Internal Medicine
The Japanese Society of Internal Medicine
doi: 10.2169/internalmedicine.5132-20
Intern Med 59: 2817-2823, 2020
http://internmed.jp

【 ORIGINAL ARTICLE 】

Serum Anti-*Helicobacter pylori* IgG Antibody Titer in *H. pylori*-negative Cases with a Different Gastric Mucosal Atrophy Status

Kyoichi Adachi[1], Kanako Kishi[1], Takumi Notsu[1], Tomoko Mishiro[1], Kazunari Sota[1], Norihisa Ishimura[2] and Shunji Ishihara[2]

Abstract:

Objective This retrospective study was performed to investigate the anti-*Helicobacter pylori* IgG antibody serum titers in *H. pylori*-negative subjects with different degrees of gastric mucosal atrophy including C0 grade atrophy.

Methods The absence of *H. pylori* infection was determined based on both negative serum anti-*H. pylori* IgG antibody test findings and no endoscopic evidence of that infection. Cases negative for the antibody and with positive endoscopic findings of *H. pylori* infection were defined as *H. pylori*-positive. The serum anti-*H. pylori* IgG antibody titers were analyzed in *H. pylori*-negative (n=1,087), -positive (n=69), and post-eradicated (n=278) subjects.

Results The serum antibody titer in subjects with *H. pylori*-positive endoscopy findings was significantly higher than that in *H. pylori*-negative subjects, even when the serum titer indicated a negative result. In addition, the anti-*H. pylori* IgG antibody serum titer was higher in *H. pylori*-negative subjects with a greater degree of gastric mucosal atrophy. In a comparison between *H. pylori*-negative C0 and C1 gastric mucosal atrophy cases, the antibody serum titer in those classified as C0 was significantly lower. An analysis of *H. pylori* post-eradicated cases showed that the serum antibody titer decreased over time after successful eradication.

Conclusion The disappearance of *H. pylori* infection in *H. pylori*-negative individuals may occur later in those with a greater degree of gastric mucosal atrophy. The serum antibody titer difference between the *H. pylori*-negative C0 and C1 groups might have been caused by the differences in distribution between *H. pylori*-uninfected subjects and those in whom the infection had disappeared, thus additional investigation is needed to clarify the significance of gastric mucosal classification including the C0 grade.

Key words: *Helicobacter pylori*, serum antibody titer, gastric mucosal atrophy, C0 grade

(Intern Med 59: 2817-2823, 2020)
(DOI: 10.2169/internalmedicine.5132-20)

Introduction

A long-term *Helicobacter pylori* infection is the most important factor for the development of gastric mucosal atrophy (1-3). However, some individuals show apparent endoscopic gastric mucosal atrophy without that infection, even though they have not undergone eradication therapy (4, 5). This condition is considered to be caused by the spontaneous disappearance of *H. pylori* or the usage of an antibiotics for another condition (6-9). *H. pylori* infection generally initiates in childhood, while the degree of gastric mucosal atrophy gradually increases with age (10, 11). Therefore, the timing of the disappearance of that infection is expected to have an effect on degree of gastric mucosal atrophy in *H. pylori*-negative individuals. Since the serum titer of the anti-*H. pylori* IgG antibody has been reported to decrease after the disappearance of infection by eradication ther-

[1]Health Center, Shimane Environment and Health Public Corporation, Japan and [2]Second Department of Internal Medicine, Shimane University Faculty of Medicine, Japan
Received: April 20, 2020; Accepted: June 3, 2020; Advance Publication by J-STAGE: July 21, 2020
Correspondence to Dr. Kyoichi Adachi, adachi@kanhokou.or.jp

