Original article

Helicobacter pylori Infection in Association with Iron Deficiency Anemia in Adults at Legacy Clinics, in Rwanda: A Clinic based Cross-Sectional Study

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ABSTRACT:

Background: Many studies link *H. pylori* infection with high depletion of serum iron in human hosts, and report it as the root leading to iron deficiency anemia (IDA). This study aimed to evaluate the association between *H. pylori* infection and iron deficiency in adults at Legacy Clinics, in Rwanda.

Methods: This was a Clinic based cross-sectional study conducted on *H. pylori*, iron and ferritin statuses, hemoglobin (Hb), mean corpuscular volume (MCV), and hematocrit (Hct). A sample of 200 adults who attended the clinic during the study period were recruited for the study. Data were collected using Microsoft Office Excel and exported to SPSS Version 25 and GraphPad Prism for analysis. A p-value less than 0.05 was considered statistically significant.

Results: Among 200 adults included in this study, 56.5% were females and 43.5% were males. The mean age of the participants was 39 (sd. = ±12.9) years old. The overall prevalence of anemia and *H. pylori* infection was 29.5% and 29.0%, respectively. The proportion of being infected by *H. pylori* and experiencing IDA was 11.9 times the proportion of having IDA without being infected by *H. pylori*.

Conclusion: This study showed significant *H. pylori* infection in participants. There was a difference in IDA in participants infected with *H. pylori* and negative *H. pylori* group. The study showed a great risk of experiencing insufficient iron levels and iron-associated anemia in the group of people infected by *H. pylori* compared to the normal group.

Keywords: *Helicobacter pylori* infection, iron deficiency, anemia, adult.

1. INTRODUCTION

Helicobacter pylori (H. pylori), previously known as *Campylobacter pylori*, is a gram-negative, microaerophilic, spiral bacterium usually found in the stomach.[1] Marshall and Warren were the first to identify *H. pylori* in 1982.[2] Ali in his study of 2019 associated this bacterium with mucosa-associated lymphoid tissue in the stomach, esophagus, colon, rectum, extra nodal marginal zone B-cell lymphoma, and so-called diffuse large B-cell lymphoma.[3] The prevalence of *H. pylori* infection is abundant where it remains high and related to socioeconomic status in developing countries.[4] The study by Burucoa and Axon in 2017 showed the prevalence of *H. pylori* infection of 30% to 50% in developed countries and 85% to 95% in developing countries including Rwanda.[5] Kabakambira *et al.* reported the prevalence of *H. pylori* infection in Rwanda to be 75% of the population and was the same in previous three decades earlier in Kigali City [6].

Iron depletion is the result of reduction in iron stores and is the early asymptomatic stage of micronutrient deficiency.[7] Like iron, ferritin is also stored intracellularly as a best indicator of iron deficiency,[8] however, it is sometimes

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acted as an acute phase protein where it is found elevated during infection, malignancy and chronic inflammation,[8] in overweight and obese people.[9] Some studies concluded that iron deficiency progresses to iron deficiency anemia (IDA)if not corrected [10] and IDA leads to irreversible health disorders including impaired immune function.[11-14] Iron has a vital role in many biological functions including energy production, respiration, and cell proliferation. Iron is an important element in living systems, as it participates in a series of metabolic processes, including DNA synthesis and oxygen and electron transport [15].

There is an assumption that most patients diagnosed of *H. pylori* infection are considered to be at high risk of iron deficiency and reduced iron reserves.[16] On the other hand, some studies have showed that resolution of *H. pylori* infection would not significantly improve the iron status or reduce iron in adults.[17] The exact relationship between *H. pylori* infection and iron deficiency anemia remains a topic of discussion. In Rwanda, similar studies were conducted among pediatric population, thus, this study aimed to evaluate the association between *H. pylori* infection and Iron deficiency anemia in adults at Legacy Clinics, in Rwanda.

