Cantero-Fortiz *et al.*
Egypt J Neurol Psychiatry Neurosurg (2021) 57:164
https://doi.org/10.1186/s41983-021-00419-9

The Egyptian Journal of Neurology, Psychiatry and Neurosurgery

RESEARCH Open Access

Helicobacter pylori antibodies and multiple sclerosis: a single-center study and a short review of the literature

Yahveth Cantero-Fortiz[1,2], Iván Murrieta-Álvarez[2,3], Andrés Aurelio León-Peña[2,4], Miguel Antonio López-Trujillo[5], Alejandra Carmina Córdova-Ramírez[3], Montserrat Rivera-Álvarez[3], Gilberto David Elias-de-la-Cruz[3], Juan Carlos Olivares-Gazca[2,3], Guillermo J. Ruiz-Delgado[2,3,5] and Guillermo J. Ruiz-Argüelles[2,3,5*]

Abstract

Background: Multiple sclerosis is an immune-mediated disease which has been associated to a great variety of mechanisms that could influence its pathogenesis. Numerous reports in the medical literature suggest that *Helicobacter pylori* may be a mediator of the disease. However, it is unknown if there is any clear association between MS and HP.

Results: We studied 144 persons with multiple sclerosis prospectively enrolled in our hematopoietic stem cell transplantation program. In 144 persons, 14% patients were positive for IgG-HP whereas 86% were negative, 8.3% pwMS were IgM-HP positive while 91.6% pwMS were negative, 18% patients were positive and 82% negative for IgA-HP. Significantly lower concentrations of anti-HP IgG were found in RRMS in comparison with SPMS (− 28.5, 95% CI 4.3–52.7). While concentrations of anti-HP IgA were significantly lower in SPMS in comparison with RRMS (0.54, 95% CI 0.1–0.9). In a multivariate analysis, positivity rate of anti-HP IgG was found to be higher in SPMS patients (OR 4.7, 95% CI 1.1–19.6).

Conclusions: There was a negative correlation between the presence of anti-HP antibodies and MS**.** Further larger studies with specific laboratory testing methods are needed to discard or confirm the potential role of anti-HP antibodies as protective for MS.

Keywords: MS, *Helicobacter pylori*, Seroprevalence, Epidemiology

Background

Multiple sclerosis (MS) is a chronic neurological inflammatory disease, associated to immune-mediated injury, destruction of the central nervous system myelin and variable axonal injury [1]. Since it is an immune mediate disease has been associated to multiple mechanisms which could influence in its onset and clinical presentation [1, 2].

Helicobacter pylori (HP) is a Gram-negative bacteria that colonizes the gastric mucosa of more than half worldwide population, and is implicated in a large variety of disorders of the upper gastrointestinal tract and alterations related to host colonization [2].

There are some reports and evidence that relate HP to extra gastrointestinal diseases, specifically brain diseases, such as stroke, migraine, Alzheimer's disease and MS [3–5]. The latter represents the most common cause of non-traumatic disability that affects young adults, and its prevalence and incidence is increasing in developing and developed countries [6–9]. Taking together, those two widely different conditions may share an association in

*Correspondence: gruiz1@hsctmexico.com
[2] Centro de Hematología y Medicina Interna de Puebla, 8B Sur 3710, 72530 Puebla, Mexico
Full list of author information is available at the end of the article

the development or progression of the disease. Although some studies have reported a possible negative correlation suggesting that a high previous exposure to HP may be a protective effect associated with a reduction in the incidence of MS, some other studies have failed in the pursuit of this association [6–15].

In this cross-sectional study we aimed to assess the previous exposure status to HP through immunological screening in persons with MS (pwMS), to correlate the seroprevalence of HP to the clinical outcome of the disease in MS patients who underwent hematopoietic stem cell transplantation (HSCT).

To test the impact of whether HP exposure is related to the progression or response of MS in a cohort of patients undergoing HSCT.

Methods

Design

This work employed a convenience sampling of MS patients who were included within our HSCT program, from January 2018 to December 2019. The study was approved by the Ethics Committee and informed consent was obtained from all the patients.

Patients

All patients were diagnosed with MS according to revised McDonald [16] criteria. Patients were subjected to a physical evaluation by the same trained neurologist that included the scoring of the Expanded Disability Status Scale (EDSS) and an in-depth neurological consultation to obtain all relevant clinical data [17]. Patients with three different phenotypes of MS: relapsing–remitting (RRMS) and primary progressive (PPMS) and secondary progressive (SPMS) were included. Other inclusion criteria were: Karnofsky performance status above 70%, EDSS score of 8 or below, and discontinuation of disease modifying therapy 3 months before the assessment [18].

