modulation of the host immune response. The study recommends conducting extensive future research

on the relationship between *H. pylori* infection and the virulence of *E. histolytica* strains, as well as identifying which strains are most prevalent among patients with *H. pylori* infection.

Author contributions

The first researcher (Khataab. M. N) supplied the samples of the study from the hospitals in Mosul city, worked and conducted the tests and the second researcher (Hamoo. R.N) provided the idea of the research.

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No funds received.

Data availability

N/A

DECLARATIONS

Conflict of interest

The authors declare no competing interests

Consent to publish

N/A

Ethical approval

This study was approved by the institutional ethics committee of the Ministry of Health/ Directorate of Health in Nineveh/ Training and Development Center. No. 58927 in 9 Dec. 2024.

Ref. 28 "Retracted"

REFERENCES

- [1] Hajare ST, Gobena RK, Chauhan NM, Eriso F. Prevalence of Intestinal Parasite Infections and Their Associated Factors among Food Handlers Working in Selected Catering Establishments from Bule Hora, Ethiopia. *BioMed Research International*. 2021;2021:6669742. [10.1155/2021/6669742](https://doi.org/10.1155/2021/6669742)
- [2] Horikoshi Y, Ibrahim UM, Morris SK. School-Based Approach for Parasitic Disease Control in Japan and Africa. *Pediatrics International*. 2021;63(3):264-9. [10.1111/ped.14535](https://doi.org/10.1111/ped.14535)
- [3] Ahmed M. Intestinal Parasitic Infections in 2023. *Gastroenterology Research*. 2023;16(3):127-40. [10.14740/gr1622](https://doi.org/10.14740/gr1622)
- [4] Haque R, Duggal P, Ali IM, Hossain MB, Mondal D, Sack RB, et al. Innate and Acquired Resistance to Amebiasis in Bangladeshi Children. *The Journal of Infectious Diseases*. 2002;186(4):547-52. [10.1086/341566](https://doi.org/10.1086/341566)
- [5] Mahmud R, Lim YAL, Amir A. *Medical Parasitology: A Textbook*. Cham, Switzerland: Springer International Publishing; 2017. Cited pages: 5–9. [10.1007/978-3-319-68795-7](https://doi.org/10.1007/978-3-319-68795-7)
- [6] Even G, Lokmer A, Rodrigues J, Audebert C, Viscogliosi E, Ségurel L, et al. Changes in the Human Gut Microbiota Associated with Colonization by *Blastocystis* sp. and *Entamoeba* spp. in Non-Industrialized Populations. *Frontiers in Cellular and Infection Microbiology*. 2021;11:533528. [10.3389/fcimb.2021.533528](https://doi.org/10.3389/fcimb.2021.533528)
- [7] Guillén N. Pathogenicity and Virulence of *Entamoeba histolytica*, the Agent of Amoebiasis. *Virulence*. 2023;14(1):2158656. [10.1080/21505594.2022.2158656](https://doi.org/10.1080/21505594.2022.2158656)
- [8] Hussein RA, Al-Ouqaili MTS, Majeed YH. Detection of *Helicobacter pylori* Infection by Invasive and Non-Invasive Techniques in Patients with Gastrointestinal Diseases from Iraq: A Validation Study. *PLOS ONE*. 2021;16(8):e0256393. [10.1371/journal.pone.0256393](https://doi.org/10.1371/journal.pone.0256393)
- [9] Muhammad MAG, Altaie AA, Abdullah BA. Invasive Methods for Detection of *Helicobacter pylori* Infection in Gastric Disorders Patients Undergoing Endoscopy from Mosul City, Iraq. *Romanian Journal of Infectious Diseases*. 2024;27(2):105-11. [10.37897/RJID.2024.2.11](https://doi.org/10.37897/RJID.2024.2.11)
- [10] Jaka H, Smith SI. Forty Years of *Helicobacter pylori*: The African Perspective. *Digestive Diseases*. 2024;42(2):161-5. [10.1159/000535263](https://doi.org/10.1159/000535263)
- [11] Yang H, Hu B. Immunological Perspective: *Helicobacter pylori* Infection and Gastritis. *Mediators of Inflammation*. 2022;2022:2944156. [10.1155/2022/2944156](https://doi.org/10.1155/2022/2944156)