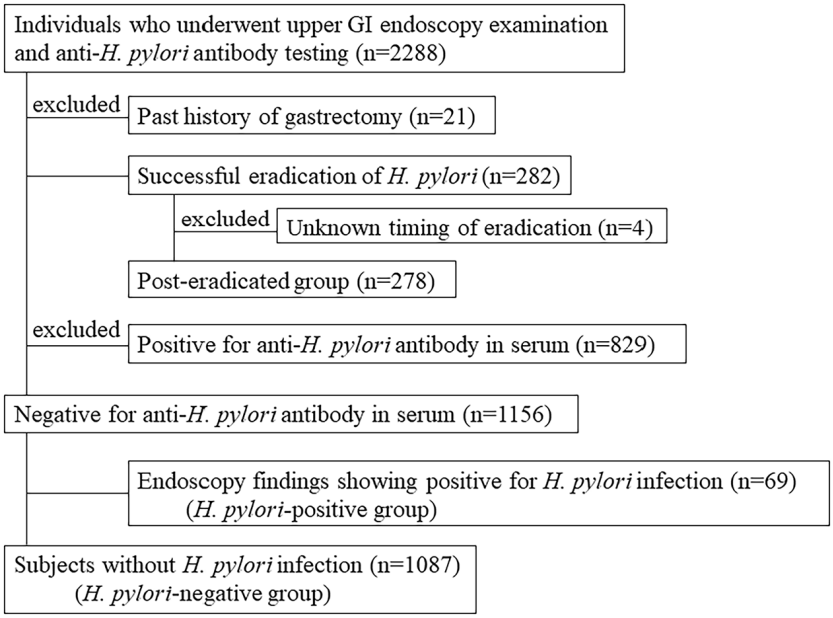

Figure 1. Flow diagram of the subject selection process.

apy (12, 13), the timing of infection disappearance may also affect that serum titer. However, the relationship between gastric mucosal atrophy degree and anti-*H. pylori* IgG antibody serum titer has not yet been investigated in *H. pylori*-negative subjects who have no history of eradication therapy.

Endoscopic gastric mucosal atrophy classification was developed by Kimura and Takemoto, and it is based on 6 grades (C1, C2, C3, O1, O2, O3) (14). In addition, cases without gastric mucosal atrophy have recently been referred to as C0 grade (4), thus the classification of gastric mucosal atrophy can be divided into 7 groups, including the C0 grade and those noted above. To the best of our knowledge, the significance of endoscopic gastric mucosal classification that includes the C0 grade has not yet been determined. We conducted this retrospective study to clarify the relationship between serum titer of the anti-*H. pylori* IgG antibody and degree of gastric mucosal atrophy in subjects without *H. pylori* infection, as well as the significance of that classification with C0 grade included.

Materials and Methods

The subjects were selected from individuals who visited the Health Center of Shimane Environment and Health Public Corporation for a detailed medical checkup examination between June 2016 and November 2019, as we have been evaluating patients based on the endoscopic gastric mucosal atrophic classification including C0 grade since June 2016. The majority were socially active and productive, and considered to be socioeconomically middle class. During the study period, a total of 2,288 individuals were simultaneously assessed by both an upper GI endoscopic examination and serum anti-*H. pylori* IgG antibody test. Those with a history of gastric surgery were excluded from the present analysis. Of the 282 individuals with an *H. pylori* post-eradication status, 4 were excluded prior to the analysis, because the timing of eradication could not be determined. Thus, the number of post-eradicated cases subjected to analyze was 278 (post-eradicated group: 184 males, 94 females; mean age 55.0 years). Serum anti-*H. pylori* IgG antibody detection was performed using the SphereLight *H. pylori* antibody J® kit (FUJIFILM Wako Pure Chemical, Osaka, Japan), originally developed as a qualitative test for the diagnosis of *H. pylori* infection, and the antibody titer was automatically determined using a chemiluminescent enzyme immunoassay method, with a value ≥4.0 U/mL defined as positive, according to the manufacturer's instructions (4, 15). The presence or absence of *H. pylori* infection, and the result of eradication therapy were also confirmed based on findings obtained in an upper GI endoscopic examination (16-20). In order to examine the serum titer of *H. pylori*-negative subjects, we excluded those found positive in a test for the anti-*H. pylori* antibody in serum. Sixty-nine subjects negative for the anti-*H. pylori* IgG antibody were placed in the *H. pylori*-positive group (47 males, 22 females; mean age 48.8 years), as they had endoscopic findings indicating the *H. pylori*-positive status, such as nodular gastritis, spotty and/or diffuse redness of fundic gland mucosa, or sticky mucous (16-20). The remaining 1,087 (606 males, 481 females; mean age 49.5 years) were placed in the *H. pylori*-negative group. The total number of subjects in this study was 1,434 (837 males, 597 females; mean age 50.5 years) (Fig. 1).