Ethics and consent to participate

The study was approved by the Ethics Committee (Conbioetica 21CEI00120130605, Registry No. 13 CEI 21 114 126). The study was conducted in accordance with Helsinki Declaration as revised in 2013, and written informed consent was obtained from the participants of the study after being fully informed about the procedure and possible complications.

Immunological screening of HP

For the identification of antibodies (IgG, IgM and IgA) against HP, a colorimetric immunoenzymatic method was used for each immunoglobulin isotype. We used a bacterial lysate of the HP strain ATCC43504 (ATTC Corp, VA, USA), the antigen was fixed at the bottom of each microtiter. In case of the samples to be positive, the specific antibody binds to the corresponding antigen. After washing the unbound proteins, a peroxidase conjugate of anti-human IgG, IgM or IgA antibodies is added to each well, correspondingly. The unbound conjugate was removed by a second wash to subsequently add a solution of 3,3′,5,5′-tetramethylbenzidine with an enzymatic substrate to track the specific binding of the color antibodies in case of positive samples, the color was measured by spectrophotometry at a wavelength of 450 nm. The antibody concentration was directly proportional to the intensity of the color displayed.

Statistical analysis

The data is displayed in frequency and percentages (%) for qualitative variables, quantitative variables are expressed in median and standard deviation (SD). The summary of data employed Mann–Whitney *U* test and chi-square for quantitative and qualitative data, respectively. One-way ANOVA was performed to detect significant differences among antibody concentrations in the three phenotypes of MS and to correct for multiple comparisons the single-step Bonferroni correction was employed [19]. To study the association of anti-*H. pylori* antibodies concentration and features of patients with MS, a linear regression model was employed for age and length of disease as long-term therapy may alter cellular and humoral immunity [20], while EDSS was studied with an ordinal linear regression. Both models used sex, phenotype of MS and history of disease modifying therapy (DMT), since there is evidence that use of DMT could impair humoral immunity [21, 22]. Regression coefficients and their 95% confidence intervals were calculated and reported accordingly. Phenotypes of MS were studied with logistic regression using RRMS as base outcome and age, sex, length of disease and previous history of DMT as covariates. Statistical procedures and analyses were completed with Prism 7 (GraphPad Software, San Diego, CA) and Stata 14 (Stata Corp, College Station, TX). For all the analyses a two-sided p value < 0.05 was set to establish statistical significance.

Results

Sample features

Between 2019 and 2020, a total of 144 pwMS enrolled in our HSCT program were assessed. The majority of the sample were female (65%), with a median age of 45 years (SD 10.6), and a mean EDSS of 4.3 (SD 1.97). The most frequent MS phenotype was RRMS (64%), followed by PPMS (20%), and SPMS (16%). The median age of onset of MS was 35 years old (SD 10.6) and a median duration of the disease from 9 years (SD 7.8). From the total number of patients, 67% reported a previous MS treatment

with some disease modifier drug 3 months before their arrival at our center (glatiramer acetate, natalizumab, dimethyl fumarate, teriflunomide, ocrelizumab, fingolimod, cladribine, etc.). The main features of the patients are displayed in Table 1.

HP screening results

From all the sample, 21 (14%) patients were positive for IgG-HP (3 PPMS, 6 SPMS, 12 RRMS) and 123 (86%) (26 PPMS, 18 SPMS, 79 RRMS) were negative. Moreover, nine (6.3%) pwMS were IgM-HP positive (1 PPMS, 0 SPMs, 11 RRMS), while 132 pwMS were negative (28 PPMS, 24 SPMS, 92 RRMS). 26 patients (18%) were positive and 118 (82%) were negative for IgA-HP (5 PPMS, 6 SPMS, 15 RRMS). There were no significant differences in proportions between the groups and positivity rate and anti-HP IgA, anti-HP IgM and anti-HP IgG. However, in the multivariate analysis SPMS presented a higher likelihood of presenting positive anti-HP IgG (*OR* 4.7, 95% CI 1.1–19.6), see Table 2.