2. MATERIALS AND METHODS

2.1. Study design and setting

This was a cross sectional, and Clinic based study that carried out for outpatients from August to September 2022 in Internal Medicine (IM), and General Medicine departments at Legacy Clinics, Kigali City – Rwanda. Legacy Clinics and Diagnostics Ltd is a private, healthcare-based institution located in the central Rwanda, Kigali City, Kicukiro District, Nyarugunga Sector.

2.2. Study Populationand sample size

Being 18 years of age or above and attending Legacy Clinics' General Medicine and/or Internal Medicine defined the population of this study. The simple random recruitment among the target population was used to estimate the sample size of the current study. Participants of the study were recruited strictly from the defined population.

2.3. Inclusion and Exclusion Criteria

Adults who attended Legacy Clinics in General Medicine and/or Internal Medicine, in the time limitations for this study, were included in the sample as the main criterion. Adult patients, whose laboratory request showed blood tests for FBC, serum iron and Ferritin and stool test for *H. pylori* Antigens, were selected. Selected patients were informed about the study and those who verbally accepted to participate were enrolled in the study.

Patients other than adults were excluded from the study. Adults who had laboratory request not including the four tests of this study were also excluded from the sample size. Emancipated minors and unconscious patients did not attend the clinic during the study period, thus they were not included in the study. Eligible participants, who refused the participation during verbal consenting, were strictly excluded from the study.

2.4. Ethical approvals

The proposal of this study was submitted at Ruhengeri Institute of Higher Education and Legacy Clinics' Ethics committee. Both institutions approved this study to be conducted as proposed. Personal identifications of participants were kept confidential hence; anonymous presentation of data was used. The participation in the study was voluntary and approved after the verbal consent is done by the selected participant.

2.5. Blood and stool Sample collection

Blood samples for participants were collected according to standard operating procedures for sample collection. Two 4 ml tubes, the first with clotting activator red top tube and the second with EDTA lavender top tube in that order, were collected for each participant. Participant's safety was professionally maintained during blood sample collection as very sensitive procedure of this study.

After blood collection, each participant was given a container well labeled with a barcode for stool sample collection. Instructions for acceptable stool sample collection were reminded to the participant. Containers had scoops attached the cap; and using the scoop, 1-3 spoons of stool from areas which appear bloody, slimy, or watery were collected into the container. If the stool is formed, the small amounts of about three grams from each end and from the middle were put into the container. Stool samples were submitted in sample collection room for verification and acceptance. Participant's blood and stool samples were timely transported by trained laboratory attendants to the testing areas for analysis. The transportation strictly respected the standard procedure of samples transportation for laboratory testing.

2.6. Laboratory analysis

Requested tests were performed at Legacy Pathology Laboratory which is accredited to ISO 15189 version 2012 by Kenya Accreditation Service (KENAS).

2.6.1. Full Blood Count

After receiving in hematology area, EDTA samples were immediately put on the roll mixer. These samples were used to analyze the full blood count on Sysmex XN 500i.Each sample was slightly mixed by inverting the tube up down clockwise 5 to 8 times. The barcode label on each tube containing whole blood sample had to be scanned prior to the analysis on the automated cells counter. The result was interfaced in the Laboratory Information System for post analytical procedures and on the other hand, recorded in the data collection Excel sheet for this study. Only hemoglobin, hematocrit, and MCV were collected from the FBC participants.

2.6.2. Analysis of Iron and Ferritin levels

Samples in red top tubes were transported in Biochemistry and Serology testing area. Samples were centrifuged at 3500 rpm for 5 minutes and cells separated from serum. Serum

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iron levels were quantified using Roche Cobas c311, the fully automated clinical biochemistry analyzer that use principal of random-access, software- controlled system for and photometric analyses intended for quantitative in vitro determinations of a wide variety of tests. Ferritin levels were quantified using Cobase411, the fully automated immunoassay analyzer. Calibration and quality control for iron and ferritin tests were performed according to the standard procedure. 300 microliters of serum were pipetted in 2 sample cups for each participant's sample and put in chemistry and immunoassay analyzers, respectively. Iron and ferritin tests were selected and run on appropriate equipment as instructed in the operating manual. Results were interfaced in the LIS and recorded in the backup hard logbook for post analytical procedures. On the other hand, results were recorded in the Excel sheet of data collection for the study.