- [12] Duan M, Li Y, Liu J, Zhang W, Dong Y, Han Z, et al. Transmission Routes and Patterns of *Helicobacter pylori*. *Helicobacter*. 2023;28(1):e12945. [10.1111/hel.12945](https://doi.org/10.1111/hel.12945)
- [13] Fahmi Y, Dyab AK, Abd Ella OH, Said MTA, Abuelwafa EA. Co-Infection between Intestinal Parasites and *Helicobacter pylori*: A Brief Review. *International Journal of Chemical and Biochemical Sciences*. 2024;25(14):553-6
- [14] Tabesh A, Antillon RA, Kondradzhyan M, Tan AZ. Prevalence and Resistance of *Helicobacter pylori* in a Predominantly Hispanic Population. *World Journal of Gastrointestinal Endoscopy*. 2024;16(9):526-32. [10.4253/wjge.v16.i9.526](https://doi.org/10.4253/wjge.v16.i9.526)
- [15] Luna LG, editor. *Manual of Histologic Staining Methods of the Armed Forces Institute of Pathology*. 3rd ed. New York: Blakiston Division, McGraw-Hill; 1968
- [16] Bickley J, Owen RJ, Fraser AG, Pounder RE. Evaluation of the Polymerase Chain Reaction for Detecting the Urease C Gene of *Helicobacter pylori* in Gastric Biopsy Samples and Dental Plaque. *Journal of Medical Microbiology*. 1993;39(5):338-44. [10.1099/00222615-39-5-338](https://doi.org/10.1099/00222615-39-5-338)
- [17] Khairnar K, Parija SC. A Novel Nested Multiplex Polymerase Chain Reaction (PCR) Assay for Differential Detection of *Entamoeba histolytica*, *E. moshkovskii* and *E. dispar* DNA in Stool Samples. *BMC Microbiology*. 2007;7(1):47. [10.1186/1471-2180-7-47](https://doi.org/10.1186/1471-2180-7-47)
- [18] Bowles J, Blair D, McManus DP. Genetic Variants within the Genus *Echinococcus* Identified by Mitochondrial DNA Sequencing. *Molecular and Biochemical Parasitology*. 1992;54(2):165-73. [10.1016/0166-6851\(92\)90109-W](https://doi.org/10.1016/0166-6851(92)90109-W)
- [19] Saeed AY, Rashad BH, Ali BN, Sulaivany AH, Ibrahim KS. *Helicobacter pylori* Infection: Prevalence, Risk Factors, and Treatment Efficacy in Symptomatic Patients in Zakho City, Kurdistan Region, Iraq. *Cureus*. 2024;16(11):e73873. [10.7759/cureus.73873](https://doi.org/10.7759/cureus.73873)
- [20] Al-Sabary AH, Abdul Razzaq MS, Wtw MA. Genotyping of *Helicobacter pylori* Isolated from Iraqi Patient with Upper Gastrointestinal Disease. *Journal of Global Pharma Technology*. 2017;9(9, Part B):121-7. Available from: <https://jgpt.co.in/index.php/jgpt/article/view/521>
- [21] Jabbar AM, Al-Obaidi LAH. Isolation and Molecular Detection of *Helicobacter pylori* from Biopsy Samples of Gastritis Patients in Iraq. *International Journal of Research Studies in Biosciences*. 2015;3(5):193-200. Available from: <https://www.arcjournals.org/article/3413>
- [22] Zamani M, Ebrahimitabar F, Zamani V, Miller WH, Alizadeh-Navaei R, Shokri-Shirvani J, et al. Systematic Review with Meta-Analysis: The Worldwide Prevalence of *Helicobacter pylori* Infection. *Alimentary Pharmacology & Therapeutics*. 2018;47(7):868-76. [10.1111/apt.14561](https://doi.org/10.1111/apt.14561)
- [23] Naqid IA, Al-Brefkani AMT, Hussein NR. A Study of Prevalence and Risk Factors for *Helicobacter pylori* Infection among Adults in Duhok Province, Kurdistan Region, Iraq. *Archives of Razi Institute*. 2024;79(2):272-8. [10.32592/ARI.2024.79.2.272](https://doi.org/10.32592/ARI.2024.79.2.272)
- [24] Bin Mohanna MA, Al-Zubairi LM, Sallam AK. Prevalence of *Helicobacter pylori* and Parasites in Symptomatic Children Examined for *Helicobacter pylori* Antibodies, Antigens, and Parasites in Yemen. *Saudi Medical Journal*. 2014;35(11):1408-11. [10.15537/1658-3175.6089](https://doi.org/10.15537/1658-3175.6089)
- [25] Ibrahim A, Ali YBM, Abdel-Aziz AA, El-Badry AA. *Helicobacter pylori* and Enteric Parasites Co-Infection among Diarrheic and Non-Diarrheic Egyptian Children: Seasonality, Estimated Risks, and Predictive Factors. *Journal of Parasitic Diseases*. 2019;43(2):198-208. [10.1007/s12639-018-1075-y](https://doi.org/10.1007/s12639-018-1075-y)
- [26] Mina S, Daher S, Mina N, Khoder G. Concomitant Infection of *Helicobacter pylori* and