Table 1 Demographic and clinical features in pwMS involved in the study

	PPMS ($n=29$)	SPMS ($n=24$)	RRMS ($n=91$)	GENERAL ($n=144$)	*p* value
Sex (%)					
Female	13 (44)	19 (79)	59 (64)	91 (63)	0.03
Male	16 (66)	5 (21)	32 (36)	53 (27)	
Age (SD)	47.6 (9.6)	50.6 (8.7)	42.3 (9.7)	44.8 (10.6)	< 0.0001
Weight (SD)	80 (19.6)	72 (14.5%)	73.4 (14.7)	74.5 (15.9)	0.1
Height (SD)	1.7 (0.09)	1.69 (0.09)	1.68 (0.1)	1.69 (0.1)	0.0888
BMI (SD)	26.5 (6)	25.2 (4.8)	25.84 (4.9)	25.87 (5.1)	0.6629
EDSS (SD)	4.9 (1.9)	5.9 (1.3)	3.6 (1.8)	4.3 (1.97)	< 0.0001
Length of disease(SD)	7 (6.6)	15 (7.6)	8.4 (7.7)	9.2 (7.9)	0.0003
Previous DMT (%)					
Yes	16 (66)	17 (70)	64 (70)	97 (67)	0.2929
No	13 (44)	7 (30)	27 (30)	47 (33)	

Table 2 Analysis of association between, results of IgA, IgM and IgG anti-HP antibodies with MS phenotype

IgG positive		**Univariate**		**Multivariate**	
		OR	**95% CI**	**OR**	**95% CI**
Type of MS	RRMS (base)	–	–	–	–
	PPMS	0.75	0.19–2.9	1.08	0.25–4.6
	SPMS	2.1	0.72–6.6	4.7	1.1–19.6
IgM positive		**Univariate**		**Multivariate**	
		OR	**95% CI**	**OR**	**95% CI**
Type of MS	RRMS (base)	–	–	–	–
	PPMS	0.25	0.03–2.1	0.39	0.04–3.3
	SPMS	–	–	–	–
IgA positive		**Univariate**		**Multivariate**	
		OR	**95% CI**	**OR**	**95% CI**
Type of MS	RRMS (base)	–	–	–	–
	PPMS	1.0	0.34–3.2	1.0	0.33–3.3
	SPMS	1.6	0.57–4.9	2.02	0.57–7.1

Age, sex, length of disease and previous history of immunomodulatory agents were used as covariates for the multivariate analysis

RRMS relapsing–remitting multiple sclerosis; *PPMS* primary-progressive multiple sclerosis; *SPMS*: secondary progressive multiple sclerosis; *OR* odds ratio; *95% CI* 95% confidence interval

Differences of antibodies concentrations among the MS phenotypes

The concentration of anti-HP IgG showed a significant difference among group (p 0.03) The Bonferroni correction post-hoc test revealed that IgG concentration was significantly lower in RRMS in comparison with SPMS. (− 28.5, 95% CI 4.3–52.7). Concentration of IgA was significantly different among the phenotypes (p 0.04) the post-hoc analysis showed that concentration of anti-HP IgA was significantly lower in SPMS group in comparison with the RRMS group (− 0.54, 95% CI 0.1–0.9), see Fig. 1. There were no significant differences in anti-HP IgM concentrations among the groups. The regression analyses did not find significant associations between anti-HP IgA, anti-HP IgM and anti-HP IgG with age, length of disease nor EDSS (Table 3).

Discussion

There is some scientific controversial evidence in support or against the role of HP as a potential trigger of a great variety of autoimmune diseases, including various neurological disorders of the central nervous system, such as MS [23].

Autoimmune diseases result from environmental exposures to bacteria, viruses and parasites in addition to genetic susceptibility [23–25]. There are several mechanisms triggered by infectious agents involved in the development of autoimmune diseases: T-regulatory imbalance, molecular mimicry, bystander effect, high levels of proinflammatory cytokines, epitope spreading, direct inflammatory damage, microbial super antigens, MHC class II expression on non-immune cells and immune complex formation [23, 26–28].

HP presents itself as a prevalent microbe, according to the medical literature it is present in 50–80% of the population worldwide being clinically implicated in gastritis, peptic ulcers, and appears as an independent risk factor for gastric carcinoma [29, 30]. Although many studies point out the fact that persistent HP infection represents a chronic inflammatory stimulus and could be a potential cause of the MS pathogenesis [29, 31, 32]. Despite the scientific evidence shown so far, the relationship between these two entities has not been clear.