2.6.3. *H. pylori* Antigens

H. pylori infection was detected by testing fresh stool sample on *H. pylori* rapid diagnostic test. Small portion of stool of about 2 grams was collected from the primary sample and transferred to the specimen collection tube containing the buffer. The mixture was mix well by gentle shaking and *incubated* for 2 minutes at room temperature. The test cassette was removed from its foil pouch by tearing along the notch. Immediately the cassette was labeled with participant ID number. Two drops of about 80 microliters were squeezed in the sample well of the cassette. The result was read in 10 minutes and not exceeding 15 minutes. The result was recorded in the stool specimen results logbook and in parallel into the study's Excel sheet for data collection. All wastes were discarded according to the safety procedures and biological waste management guidelines.

2.7. Statistical analyses

Data were collected using Microsoft Office Excel and exported to SPSS (Version 25.0) and GraphPad Prism 7.0 Software (GraphPad Software Inc., San Diego, CA, USA) for analysis and a p-value less than 0.05 was considered as statistically significant. Chi-square test was used to assess the significance of the association between different qualitative variables (*H. pylori*, anemia, and IDA). ANOVA and t test were used to compare the means of Hb, MCV and Hct between different groups of participants.

3. RESULTS

3.1. Clinical characteristics of the study participants

The overall size of the study participants was 200 in which 56.5% and 43.5% were female and male, respectively. The mean-age of the participants was 39 (sd. = ±12.9) years old, ranging from 18 to 78 years. The dominant age group was 34-49 years (43.0%), followed by 18-33 (39.5), 50-65 (12.0%). The lowest presented group was participants aged 66 years and above (Table 1).

Results from clinical characteristics showed that the mean value of Hb, MVV and Hct were not significantly different across the age groups of the participants. The difference in mean value of Hb, MCV and Hct was statistically significant across gender ($p < 0.0001$)(Figure 1).

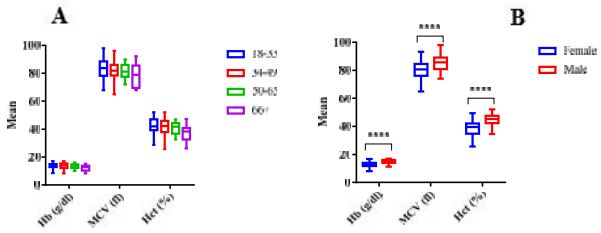

Fig 1: Mean hemoglobin, MCV and hematocrit values according to age (A) and gender (B). (One-way ANOVA test for mean comparison across age groups, and unpaired t test for mean comparison across gender, ****p <0.0001 and the error bars represent ± SEM).

Table 1: Demographic characteristics of participants (n=200).

		Frequency	Percentage (%)
Age at diagnosis		39 (mean)	± 12.9 (sd.)
Age group			
	18-33	79	39.5
	34-49	86	43.0
	50-65	24	12.0
	66+	11	5.5
Gender			
	Female	113	56.5
	Male	87	43.5
	Total	200	100

3.2. Prevalence of H. pylori and anemia among the study participants

This study assessed the prevalence of H. pylori and anemia and their associated clinical parameters. The overall prevalence of anemia and *H. pylori* in the study participants was closely equal (29.5% and 29.0%, respectively). Anemia was much more prevalent in females (47.8%) than in males (5.7%) ($p< 0.001$). According to age groups, anemia was more prevalent in oldest adult participants (54.5%), followed by participants aged 34-49 years (30.2%). Anemia prevalence was closely equal in participants' age group 50-65 and the youngest adult group at proportions of 25.0% and 26.6%, respectively. The prevalence of *H. pylori* in female and males was closely equal (30.1% and 27.6%, respectively), and approximately equal across age groups, ranging from 22.8% to 33.7% (Table 2). A higher prevalence of anemic participants was in elderly subjects with 54.5% in people of 66 years and above (Table 2).