- Intestinal Parasites: Burden, Sociodemographic and Clinical Characteristics in Hospitalized Children and Adolescents in Northern Lebanon. *F1000Research*. 2024;13:500. Version 2. [10.12688/f1000research.148550.2](https://doi.org/10.12688/f1000research.148550.2)
- [27] Almaw A, Berhan A, Ayele A, Fentie A, Abebaw A, Malkamu B, et al. Prevalence of Intestinal Parasites and *Helicobacter pylori* Coinfection, and Contributing Factors among Patients with Gastrointestinal Manifestations at Addis Zemen Primary Hospital, Northwest Ethiopia. *Gut Pathogens*. 2024;16(1):48. [10.1186/s13099-024-00642-8](https://doi.org/10.1186/s13099-024-00642-8)
- [28] Toma RS, Al-Hadraawy SK, Aljanaby AAJ. Study Role of Cytokine in Patients with Co-Infection *H. pylori* and *E. histolytica*. In: AIP Conference Proceedings. vol. 2977. AIP Publishing; 2023. p. 040004. Retracted by AIP Publishing; do not use as supporting scientific evidence. [10.1063/5.0182315](https://doi.org/10.1063/5.0182315)
- [29] Elfadul ME, Lowaty JY, Alkhidir AAI. Determination of Possible Association between *Helicobacter pylori* and the Infection of *Entamoeba histolytica*, *Giardia lamblia* and *Entamoeba coli* in Omdurman-Sudan. *Sudan Medical Laboratory Journal*. 2022;10(1):39-44. The published title spells *Giardia* as “*Gardia*”. [10.52981/smlj.v10i1.2776](https://doi.org/10.52981/smlj.v10i1.2776)
- [30] Habeeb FF, Abed AM. The Relationship between *Helicobacter pylori* Infection and Some Intestinal Parasites for Patients Attending to Shirqat General Hospital/Salah Al-Din Governorate. *International Journal for Research in Applied Sciences and Biotechnology*. 2021;8(5):125-9. The DOI is printed as 10.31033/ijrasb.8.4.17 in the article PDF, although some secondary metadata lists 10.31033/ijrasb.8.5.17. [10.31033/ijrasb.8.4.17](https://doi.org/10.31033/ijrasb.8.4.17)
- [31] Krzyżek P, Gościński G. Frequency and Immunological Consequences of *Helicobacter pylori* and Intestinal Parasite Co-Infections: A Brief Review. *Annals of Parasitology*. 2017;63(4):255-63. [10.17420/ap6304.112](https://doi.org/10.17420/ap6304.112)
- [32] Tilahun M, Gedefie A, Belayhun C, Sahle Z, Abera A. *Helicobacter pylori* Pathogenicity Islands and *Giardia lamblia* Cysteine Proteases in Role of Coinfection and Pathogenesis. Infection and Drug Resistance. 2022;15:21-34. [10.2147/IDR.S346705](https://doi.org/10.2147/IDR.S346705)
- [33] Abd El Hameed YF, Boghdadi AM, Ghobrial CM, Hassan MA. Association of *Helicobacter pylori* and Parasitic Infections in Childhood: Impact on Clinical Manifestations and Implications. *Journal of Parasitic Diseases*. 2021;45(3):790-6. [10.1007/s12639-021-01362-5](https://doi.org/10.1007/s12639-021-01362-5)

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