All upper GI endoscopic examinations were performed by experienced licensed endoscopists using an EG-L580NW endoscope (Fujifilm, Tokyo, Japan). At our institution, an upper GI endoscopic examination is performed with the subject in an unsedated condition without anti-cholinergic drug administration, with the endoscope typically inserted in a transnasal manner. When gastric mucosal atrophy was endo-

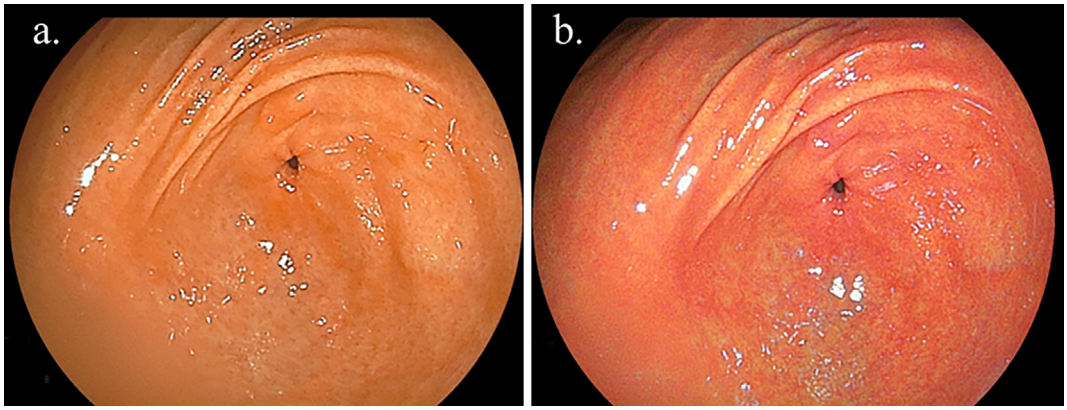

Figure 2. **Representative endoscopic images showing C0 grade gastric mucosal atrophy. No atrophic mucosal area can be seen in the antrum with ordinary imaging (a) or linked color imaging (LCI) (b).**

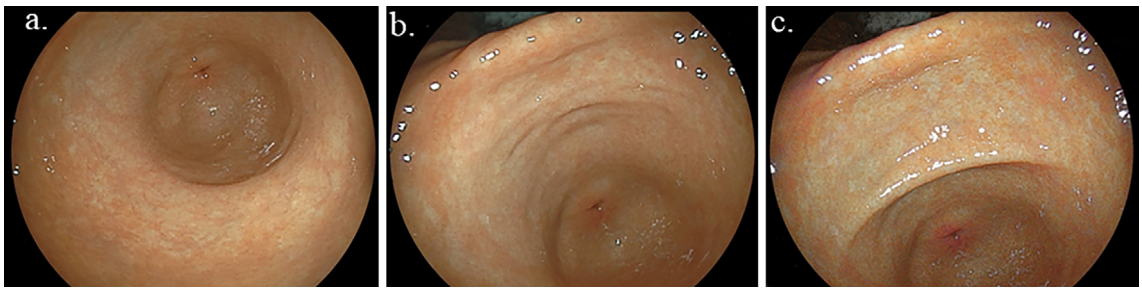

Figure 3. **Representative endoscopic findings demonstrating C1 grade gastric mucosal atrophy. An atrophic border is shown as differences in color in the antrum by ordinary imaging (a, b) and linked color imaging (LCI) (c). The atrophic area and border were easily recognized and visualized by LCI.**

scopically observed, the degree was evaluated using the classification of Kimura and Takemoto, in which gastric mucosal atrophy is classified into 6 grades (C1, C2, C3, O1, O2, O3) (14). The degree of gastric mucosal atrophy was carefully determined by recognition of an atrophic border. Cases without gastric mucosal atrophy were determined to be C0 for this study. Therefore, subjects with C1 atrophy according to the original Kimura and Takemoto classification were classified as C0 or C1 (Fig. 2, 3), while gastric mucosal atrophy was classified into 7 grades, including C0 and the others noted above. All endoscopic images from each subject were simultaneously reviewed by three expert endoscopists to determine the degree of gastric mucosal atrophy and the absence of endoscopic findings indicating positive for *H. pylori* infection was confirmed by consensus.

The serum anti-*H. pylori* IgG antibody titers in *H. pylori*-negative subjects with different degrees of gastric mucosal atrophy were analyzed and compared. The subjects with gastric mucosal atrophy graded from C3-O3 were combined into a single group for the present analysis, since the number with C3, O1, O2 and O3 atrophy was small (19, 3, 6, 13, respectively). In addition, the serum antibody titers were analyzed in subjects with *H. pylori*-positive endoscopic findings. In post-eradicated subjects, the serum antibody titers were compared after dividing them based on the duration after eradication to examine the relationship between the degree of gastric mucosal atrophy and age at time of successful eradication.