Table 2: Anemia and *H. pylori* infection prevalence according to age and gender

Characteristics	N	H. pylori			Anemia		
		Negative N (%)	Positive N (%)		Negative N (%)	Positive N (%)	
Overall prevalence		142 (71.0)	58 (29.0)		141 (70.5)	59 (29.5)	
Age group							
18-33	79	61 (77.2)	18 (22.8)	0.449	58 (73.4)	21 (26.6)	0.273
34-49	86	57 (66.3)	29 (33.7)		60 (69.8)	26 (30.2)	

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50-65	24	16 (66.7)	8 (33.3)	18 (75.0)	6 (25.0)
66+	11	8 (72.7)	3 (27.3)	5 (45.5)	6 (54.5)
Gender					
Female	113	79 (69.9)	34 (30.1)	0.699	59 (52.2)	54 (47.8)	<0.001*
Male	87	63 (72.4)	24 (27.6)	82 (94.3)	5 (5.7)

***:** statistically significant; N: actual number; %: percentages

3.3. Association between anemia and *H. pylori* infection in the study population

In this study, the association between anemia and *H. pylori* was assessed according to age groups and gender. Anemia was significantly associated to *H. pylori* according to 2 youngest groups, 18-49 years old (p < 001). Regarding *gender*, anemia was significantly associated to *H. pylori* across females and not for males. A significant association was also observed across normal and low (abnormal) iron values of the participants (p = 0.006 and p = 0.030). The association between anemia and *H. pylori* was also statistically significant in the participants with normal ferritin level (p < 001)). Anemia was observed in people with positive *H. pylori* with a significant difference compared to people with negative *H. pylori*, but no significant difference was observed in participant with low ferritin (Table 3).

Table 3: Relationship between anemia and *H. pylori* infection according to age, gender, iron and ferritin levels (n=200)

Characteristics				Anemia prevalence	
	H. pylori	(-) / (+)	N	N (%)	p-value
Age group					
18-33	H. pylori	(-)	61	10 (16.4)	< 0.001*
		(+)	18	11 (61.1)	
34-49	H. pylori	(-)	57	7 (12.3)	< 0.001*
		(+)	29	19 (65.5)	
50-65	H. pylori	(-)	16	3 (18.8)	0.317
		(+)	8	3 (37.5)	
66+	H. pylori	(-)	8	4 (50.0)	0.621
		(+)	3	2 (66.7)	
Gender					
Female	H. pylori	(-)	79	22 (27.8)	< 0.001*
		(+)	34	32 (94.1)	
Male	H. pylori	(-)	63	2 (3.2)	0.095
		(+)	24	3 (12.5)	
Iron					
Normal	H. pylori	(-)	128	10 (7.8)	0.006*
		(+)	17	5 (29.4)	
Low	H. pylori	(-)	14	14 (100.0)	0.030*
		(+)	41	30(73.2)	
Ferritin					
Normal	H. pylori	(-)	137	19 (13.9)	< 0.001*
		(+)	35	15 (42.9)	
Low	H. pylori	(-)	5	5 (100)	0.393
		(+)	23	20 (87.0)	

*: statistically significant; (-): negative; (+): positive; N: actual number, %: percentages