In general population, HP seroprevalence determined by IgG anti-HP titles ranges between 30 and 50% in high income countries (HIC) and between 85 and 95% in low- and middle-income countries (LMIC) [33, 34], The

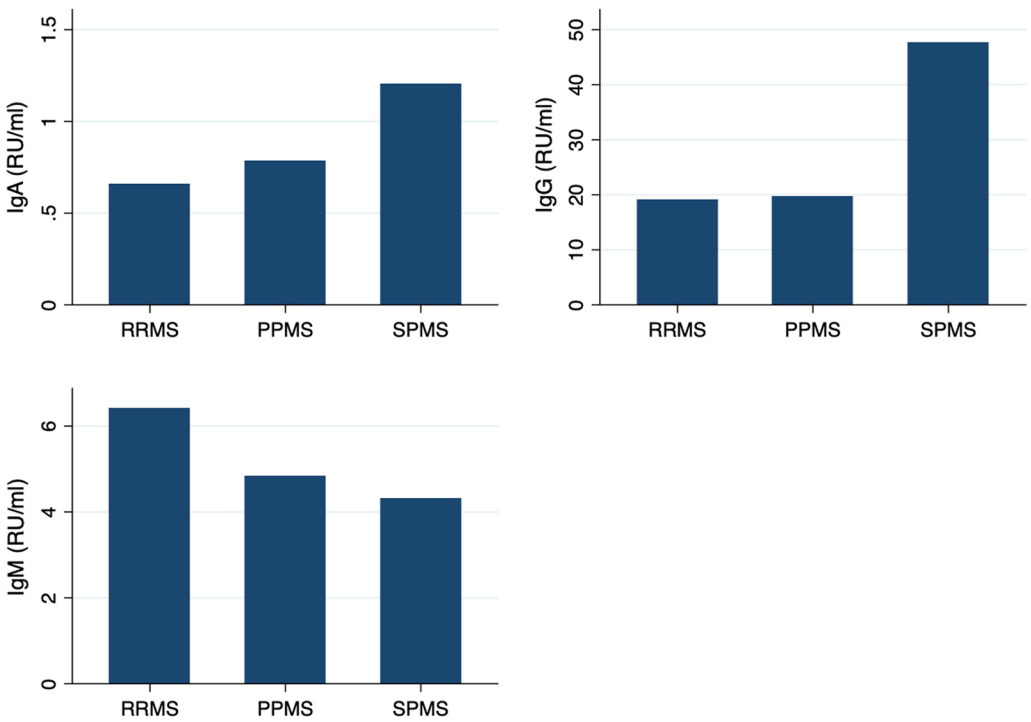

Fig. 1 Comparison of IgA, IgG and IgM titers by type of multiple sclerosis. After multiple comparison correction, a significant difference was observed in anti-HP IgA concentration between RRMS and SPMS (— 0.54 UR/ml, 95% CI 0.002–1.09). *RR/ml* relative units per milliliter; *RRMS* relapsing–remitting multiple sclerosis; *PPMS* primary-progressive multiple sclerosis; *SPMS* secondary-progressive multiple sclerosis

Table 3 Regression analysis between concentration of IgA, IgM and IgG anti-HP antibodies with EDSS, age and length of disease

IgG (RU/ml)	**Univariate**		**Multivariate**	
	Coef	**95% CI**	**Coef**	**95% CI**
EDSS[a]	−0.04	−0.1 to 0.09	−0.1	−0.3 to 0.01
Age	−0.4	−1.3 to 0.4	−0.81	−1.7 to 0.1
Length of disease	0.1	−0.9 to 1.3	−0.3	−1.2 to 1.1
IgM (RU/ml)	**Univariate**		**Multivariate**	
	Coef	**95% CI**	**Coef**	**95% CI**
EDSS[a]	−0.1	−0.2 to 0.1	−0.1	−0.2 to 0.06
Age	−0.07	−0.1 to -0.005	−0.06	−0.1 to 0.01
Length of disease	−0.02	−0.1 to 0.06	−0.01	−0.1 to 0.08
IgA (RU/ml)	**Univariate**		**Multivariate**	
	Coef	**95% CI**	**Coef**	**95% CI**
EDSS[a]	0.04	−0.1 to 0.1	−0.04	−0.2 to 0.1
Age	−0.0004	−0.01 to 0.01	−0.006	−0.02 to 0.01
Length of disease	0.001	−0.01 to 0.02	−0.001	−0.02 to 0.02

Phenotype of multiple sclerosis, sex, and previous history of immunomodulatory agents were used as covariates for the multivariate analysis

RU/ml Relative units per milliliter; *EDSS* Expanded disability status scale; *Coef* Regression coefficient; *95% CI* 95% confidence interval

[a] EDSS was considered as an ordinal variable and ordinal linear regression was conducted for this case

findings of the present study showed a low prevalence of IgG (14%), IgM (8.3%) and IgA (18%) HP antibodies in pwMS, these data differ from the worldwide prevalence and with a study by Long et al. [27] in which they report the presence of HP antibodies in 73% of the pwMS analyzed.