Statistical analyses were performed using a chi-squared test, Mann-Whitney U test, or the Kruskal-Wallis test, as appropriate. All calculations were done using the StatView 5.0 software program for Macintosh (Abacus Concepts, Berkeley, USA), with a p level <0.05 considered to indicate statistical significance.

This study was performed in accordance with the Declaration of Helsinki, and the protocol was approved by the ethics committee of the Shimane Environment and Health Public Corporation. Written informed consent indicating that obtained clinical data would be used for a clinical study without release of individual information was received from all subjects before performing the medical checkup examinations.

Results

The characteristics of the present *H. pylori*-negative and -positive subjects are shown in Table 1. The numbers of negative subjects with C0, C1, C2, and C3-O3 grade gastric mucosal atrophy were 716, 172, 158 and 41, respectively. Further analysis showed a female predominance in those

Table 1. Characteristics of the *H. Pylori*-negative and -positive Subjects.

Gastric mucosal atrophy grade	*H. pylori*-negative (n=1,087)				*H. pylori*-positive (n=69)
	C0 (n=716)	C1 (n=172)	C2 (n=158)	C3-O3 (n=41)	
Gender, male/female	420/296	95/77	68/90#1, 2	23/18#3	47/22#3
Age, years	49.3±0.4	47.6±0.8#1	49.1±0.8	62.0±1.4#1, 2, 3	48.8±1.3#4
Serum titer of anti-*H. pylori* IgG antibody (U/mL)	0.81±0.02	0.99±0.06#1	1.07±0.07#1	1.54±0.13#1, 2, 3	2.63±0.12#1, 2, 3, 4
≤0.9	526 (73.5%)	112 (65.1%)	88 (55.7%)	13 (31.7%)	5 (7.3%)
1.0-1.9	152 (21.2%)	40 (23.3%)	47 (29.7%)	16 (39.0%)	12 (17.4%)
2.0-2.9	25 (3.5%)	15 (8.7%)	18 (11.4%)	10 (24.4%)	21 (30.4%)
3.0-3.9	13 (1.8%)	7 (2.9%)	5 (3.2%)	2 (4.9%)	31 (44.9%)

H. pylori-negative: negative in anti-*H. pylori* antibody test results and absence of *H. pylori* infection in endoscopic findings. *H. pylori*-positive: negative in anti-*H. pylori* antibody test results and positive for *H. pylori* infection in endoscopic findings. Values are expressed as the mean±SE or number of subjects. Gastric mucosal atrophy was evaluated using the modified classification of Kimura and Takemoto. #1, 2, 3, 4: significantly different in comparison to subjects with C0, C1, C2, C3-O3 grade, respectively.

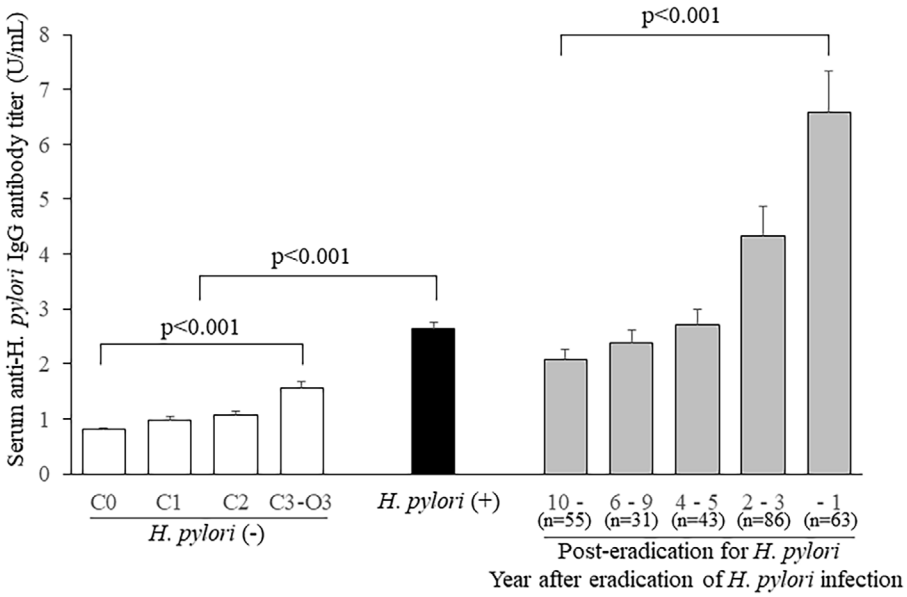

Figure 4. The serum titers of anti-*H. pylori* IgG antibody in *H. pylori*-negative, -positive, and post-eradicated cases. *H. pylori*-negative: negative in anti-*H. pylori* antibody test results and absence of *H. pylori* infection in endoscopic findings. *H. pylori*-positive: negative in anti-*H. pylori* antibody test results and positive for *H. pylori* infection in endoscopic findings. The numbers of *H. pylori*-negative, -positive, and post-eradicated subjects were 1,087, 69, and 278, respectively. Values are expressed as the mean±SE.