3.4. Association between IDA in *H. pylori* infection in the study population

The association between iron deficiency anemia (IDA) and *H. pylori* was evaluated according to age group and gender. The association between IDA and *H. pylori* was statistically significant across young adults (18-33years) (p < 0.001). For young age group, among 61 subjects were not infected by*H. pylori* only 1 had IDA, whereas in 18 infected ones 7 were IDA positive. Similar proportions were observed in participants aged 34-49, where among 57 *H. pylori* negative subjects only 2 were IDA positive while in 29 *H. pylori*-infected participant more than half were IDA positive (65.5%). For the other age groups (50-65 and ≥ 66 years), the association was not statistically significant(p = 0.439; p = 0.425).According to gender, majority *H. pylori*-infected subject were IDA positive (79.5%), while only 8.2% were IDA positive among negative *H. pylori*. The proportions of IDA positive in negatively diagnosed participants for *H. pylori* were very low (6.3% and 1.6%) for both female and males, respectively. The association between IDA and *H. pylori* was statistically significant in females (p < 0.001) and not in males (p = 0.123) (Table 4).

Table 4: Relationship between iron deficiency anemia and H. pylori infection, according to age and gender.

Characteristics				Iron deficiency anemia	
	H. pylori	(-) / (+)	**N**	**N (%)**	**p-value**
Age group					
18-33	H. pylori	(-)	61	1 (1.6)	< 0.001*
		(+)	18	7 (38.9)	
34-49	H. pylori	(-)	57	2 (3.5)	< 0.001*
		(+)	29	19 (65.5)	
50-65	H. pylori	(-)	16	2 (12.5)	0.439
		(+)	8	2 (25.0)	
66+	H. pylori	(-)	8	1 (12.5)	0.425
		(+)	3	1 (33.3)	
Gender					
Female	H. pylori	(-)	79	5 (6.3)	< 0.001*
		(+)	34	27 (79.4)	
Male	H. pylori	(-)	63	1 (1.6)	0.123
		(+)	24	2 (8.3)	

***:** statistically significant; (-): negative; (+): positive; N: actual number; %: percentages

4. DISCUSSIONS

In this study, anemia was confirmed based in participants with low level of Hb, less than 12 g/dl for females and less than 13 g/dl for males, the levels defined more than 50 years ago by the World Health Organization (WHO).[18] The latter also defined anemia with cutoff value as an Hb concentration less than 12 g/dl in women and less than 13 g/dl in men. Anemia is grouped as mild when patient has Hb of between 11 and 11.9 g/dl in women and 11 to 12.9 g/dl in men, as moderate with values of 8 to 10.9 g/dl in both women and men, and as severe when Hb levels are less than 8 g/dl in both women and men.[19] Participants experienced 33.0% of both microcytic and hypochromic anemia and the rest continued to normocytic and normochromicprofiles.MCV values less than 80 fL are classified as microcytic anemia, normocytic when they range from 80 to 100 fL and macrocytic with values greater than 100 fL. [20] The abnormal or low MCV is commonly

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observed in several conditions, including chronic iron-deficiency anemia [21]. In this study, MCV was used to confirm IDA in the study participants in addition to Hb level. Bryant *et al*. reported iron deficiency in apheresis donors with low MCV and normal Hb level.[22] This showed that not all cases of iron deficiency lead to anemia, other factors may come into action. This situation was prolonged in participants with advanced age compared to young adults. Loss or malabsorption of iron could result from chronic gastrointestinal diseases [23] which come from alteration of microbiota in elderly.[24]The decrease to below lower reference limits in iron stores and hematological values especially hemoglobin and MCV defined the IDA in other studies [25].

In this study, the prevalence of *H. pylori* was slightly equal in all age groups. This is in contrast to the findings reported by Forman *et al*. when assessing the rates of *H. pylori* in accordance to age and social class, showing an increase of *H. pylori* cases with increase in age.[26] The prevalence of *H. pylori* was approximately 30% which is quietly lower than 56.9% reported by Forman *et al*. [26] and the globally estimated prevalence (50%). [27] According to gender, the prevalence of *H. pylori*was slightly different between females and males (30.1% and 27.6%, respectively). Similarly, in an epidemiological study conducted in Romania to characterize *H. pylori* in children, females were 32.8% whereas males were 26.5% of diagnosed subjects in each gender, respectively.[28] Contrary, a study conducted in non-elderly people (≤ 65) to assess the association between *H. pylori* and decline in iron stores, reported a higher prevalence in both females and males (74.3% and 61.9%) [29].