There are some other reports in which they found significative association between HP and MS, as seen in a study by Mohebi et al. [35]. They analyzed blood samples for IgG and IgM anti HP antibodies, but there was no significant difference in seropositivity with respect to age nor sex. In spite of these findings, they report a significant difference in EDSS value between seropositivity and seronegativity, concluding that HP infection could have a protective influence on MS pathogenesis [35]. This observation could be supported by our findings, since the prevalence of anti-HP antibodies in the cohort is substantially lower than that of the general population. Positive serology for anti-HP antibodies is higher in LMIC: In México, the prevalence of IgG anti-HP is 66–80%, figures that are higher than those found in this study, where most patients live in HIC [36]. Since the prevalence of MS is substantially higher in HIC as compared to LMIC [37], we could speculate that exposure to HP could be one of the possible explanations to this difference.

Since IgG against HP is the more used and accurate marker of previous exposure and none of the patients had significative titles of IgM and IgA nor clinical manifestations of HP infection at the moment of the study which indicates active infection, we decided to analyze IgG titles to compare seroprevalence of HP with the main demographic and clinical features in pwMS [34]. In contrast with the observations reported in the study by Mohebi et al. [35], we found significantly higher concentration of anti-HP IgG in SPMS in comparison with RRMS; however, we did not find significant association between seropositivity nor concentration of IgG against HP among inflammatory phenotype of MS, age, length of disease nor EDSS (Tables 2 and 3).

A meta-analysis performed by Yao et al. [3] addressed 9 prospective articles that included 1553 cases of MS, all these articles were case–control studies in which different diagnostic methods were employed to detect HP infection (ELISA, Western Blot, immunofluorescence and latex agglutination). They reported a lower prevalence of HP infection of almost 25% in patients with MS with no significant differences among the HP identification methods employed. The prevalence found by Yao et al. [3] is considerably higher to the observed prevalence reported in our study. The study reports important limitations such as the difficulty to distinguish previous and active infection employing serological tests and the lack of subgroup analysis of gender, MS phenotype and length of the disease [3]. This metanalysis concluded that HP infection and MS may have a

negative correlation, but also suggests the existence of a potential protective factor against MS [3].

Some reports found a negative correlation on the laboratory method employed to the IgG anti HP detection [3], although other studies suggest high relevance on several specific IgG HP antigens which are important on the loss of immunological tolerance to myelin antigens particularly heat shock proteins (hsp), such as hsp60 and hsp70 [11]. In an Egyptian study Gerges et al. [11] reported a high correlation between anti HP hsp60 IgG level and SPMS and propose anti HP hsp60 IgG as a biomarker for progression of MS, being correlated to age and length of the disease in a moderate level and with EDSS in a strong level, moreover, low levels of HP infection in western countries.

However, these specific IgG antibodies have been poorly studied, partly because of the limited access and availability of these particular laboratory method in certain regions of the globe, being an issue that may represent a great disparity among studies which have tried to demonstrate a positive correlation with MS [11].

There are other recent studies that could support our results, such is the case of the study performed by Ranjbar et al. [38] in which authors determined the seroprevalence of HP in 387 pwMS vs 420 healthy subjects, which represents a larger patient cohort compared to other reports. The authors found a significantly lower seropositivity in pwMS than the healthy individuals, a lower EDSS in HP seropositive pwMS compared with seronegative pwMS, proinflammatory cytokines (IFN-y, TNF-a, IL-6, IL-7) significantly lower as compared to seronegative patients and increased levels of IL-4 and IL-10 compared to seronegative patients, concluding that HP infection has a negative correlation in MS and may act as a protective agent in MS [38]. The exact pathogenic mechanism remains unclear, although these findings could suggest that a poor HP exposition in persons predisposed to develop MS could trigger the onset and the clinical length of the disease.

