

RESEARCH ARTICLE

# The Role of Morbid Obesity in the Promotion of Metabolic Disruptions and Non-Alcoholic Steatohepatitis by Helicobacter Pylori

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## Abstract

### Background

Helicobacter pylori (HP) infection has been associated to an increased rate of type 2 diabetes (T2D) and liver disease through its effect on insulin resistance and systemic inflammation. However, results are inconstant and no studies exist in morbidly obese patients, in which both insulin resistance and inflammation coexist.

### Material and Methods

Cross-sectional study to evaluate the relationship between HP infection and alterations in carbohydrate metabolism, lipid profile, inflammation markers, and liver disease in patients awaiting for bariatric surgery. HP infection was histologically assessed in gastric antrum biopsy from 416 subjects. Liver biopsy was also available in 93 subjects.

### Results

Both impaired fasting glucose and T2D were similar when comparing subjects with and without HP infection (24.2% vs. 22%,  $p = 0.290$  and 29.4% vs. 29.1%,  $p = 0.916$ , respectively), with no differences between groups in the HOMA-IR, lipid profile neither inflammatory parameters. However, HP infection was higher among subjects with a BMI  $\geq 40.0$  kg/m<sup>2</sup> in comparison with lower degrees of obesity (71.7% vs. 60.0%,  $p = 0.041$ ). In addition, subjects without

HP infection showed higher degrees of steatosis ( $44.1 \pm 26.4\%$  vs.  $32.0 \pm 20.7\%$ ,  $p = 0.038$ ), as well as a lower prevalence of non-alcoholic steatohepatitis ( $9.3\%$  vs.  $30.7\%$ ,  $p = 0.023$ ).

## Conclusions

In patients with morbid obesity, HP infection does not seem to be associated with abnormal carbohydrate metabolism. In addition, less advanced degrees of non-alcoholic fatty disease were observed. We suggest that low-grade inflammation that accompanies obesity mitigates the diabetogenic effect of HP, so the presence of obesity should be considered in studies that evaluate the HP metabolic effects.

## Introduction

Helicobacter pylori (HP) is a micro-aerophilic, flagellated, Gram-negative pathogen, highly adapted for survival in the acidic pH [1]. HP successfully colonizes human stomach, and is present in more than half of worldwide population, being more common in societies with low socioeconomic development [1]. Colonization of the gastric mucosa by HP is acquired early in life, it will persist unless antibiotic therapy, and induce not only gastric diseases, but also metabolic manifestations [2, 3]. In this way, HP seropositivity has been related with an increased rate of incident type 2 diabetes (T2D) after 10 years' follow-up period [4]. In accordance with this finding, cross-sectional studies have also found a positive association between HP infection and the prevalence of T2D in selected populations [5, 6]. However, in contrast with these results, other studies have failed to report this relationship [7, 8]. Considering that HP does not pass into systemic circulation, it has been suggested that extragastric manifestations are mediated by insulin resistance, in relation with proinflammatory cytokines and acute phase reactants produced by the inflamed mucosa [3].

HP infection has also been associated with food intake regulation through the effect on hormones that regulate appetite, with lower production of ghrelin, and higher concentrations of leptin in comparison with HP negative subjects [9, 10]. In this way, existing data show that HP eradication in patients with peptic ulcer disease significantly increases the incidence of obesity [10, 11]. In addition, morbidly obese subjects from United States showed a 1.7-fold increase in HP infection in comparison with non-obese controls (95% CI: 1.3 to 2.2) [12].

Finally, a role for HP infection in the development of non-alcoholic fatty liver disease (NAFLD) has also been suggested [13]. In fact, the inoculation with HP in an animal model induced a significant increase in the fibrotic score and aminotransferase activity [14]. However, studies with human liver biopsy according to HP infection are scarce.

On this basis, our aim was to go deeper on assessing the relationship between HP infection and alterations in carbohydrate metabolism, lipid profile and inflammatory markers in a homogeneous sample of 416 subjects with morbid obesity. For this purpose, we have chosen this population of severely obese subjects in which metabolic disruptions and low-grade chronic inflammation are usual features. In addition, the relationship between HP and NAFLD was assessed in 93 subjects with both gastric and liver biopsies.

## Material and Methods

### Ethics statement

Informed written consent was obtained from all participants, and the human ethics committee of the two participating University Hospitals (Anau de Vilanova and Vall d'Hebron) approved

the study. An additional informed written consent was obtained from patients who underwent liver biopsy.

## Description of patients

A total of 416 patients of Caucasian origin attending to the outpatient Obesity Unit of two University hospitals (Arnau de Vilanova and Vall d'Hebron) were enrolled at the time of a regular visit between July 2010 and June 2014. Patients were recruited irrespective of the presence of gastrointestinal symptoms. The study was conducted according to the ethical guidelines of the Helsinki Declaration.

The next exclusion criteria were considered: type 1 diabetes, previous HP eradication therapy, requirements of corticosteroid treatment, malignancy, end stage renal disease, chronic liver disease, consuming more than 200 g of alcohol per week, or absence of pathology data for the study.