Basing on the IDA of 50% inthe group of people infected with *H. pylori* and 4% of IDA in the group of subjects un infected with *H. pylori*, the study showed that there is a higher risk of having anemia while you are infected by *H. pylori* compared to individuals not infected. The association of *H. pylori* infections to IDA predicted11.9 times the risk of having IDA while you are not infected with *H. pylori*. A study in Ethiopia showed that there is a great risk of being anemic due to infection by *H. pylori* [30] which agreed with similar studies in China [31, 32] that reported 2.53 times increased risk of being anemic when infected by *H. pylori* compared to *H. pylori* – negative group.

5. CONCLUSION

The prevalence of *Helicobacter pylori* infection among participants of this study was high. There was a significant difference of iron decrease in stores among participants infected with *H. pylori* compared to negative *H. pylori* participants. The study showed a great risk of experiencing insufficient iron levels and iron associated anemia in group of people infected by *H. pylori* compared to non-infected people. Long-term, and increased sample size at a nationwide population-based studies, are advised to investigate the IDA and other possible risk factors associated with *H. pylori* infection.

6. REFERENCES

1. de Lacy Clancy C. Helicobacter pylori: comparative genomics and structure-function analysis of the flagellum biogenesis protein HP0958 [Doctoral thesis]. Ireland: University College Cork; 2014.
2. Marshall BJ, Warren JR. Unidentified curved bacilli in the stomach of patients with gastritis and peptic ulceration. Lancet. 1984;1(8390):1311-5.
3. Serag M. Molecular Detection of Metronidazole Resistant Genes (RdxA and FrxA) of Helicobacter pylori Isolated from Stool Specimen at Alinjaz Diagnostic Center in Khartoum State 2019.
4. Malaty HM. Epidemiology of Helicobacter pylori infection. Best Pract Res Clin Gastroenterol. 2007;21(2):205-14.
5. Burucoa C, Axon A. Epidemiology of Helicobacter pylori infection. Helicobacter. 2017;22 Suppl 1.
6. Kabakambira JD, Hategeka C, Page C, Ntirenganya C, Dusabejambo V, Ndoli J, et al. Efficacy of Helicobacter pylori eradication regimens in Rwanda: a randomized controlled trial. BMC Gastroenterol. 2018;18(1):134.
7. Tympa-Psirropoulou E, Vagenas C, Psirropoulos D, Dafni O, Matala A, Skopouli F. Nutritional risk factors for iron-deficiency anaemia in children 12-24 months old in the area of Thessalia in Greece. Int J Food Sci Nutr. 2005;56(1):1-12.
8. Bouri S, Martin J. Investigation of iron deficiency anaemia Clin Med (Lond). 2018;18(3):242-4.
9. Khan A, Khan WM, Ayub M, Humayun M, Haroon M. Ferritin Is a Marker of Inflammation rather than Iron Deficiency in Overweight and Obese People. J Obes. 2016;2016:1937320.
10. 10. Bermejo F, García-López S. A guide to diagnosis of iron deficiency and iron deficiency anemia in digestive diseases. World J Gastroenterol. 2009;15(37):4638-43.
11. Miller CJ, Dunn EV, Abdouni SF, Shaheen HM, Ullah MS. Factors associated with iron depletion and iron deficiency anemia among Arabic preschool children of the United Arab Emirates. Saudi Med J. 2004;25(7):843-7.
12. Mwangi MN, Phiri KS, Abkari A, Gbané M, Bourdet-Sicard R, Braesco VA, et al. Iron for Africa-Report of an Expert Workshop. Nutrients. 2017;9(6).
13. Simbauranga RH, Kamugisha E, Hokororo A, Kidenya BR, Makani J. Prevalence and factors associated with severe anaemia amongst under-five children hospitalized at Bugando Medical Centre, Mwanza, Tanzania. BMC Hematology. 2015;15(1):13.
14. Nguyen ND, Allen JR, Peat JK, Beal P, Webster BH, Gaskin KJ. Iron status of young Vietnamese children in Australia. J Paediatr Child Health. 2004;40(8):424-9.