Regarding the geographical location, we stratified the population according to the country of origin of all the pwMS detecting that the majority of them (99.3%) belongs to Western countries. This scenario made it impossible to perform a comparison between the eastern and western regions to contrast what was established in the current scientific literature. However, our data are consistent with the low prevalence in pwSM specially in Western countries reported in other studies and could support the hypothesis in which HP plays a role as a protective effect reducing the risk of MS [3, 38] (Fig. 2).

This study showed some limitations related to the heterogeneity of the sample: Since MS is a disease with higher latitudinal prevalence [39], all the patients included in the study and treated at our center were from different regions of the globe, in addition to these differences they coursed with aggressive and rapidly progressive forms of the disease. This scenario made it difficult to create a control group with homogeneous characteristics as compared with other studies in which authors analyzed pwMS from a single population. However, patients

Fig. 2 Geographic distribution of participants of the study

were studied, analyzed, and compared with the current scientific data.

Conclusions

In spite of the poor association among IgG anti HP antibodies and gender, age, weight, height, BMI, EDSS and length of the disease in pwMS that here we report, there is still controversial data in support or against this correlation. Beside this, it is important to remark that further studies with specific laboratory testing methods and larger cohorts of patients are needed to discard or confirm the potential role of the previous exposure to HP and the presence of anti-HP hsp60 IgG as a clinical biomarker in the pathogenesis of MS and if there is a geographical influence on the presence of this agent.

Abbreviations

MS: Multiple sclerosis; HP: *Helicobacter pylori*; pwMS: Persons with MS; HSCT: Hematopoietic stem cell transplantation; EDSS: The Expanded Disability Status Scale; RRMS: Relapsing–remitting; PPMS: Primary progressive; SPMS: Secondary progressive; DMT: Disease modifying therapy; BMI: Body Mass Index; HIC: High income countries; LMIC: Low and middle-income countries.

Acknowledgements

Not applicable.

Authors' contributions

YC-F, GJR-D, and GJR-A: conceptualization, design, analysis and writing. IM-A, AAL-P, MAL-T, MR-A, GDE-C, JCO-G and ACC-R: analysis, writing and review. All authors have read and approved the manuscript.

Funding

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The study was conducted in accordance with Helsinki Declaration as revised in 2013, and written informed consent was obtained from the participants of the study.
The study was approved by the Ethics Committee (Conbioetica 21CEI00120130605, Registry No. 13 CEI 21 114 126). All patients signed a consent form after being fully informed about the procedure and possible complications.

Consent for publication

Not applicable.

Competing interests

The authors declare no conflicts of interest.

Author details

[1]Universidad de las Américas Puebla, Puebla, Mexico. [2]Centro de Hematología y Medicina Interna de Puebla, 8B Sur 3710, 72530 Puebla, Mexico. [3]Universidad Popular Autónoma del Estado de Puebla, Puebla, Mexico. [4]Benemérita Universidad Autónoma de Puebla, Puebla, Mexico. [5]Laboratorios Clínicos de Puebla, Puebla, Mexico.

Received: 10 March 2021 Accepted: 19 November 2021
Published online: 05 December 2021