with C2 atrophy, while those in the C3-O3 group were older in comparison to the others.

The anti-*H. pylori* IgG antibody serum titer was greater in *H. pylori*-negative subjects with a higher degree of gastric mucosal atrophy. The serum titer levels were divided into four subgroups (<0.9, 1.0-1.9, 2.0-2.9, 3.0-3.9 U/mL) and analysis in *H. pylori*-negative subjects showed that the anti-*H. pylori* IgG antibody titer was significantly different among the gastric mucosal atrophy grades (p<0.001). Furthermore, when the anti-*H. pylori* IgG antibody titer was examined in *H. pylori*-negative subjects with C0 and C1 gastric mucosal atrophy, it was found to be significantly lower in subjects with the C0 grade, while the distribution of the serum titers was also significantly different between C0 and C1 (p=0.013). The anti-*H. pylori* IgG antibody titer in *H. pylori*-positive subjects was significantly higher than that in the *H. pylori*-negative subjects, while the antibody titer was also more largely distributed from 2.0-3.9 in the positive group (Table 1, Fig. 4).

The further analyze the anti-*H. pylori* IgG antibody serum titer, *H. pylori*-negative subjects were divided based on gender, and older age and higher level of serum titer were noted in both males and females with a higher degree of gastric mucosal atrophy. Furthermore, the distribution of the anti-*H. pylori* IgG antibody titer was significantly different among the gastric mucosal atrophy grades in both genders (p< 0.001). Also, a comparison between the C0 and C1 gastric mucosal atrophy groups showed that the anti-*H. pylori* IgG

Table 2. Characteristics of the Subjects without *H. Pylori* Infection Divided by Gender.

Gastric mucosal atrophy grade	C0	C1	C2	C3-O3
Male				
Age, years	50.0±0.5	48.2±1.1	50.0±1.2	63.2±1.9#1, 2, 3
Serum titer of anti-*H. pylori* IgG antibody (U/mL)	0.77±0.03	1.01±0.08#1	1.21±0.11#1	1.38±0.16#1, 2
≤0.9	324 (77.1%)	61 (64.2%)	32 (47.1%)	9 (39.1%)
1.0-1.9	77 (18.3%)	20 (21.1%)	23 (33.8%)	9 (39.1%)
2.0-2.9	12 (2.9%)	11 (11.6%)	9 (13.2%)	4 (17.4%)
3.0-3.9	7 (1.7%)	3 (3.2%)	4 (5.9%)	1 (4.3%)
Female				
Age, years	48.3±0.6	46.9±1.2	48.5±1.0	60.4±2.1#1, 2, 3
Serum titer of anti-*H. pylori* IgG antibody, U/mL)	0.87±0.04	0.95±0.08#1	0.95±0.08	1.77±0.22#1, 2, 3
≤0.9	202 (68.2%)	51 (66.2%)	56 (62.2%)	4 (22.2%)
1.0-1.9	75 (25.3%)	20 (26.0%)	24 (26.7%)	7 (38.9%)
2.0-2.9	13 (4.4%)	4 (5.2%)	9 (10.0%)	6 (33.3%)
3.0-3.9	6 (2.0%)	2 (2.6%)	1 (1.1%)	1 (5.6%)

Data are expressed as the mean±SE or number of subjects. Gastric mucosal atrophy was evaluated using the modified classification of Kimura and Takemoto. #1, 2, 3: significantly different in comparison to subjects with C0, C1, C2 grade, respectively.

Table 3. Relationship between the Age at Time of Eradication and the Degree of Gastric Mucosal Atrophy in *H. Pylori* Post-eradicated Cases.