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15. Johnson DC, Dean DR, Smith AD, Johnson MK. Structure, function, and formation of biological iron-sulfur clusters. Annu Rev Biochem. 2005;74:247-81.
16. Muhsen K, Cohen D. Helicobacter pylori infection and iron stores: a systematic review and meta-analysis. Helicobacter. 2008;13(5):323-40.
17. DuBois S, Kearney DJ. Iron-deficiency anemia and Helicobacter pylori infection: a review of the evidence. Am J Gastroenterol. 2005;100(2):453-9.
18. Nutritional anaemias. Report of a WHO scientific group. World Health Organ Tech Rep Ser. 1968;405:5-37.
19. Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity. Vitamin and Mineral Nutrition Information System [press release]. Geneva, Switzerland: World Health Organization2011.
20. Moreno Chulilla JA, Romero Colás MS, Gutiérrez Martín M. Classification of anemia for gastroenterologists. World J Gastroenterol. 2009;15(37):4627-37.
21. Maner BS, Moosavi L. Mean Corpuscular Volume. [Updated 2022 Jul 4]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2023. Available online: https://www.ncbi.nlm.nih.gov/books/NBK545275/ (Accessed on 22 May, 2023). 2022.
22. Bryant BJ, Hopkins JA, Arceo SM, Leitman SF. Evaluation of low red blood cell mean corpuscular volume in an apheresis donor population. Transfusion. 2009;49(9):1971-6.
23. Ngum R, Grigos A, Daoud M. Iron-Deficiency Anemia in an Elderly Caucasian Female: An Unusual Colonoscopic Finding. Cureus. 2021;13(11):e19564.
24. De Vuyst L, Leroy F. Cross-feeding between bifidobacteria and butyrate-producing colon bacteria explains bifdobacterial competitiveness, butyrate production, and gas production. Int J Food Microbiol. 2011;149(1):73-80.
25. Camaschella C. Iron-deficiency anemia. N Engl J Med. 2015;372(19):1832-43.
26. Sitas F, Forman D, Yarnell JW, Burr ML, Elwood PC, Pedley S, et al. Helicobacter pylori infection rates in relation to age and social class in a population of Welsh men. Gut. 1991;32(1):25-8.
27. Suerbaum S, Michetti P. Helicobacter pylori infection. N Engl J Med. 2002;347(15):1175-86.
28. Lupu A, Miron IC, Cernomaz AT, Gavrilovici C, Lupu VV, Starcea IM, et al. Epidemiological Characteristics of Helicobacter pylori Infection in Children in Northeast Romania. Diagnostics (Basel). 2023;13(3).
29. Lee JY, Kim SE, Park SJ, Park MI, Moon W, Kim JH, et al. Helicobacter pylori infection and iron deficiency in non-elderly adults participating in a health check-up program. Korean J Intern Med. 2022;37(2):304-12.
30. Kibru D, Gelaw B, Alemu A, Addis Z. Helicobacter pylori infection and its association with anemia among adult dyspeptic patients attending Butajira Hospital, Ethiopia. BMC Infect Dis. 2014;14:656.
31. Xu MY, Cao B, Yuan BS, Yin J, Liu L, Lu QB. Association of anaemia with Helicobacter pylori infection: a retrospective study. Sci Rep. 2017;7(1):13434.
32. Hou B, Zhang M, Liu M, Dai W, Lin Y, Li Y, et al. Association of active Helicobacter pylori infection and anemia in elderly males. BMC Infectious Diseases. 2019;19(1):228.

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