References

1. Ruiz-Argüelles GJ, Olivares-Gazca JC, Olivares-Gazca M, Leon-Peña AA, Murrieta-Alvarez I, Cantero-Fortiz Y, et al. Self-reported changes in the expanded disability status scale score in patients with multiple sclerosis after autologous stem cell transplants: real-world data from a single center. Clin Exp Immunol. 2019;198(3):351–8. https://doi.org/10.1111/cei.13358.
2. Sgouras DN, Trang TT, Yamaoka Y. Pathogenesis of *Helicobacter pylori* Infection. Helicobacter. 2015;20(1):8–16. https://doi.org/10.1111/hel.12251.
3. Gang Y, Ping W, Xiang-Dan L, Ting-Min Y, Harris RA. Meta-analysis of association between *Helicobacter pylori* infection and multiple sclerosis. Neurosci Lett. 2016;620:1–7. https://doi.org/10.1016/j.neulet.2016.03.037.
4. Huang WS, Tseng CH, Lin CL, Tsai CH, Kao CH. *Helicobacter pylori* infection increases subsequent ischemic stroke risk: a nationwide population-based retrospective cohort study. QJM. 2014;107(12):969–75. https://doi.org/10.1093/qjmed/hcu117.
5. Kountouras J, Zavos C, Polyzos SA, Katsinelos P, Deretzi G. Association between cirrhosis and *Helicobacter pylori*-related brain pathologies. Eur J Gastroenterol Hepatol. 2015;27(2):183. https://doi.org/10.1097/MEG.0000000000000248.
6. Dobson R, Giovannoni G. Multiple sclerosis- a review. Eur J Neurol. 2019;26(1):27–40. https://doi.org/10.1111/ene.13819.
7. Kobelt G, Thompson A, Berg J, Gannedahl M, Eriksson J. European multiple sclerosis platform. New insights into the burden and costs of multiple sclerosis in Europe. Mult Scler. 2017;23(8):1123–36. https://doi.org/10.1177/1352458517694432.
8. Ascherio A. Environmental factors in multiple sclerosis. Expert Rev Neurother. 2013;13:3–9. https://doi.org/10.1586/14737175.2013.865866.
9. Browne P, Chandraratna D, Angood C, Tremlett H, Baker C, Taylor BV, et al. Atlas of Multiple Sclerosis 2013: a growing global problem with widespread inequity. Neurology. 2014;83(11):1022–4. https://doi.org/10.1212/WNL.0000000000000768.
10. Solaro C, Messmer UM. Management of pain in multiple sclerosis: a pharmacological approach. Nat Rev Neurol. 2011;7(9):519–27. https://doi.org/10.1038/nrneurol.2011.120.
11. Gerges SE, Alosh TK, Khalil SH, El Din MMW. Relevance of *Helicobacter pylori* infection in Egyptian multiple sclerosis patients. Egypt J Neurol Psychiatr Neurosurg. 2018;54(1):41. https://doi.org/10.1186/s41983-018-0043-x.
12. Pedrini MJ, Seewann A, Bennett KA, Wood AJ, James I, Burton J, et al. *Helicobacter pylori* infection as a protective factor against multiple sclerosis risk in females. J Neurol Neurosurg Psychiatry. 2015;86(6):603–7. https://doi.org/10.1136/jnnp-2014-309495.
13. Mohebi N, et al. Relation of *Helicobacter pylori* infection and multiple sclerosis in Iranian patients. Neurol Int. 2013;5(2):31–3. https://doi.org/10.4081/ni.2013.e10.
14. Bennett KA, et al. Western Australian multiple sclerosis patients exhibit a lower prevalence of *Helicobacter pylori* infection. Mult Scler. 2012;18(4):521.
15. Gavalas E, Kountouras J, Boziki M, Zavos C, Polyzos SA, Vlachaki E, et al. Relationship between *Helicobacter pylori* infection and multiple sclerosis. Ann Gastroenterol. 2015;28(3):353–6.
16. Thompson AJ, Banwell BL, Barkhof F, Carroll WM, Coetzee T, Comi G, Correale J. Diagnosis of multiple sclerosis: 2017 revisions of the McDonald criteria. Lancet Neurol. 2018;17(2):162–73. https://doi.org/10.1016/S1474-4422(17)30470-2.
17. Kurtzke JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology. 1983;33(11):1444–52.
18. Péus D, Newcomb N, Hofer S. Appraisal of the Karnofsky Performance Status and proposal of a simple algorithmic system for its evaluation. BMC Med Inform Decis Mak. 2013;13:72.
19. Ludbrook J. Multiple comparison procedures updated. Clin Exp Pharmacol Physiol. 1998;25(12):1032–7.