	Degree of gastric mucosal atrophy (p<0.001)					
Age (years)	C1	C2	C3	O1	O2	O3
≤39 (n=44)	8 (18.2%)	24 (54.5%)	11 (25.0%)	1 (2.3%)	0	0
40-49 (n=97)	14 (14.4%)	37 (38.1%)	35 (36.1%)	8 (8.2%)	3 (3.1%)	0
50-59 (n=90)	8 (8.9%)	35 (38.9%)	25 (27.8%)	12 (13.3%)	5 (5.6%)	5 (5.6%)
≥60 (n=47)	1 (2.1%)	13 (27.7%)	8 (17.0%)	7 (14.9%)	9 (19.1%)	9 (19.1%)

Data are expressed as the number of subjects, unless otherwise noted. Gastric mucosal atrophy was evaluated using the classification of Kimura and Takemoto.

antibody titer tended to be lower in both males and females with C0 grade, with a similar tendency observed in the distribution of serum titers between the genders (Table 2).

An analysis of the anti-*H. pylori* IgG antibody serum titer in the *H. pylori*-eradicated group showed that it decreased for a longer duration after successful eradication (Fig. 4). In addition, the degree of gastric mucosal atrophy when successful eradication was achieved was significantly lower in younger subjects (Table 3).

Discussion

In this study, we determined the anti-*H. pylori* IgG antibody titer in serum obtained from *H. pylori*-negative, -positive, and post-eradicated subjects. An analysis of *H. pylori*-negative subjects found a higher titer in those with a higher degree of gastric mucosal atrophy, while comparisons between subjects with C0 and C1 gastric mucosal atrophy also revealed a significant difference regarding that antibody titer. The timing of the disappearance of *H. pylori* infection is considered to affect not only the degree of gastric mucosal atrophy but also serum anti-*H. pylori* IgG antibody titer. The progression of gastric mucosal atrophy induced by such infection discontinues after successful eradication of the bacterium. Thus, the degree of gastric mucosal atrophy is considered to reflect that at the time of disappearance of infection, though improvement may actually occur quite slowly over a long period. Indeed, findings for the *H. pylori*-eradicated group demonstrated that the degree of gastric mucosal atrophy was significantly lower in those who were younger at the time of successful eradication. Other studies have noted that the anti-*H. pylori* IgG antibody titer in the serum was reduced after successful eradication (6, 7), while the present results also demonstrated that the titer in *H. pylori*-eradicated subjects had a greater decrease in those with a longer duration following eradication. Thus, a reduced titer in individuals with a milder degree of gastric mucosal atrophy is considered to be related to a long duration after the disappearance of infection. As a result, the disappearance of *H. pylori* infection in *H. pylori*-negative patients with a greater degree of gastric mucosal atrophy may occur later, though that timing could not to be determined in this study.

H. pylori-negative subjects did not have a current *H. pylori* infection during the test period, but they consisted of two types; those who had never been infected and those who had a previous infection that had disappeared prior to our examination. The disappearance of *H. pylori* can be

caused by targeted eradication therapy, a spontaneous disappearance, or the usage of antibiotics for another disease condition (6-9). A spontaneous disappearance is considered to occur in individuals with severe gastric mucosal atrophy, as the bacterial organisms cannot survive in the mucosa of intestinal metaplasia induced with progression of the disease. Among the present study subjects without *H. pylori* infection, only 13 had an O3 grade of gastric mucosal atrophy, while the 186 with a grade ranging from C2-O2 showed a non-atrophic fundic gland area in endoscopic findings, in which *H. pylori* can survive. Thus, an *H. pylori*-negative status in individuals with gastric mucosal atrophy is likely not caused by disease progression, with the usage of antibiotics for another condition considered to be the main reason for bacterial negativity.

The present subjects without *H. pylori* infection were divided into those who had never been infected (uninfected), and those with an infection that occurred during childhood and then later disappeared for some reason (disappeared). Interestingly, the values for both the titer and distribution of the anti-*H. pylori* IgG antibody in serum were significantly different between the C0 and C1 groups in the uninfected subjects. In the subjects with C0 atrophy, the serum titer was lower and those subjects were also more predominantly represented in the group with a lower serum titer of the antibody in comparison with the C1 group. These findings suggest that the distribution of *H. pylori*-uninfected and disappeared subjects is different when divided between the C0 and C1 atrophy status.