20. Paghera S, Sottini A, Previcini V, Capra R, Imberti L. Age-related lymphocyte output during disease-modifying therapies for multiple sclerosis. Drugs Aging. 2020;37(10):739–46.
21. Selter RC, Biberacher V, Grummel V, Buck D, Einbröker C, Oertel WH, et al. Netalizumab treatment decreases serum IgM and IgG levels in multiple sclerosis patients. Mult Scler. 2013;19(11):1454–61.
22. Pellegrino P, Carnovale C, Perrone V, Pozzi M, Antoniazzi S, Radice S, et al. Efficacy of vaccination against influenza in patients with multiple sclerosis: the role of concomitant therapies. Vaccine. 2014;32(32):4730–5.
23. Smyk DS, Koutsoumpas AL, Mytilinaiou MG, Rigopoulou EI, Sakkas LI, Bogdanos DP. *Helicobacter pylori* and autoimmune disease: cause or bystander. World J Gastroenterol. 2014;20(3):613–29. https://doi.org/10.3748/wjg.v20.i3.613.
24. Shoenfeld Y, Zandman-Goddard G, Stojanovich L, Cutolo M, Amital H, Levy Y. The mosaic of autoimmunity: hormonal and environmental factors involved in autoimmune diseases–2008. Isr Med Assoc J. 2008;10(1):8–12.
25. Smyk D, Rigopoulou EI, Baum H, Burroughs AK, Vergani D, Bogdanos DP. Autoimmunity and environment: am I at risk? Clin Rev Allergy Immunol. 2012;42(2):199–212. https://doi.org/10.1007/s12016-011-8259-x.
26. Getts MT, Miller SD. 99th Dahlem conference on infection, inflammation and chronic inflammatory disorders: triggering of autoimmune diseases by infections. Clin Exp Immunol. 2010;160(1):15–21. https://doi.org/10.1111/j.1365-2249.2010.04132.x.
27. Fujinami RS, von Herrath MG, Christen U, Whitton JL. Molecular mimicry, bystander activation, or viral persistence: infections and autoimmune disease. Clin Microbiol Rev. 2006;19(1):80–94. https://doi.org/10.1128/CMR.19.1.80-94.2006.
28. Röner S, Zinser E, Menges M, Wiethe C, Littmann L, Hänig J. Minor role of bystander tolerance to fetal calf serum in a peptide-specific dendritic cell vaccine model against autoimmunity: comparison with serum-free cultures. J Immunother. 2008;31(7):656–64. https://doi.org/10.1097/CJI.0b013e31818283ef.
29. Long Y, Gao C, Qiu W, Hu X, Shu Y, Peng F, Lu Z. *Helicobacter pylori* infection in neuromyelitis optica and multiple sclerosis. NeuroImmunoModulation. 2013;20(2):107–12. https://doi.org/10.1159/000345838.
30. Hasni S, Ippolito A, Illei GG. *Helicobacter pylori* and autoimmune diseases. Oral Dis. 2011;17(7):621–7. https://doi.org/10.1111/j.1601-0825.2011.01796.x.
31. Kountouras J, Zavos C, Gavalas E, et al. *Helicobacter pylori* may be a common denominator associated with systemic and multiple sclerosis. Joint Bone Spine. 2011;78(2):222–3. https://doi.org/10.1016/j.jbspin.2011.01.006.
32. Prasad H, Krishnaprasad MS, Karnaker VK. Therapeutic induction of *Helicobacter pylori* bacteraemia in multiple sclerosis: how far from reality. Med Hypotheses. 2008;71(4):610–609. https://doi.org/10.1016/j.mehy.2008.05.025.
33. Khoder G, Muhammad JS, Mahmoud I, Soliman SSM, Burucoa C. Prevalence of *Helicobacter pylori* and its associated factors among healthy asymptomatic residents in the United Arab Emirates. Pathogens. 2019;8(2):44. https://doi.org/10.3390/pathogens8020044.
34. Hooi JKY, Lai WY, Ng WK, Suen MMY, Underwood FE, Tanyingoh D, et al. Global prevalence of *Helicobacter pylori* infection: systematic review and meta-analysis. Gastroenterology. 2017;153(2):420–9. https://doi.org/10.1053/j.gastro.2017.04.022.
35. Mohebi N, Mamarabadi M, Moghaddasi M. Relation of *Helicobacter pylori* infection and multiple sclerosis in Iranian patients. Neurol Int. 2013;5(2):31–3. https://doi.org/10.4081/ni.2013.e10.
36. Alarid-Escudero F, Enns EA, MacLehose RF, Parsonnet J, Torres J, Kuntz KM. Force of infection of *Helicobacter pylori* in Mexico: evidence from a national survey using a hierarchical Bayesian model. Epidemiol Infect. 2018;146(8):961–9. https://doi.org/10.1017/S0950268818000857.
37. Walton C, King R, Rechtman L, Kaye W, Leray E, Marrie RA, et al. Rising prevalence of multiple sclerosis worldwide: Insights from the Atlas of MS, third edition. Mult Scler. 2020;26(14):1816–21. https://doi.org/10.1177/1352458520970841.
38. Ranjbar R, Karampoor S, Jalilian FA. The protective effect of *Helicobacter pylori* infection on the susceptibility of multiple sclerosis. J Neuroimmunol. 2019;337: 577069. https://doi.org/10.1016/j.jneuroim.2019.577069.
39. Wood H. Multiple sclerosis: latitude and vitamin D influence disease course in multiple sclerosis. Nat Rev Neurol. 2017;13(1):3. https://doi.org/10.1038/nrneurol.2016.181.

Publisher's Note