The present *H. pylori* negative subjects with C3-O3 atrophy were older than those with milder atrophy. Patients with C3-O3 atrophy are considered to have been previously infected by *H. pylori* for a long period and its disappearance likely occurred later in comparison to those with a milder degree of gastric mucosal atrophy. A female predominance was also noted in the C2 atrophy group in comparison to the others, though our findings could not clearly explain this phenomenon. However, the values for the titer and distribution of the anti-*H. pylori* IgG antibody were not different between males and females without *H. pylori* infection. Gender difference in regard to the progression of gastric mucosal atrophy during *H. pylori* infection might have been a factor, since the events correlated with disappearance of *H. pylori* infection such as the usage of antibiotics for another condition were considered to be similar in the female and male subjects.

In this study, 69 (6.0%) of 1,156 subjects negative for the anti-*H. pylori* IgG antibody were diagnosed as being positive for *H. pylori* infection. Testing was done with the SphereLight *H. pylori* antibody J® test, which has been demonstrated to have a high level of sensitivity (4, 15). We noted that the anti-*H. pylori* IgG antibody titer in the *H. pylori*-positive subjects was significantly higher than that in the negative subjects, and their results were more largely distributed near the cut-off value of this test. Endoscopic *H. pylori*-positive findings, including nodular gastritis, spotty and/or diffuse redness of fundic gland mucosa, and sticky mucous, were carefully reviewed simultaneously by three endoscopy experts. Although *H. pylori* infection was not examined by other diagnostic methods, the higher anti-*H. pylori* IgG antibody titer in subjects with endoscopic findings indicating positive for *H. pylori* infection is considered to indicate the importance of a detailed endoscopic examination in the cases found to be negative in a serum antibody test. Recently, patients with a non-*Helicobacter pylori Helicobacter* (NHPH) infection have been reported to have endoscopic findings of nodular gastritis and a white marbled appearance, which appeared to demonstrate atrophic change (21, 22). Therefore, some subjects in the present *H. pylori*-positive group may have had an NHPH infection. Additional investigations are needed to clarify the influence of NHPH infection on the endoscopic diagnostic findings indicating positive for *H. pylori* infection.

The present study is associated with several limitations. It was not performed in a population-based manner, as the subjects initially visited our center for a medical check-up. Also, the investigation was performed in a cross-sectional manner, thus we were unable to determine events that might have caused disappearance of *H. pylori* infection in the subjects. Furthermore, we did not perform histological examinations of the gastric mucosa samples obtained from the subjects, since the endoscopic examinations in this study were performed as part of a medical checkup. Thus, we could not obtain histological findings regarding the presence of NHPH, nor showing non-atrophic fundic or pyloric gland areas. In addition, we used the results from a single type of anti-*H. pylori* IgG antibody test and the results of different tests might not be identical to those found in this study. The present results should be confirmed by a future large-scale study that includes histological examinations and other serological testing methods in addition to the SphereLight *H. pylori* antibody J® test.

In conclusion, the anti-*H. pylori* IgG antibody titer in serum in *H. pylori*-negative subjects with a higher degree of gastric mucosal atrophy was higher in comparison to those with a lower degree. We concluded that a later disappearance of *H. pylori* infection may occur in patients with a high degree of gastric mucosal atrophy. Additionally, a lower serum titer was seen in *H. pylori*-negative subjects with C0 atrophy and those subjects were more predominantly represented in the group with a lower serum titer of the antibody. Thus, the distribution of subjects classified as *H. pylori*-uninfected and disappeared may be different between those with a C0 and C1 atrophy status, and additional investigation is needed to clarify the significance of the gastric mucosal classification including the C0 grade.

The authors state that they have no Conflict of Interest (COI).

Acknowledgement

We wish to thank Kiyoe Kawase, Marie Ishida, Yuki Funaki, Noriko Yamauchi, and Naoyuki Notsu of the Shimane Environ-

ment and Health Public Corporation, as well as Keiko Masuzaki of the Second Department of Internal Medicine, Shimane University Faculty of Medicine, for their helpful technical support.

References

1. Correa P. Human gastric carcinogenesis: a multistep and multifactorial process--First American Cancer Society Award Lecture on Cancer Epidemiology and Prevention. Cancer Res **52**: 6735-6740, 1992.
2. Kuipers EJ, Uyterlinde AM, Peña AS, et al. Long-term sequelae of *Helicobacter pylori* gastritis. Lancet **345**: 1525-1528, 1998.
3. Valle J, Kekki M, Sipponen P, Ihamäki T, Siurala M. Long-term course and consequences of *Helicobacter pylori* gastritis. Results of a 32-year follow-up study. Scand J Gastroenterol **31**: 546-550, 1996.
4. Adachi K, Mishiro T, Tanaka S, Kinoshita Y. Analysis of negative result in serum anti-*H. pylori* IgG antibody test in cases with gastric mucosal atrophy. J Clin Biochem Nutr **59**: 145-148, 2016.
5. Toyoshima O, Nishizawa T, Arita M, et al. *Helicobacter pylori* infection in subjects negative for high titer serum antibody. World J Gastroenterol **24**: 1419-1428, 2018.
6. Kokkola A, Kosunen TU, Puolakkainen P, et al. Spontaneous disappearance of *Helicobacter pylori* antibodies in patients with advanced atrophic corpus gastritis. APMIS **111**: 619-624, 2003.
7. Ono S, Kato M, Suzuki M, et al. Frequency of *Helicobacter pylori*-negative gastric cancer and gastric mucosal atrophy in a Japanese endoscopic submucosal dissection series including histological, endoscopic and serological atrophy. Digestion **86**: 59-65, 2012.
8. Hiyama T, Quach DT, Le QD, et al. Rate of unintended *Helicobacter pylori* eradication in the Vietnamese. Helicobacter **20**: 156-157, 2015.
9. Aoyama N, Shigeta S. Incidence and characteristics of *Helicobacter pylori* disappearance cases without "Eradication therapy"; spontaneous disappearance by gastric mucosal atrophy/intestinal metaplasia or unintended eradication by using antibiotics for other infectious diseases. Jpn J Helicobacter Res **19**: 112-116, 2018 (in Japanese).
10. Graham DY, Malaty HM, Evans DG, Evans DJ Jr, Klein PD, Adam E. Epidemiology of *Helicobacter pylori* in an asymptomatic population in the United States. Effect of age, race, and socioeconomic status. Gastroenterology **100**: 1495-1501, 1991.
11. Asaka M, Kimura T, Kudo M, et al. Relationship of *Helicobacter pylori* to serum pepsinogens in an asymptomatic Japanese population. Gastroenterology **102**: 760-766, 1992.
12. Kosunen TU, Seppälä K, Sarna S, Sipponen P. Diagnostic value of decreasing IgG, IgA, and IgM antibody titres after eradication of *Helicobacter pylori*. Lancet **339**: 893-895, 1992.
13. Cutler AF, Prasad VM. Long-term follow-up of *Helicobacter pylori* serology after successful eradication. Am J Gastroenterol **91**: 85-88, 1996.
14. Kimura K, Takemoto T. An endoscopic recognition of the atrophic border and its significance in chronic gastritis. Endoscopy **1**: 87-97, 1969.
15. Karasawa H, Sugiyama A, Takeda M, et al. ABC classification of gastric cancer risk based on a new kit to detect serum *Helicobacter pylori* antibodies: comparison with another antibody detection kit. J Gastrointestinal Cancer Screen **54**: 18-29, 2016 (in Japanese, Abstract in English).
16. Kato T, Yagi N, Kamada T, et al. Diagnosis of *Helicobacter pylori* infection in gastric mucosa by endoscopic features: a multicenter prospective study. Dig Endosc **25**: 508-518, 2013.
17. Watanabe K, Nagata N, Nakashima R, et al. Predictive findings for *Helicobacter pylori*-uninfected, -infected and -eradicated gastric mucosa: validation study. World J Gastroenterol **19**: 4374-4379, 2013.
18. Kato M, Terao S, Adachi K, et al. Changes in endoscopic findings of gastritis after cure of *H. pylori* infection: multicenter prospective trial. Dig Endosc **25**: 264-273, 2013.
19. Kamada T, Haruma K, Inoue K, Shiotani A. *Helicobacter pylori* infection and endoscopic gastritis -Kyoto classification of gastritis. Nihon Shokakibyo Gakkai Zasshi (Jpn J Gastroenterol) **112**: 982-993, 2015 (in Japanese, Abstract in English).
20. Kyoto classification of gastritis. Haruma K, Ed. Nihon Medical Center, Tokyo, 2017.
21. Goji S, Tamura Y, Sasaki M, et al. *Helicobacter suis*-infected nodular gastritis and a review of diagnostic sensitivity for *Helicobacter heilmannii*-like organisms. Case Rep Gastroenterol **9**: 179-187, 2015.
22. Shiratori S, Mabe K, Yoshii S, et al. Two cases of chronic gastritis with *non-Helicobacter pylori Helicobacter* infection. Intern Med **55**: 1865-1869, 2016.