All patients underwent an established protocol before surgery, including upper gastrointestinal endoscopy and gastric antrum biopsy. HP status was evaluated in each patient by histopathology examination by the Diff-Quick method [15]. Laboratory tests included fasting plasma glucose, glycated hemoglobin A1c (HbA1c), fasting insulin, liver function tests (alanine aminotransferase, aspartate aminotransferase, and gamma-glutamyl transpeptidase), lipid profile (triglycerides, high and low-density lipoprotein), and markers of inflammation [(leukocyte count, ferritin, erythrocyte sedimentation rate (ESR), and C-reactive protein (CRP)]. Demographic data (gender and age), BMI and waist circumference, and prior diagnosis of T2D were also collected. Insulin resistance was estimated using the homeostatic model assessment method (HOMA) [16]. T2D and impaired fasting glucose (IFG) were defined according to the criteria recommended by the Expert Committee on the Diagnosis and Classification of Diabetes [17].

Liver biopsy was taken at the time of bariatric surgery with a Hepafix needle in 93 subjects. All biopsies were at least 2 cm long and contained at least eight portal tracts. Liver specimens were stained with hematoxylin eosin, picosiriums for fibrosis and periodic acid Schiff (PAS) with diastase to help clarify the degree of inflammation. Liver histology was assessed using a systemic approach of necroinflammatory grading and fibrosis staging as described by Brunt et al [18] and modified by Kleine et al [19]. Individual histological features were observed and scored separately. Finally, all were graded and staged for non-alcoholic steatohepatitis (NASH) according to the system proposed at the American Association for the Study of Liver Diseases single topic conference in September 2002 [20].

## Statistical analysis

The normal distribution of the variables was evaluated using the Kolmogorov-Smirnov test. Data were expressed either as the mean  $\pm$  SD, percentage or median (total range). Comparisons between groups were performed using the Student's *t* test or the Mann-Whitney *U* test for continuous variables, as well as the  $\chi^2$  test for categorical variables. All "p" values were based on a two-sided test of statistical significance. Significance was accepted at the level of  $p < 0.05$ . Statistical analyses were performed using the SPSS statistical package (SPSS, Chicago, IL, USA).

## Results

The prevalence of HP infection in the whole study population was 69.5%, without differences between those with and without T2D (69.1% vs. 69.6%,  $p = 0.908$ ). However, HP infection was

**Table 1. Main clinical characteristics, metabolic data, liver function, lipid profile, and inflammatory parameters according to HP positivity.**

	HP positive	HP negative	P value
<b>N</b>	289	127	-
<b>Women, n (%)</b>	210 (72.6)	94 (74.0)	0.775
<b>Age (yrs)</b>	45.9 ± 9.8	44.6 ± 11.0	0.261
<b>BMI (kg/m<sup>2</sup>)</b>	44.4 ± 5.1	44.1 ± 5.6	0.600
<b>IFG, n (%)</b>	70 (24.2)	28 (22.0)	0.290
<b>T2D, n (%)</b>	85 (29.4)	38 (29.9)	0.916
<b>Fasting plasma glucose (mmol/l)</b>	6.4 ± 2.5	6.1 ± 1.9	0.304
<b>HbA1c (%)</b>	5.7 (4–12.3)	5.6 (4.7–11.4)	0.498
<b>HOMA-IR<sup>a</sup></b>	5.9 ± 7.0	5.6 ± 3.5	0.691
<b>AST (IU)</b>	22.5 ± 8.5	27.4 ± 16.7	0.316
<b>ALT (IU)</b>	25.0 ± 12.7	29.0 ± 23.6	0.264
<b>GGT (IU)</b>	36.8 ± 40.7	44.1 ± 31.2	0.375
<b>Total cholesterol (mmol/l)</b>	5.9 ± 1.0	5.0 ± 0.9	0.128
<b>HDL cholesterol (mmol/l)</b>	1.2 ± 0.2	1.1 ± 0.2	0.121
<b>LDL cholesterol (mmol/l)</b>	3.1 ± 1.9	3.1 ± 0.8	0.409
<b>Triglycerides (mmol/l)</b>	1.7 ± 0.9	1.7 ± 0.9	0.623
<b>C-reactive protein (mg/l)</b>	1.0 ± 0.9	0.8 ± 0.6	0.130
<b>Erythrocyte sedimentation rate (mm/h)</b>	27.6 ± 19.8	24.5 ± 16.0	0.192
<b>White blood cell count (x10<sup>9</sup>/l)</b>	8.2 ± 2.2	8.0 ± 2.0	0.531
<b>Ferritin (ng/ml)</b>	190.1 (8–591)	186.2 (8–550)	0.755

BMI: body mass index; IFG: impaired fasting glucose; T2D: type 2 diabetes; AST: aspartate aminotransferase; ALT: alanine aminotransferase; GGT: gamma-glutamyl transpeptidase.

<sup>a</sup>: HOMA-IR was calculated only in subjects without T2D.

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significantly higher among subjects with a BMI ≥ 40.0 kg/m<sup>2</sup> in comparison with lower degrees of obesity (71.7% vs. 60.0%, p = 0.041).

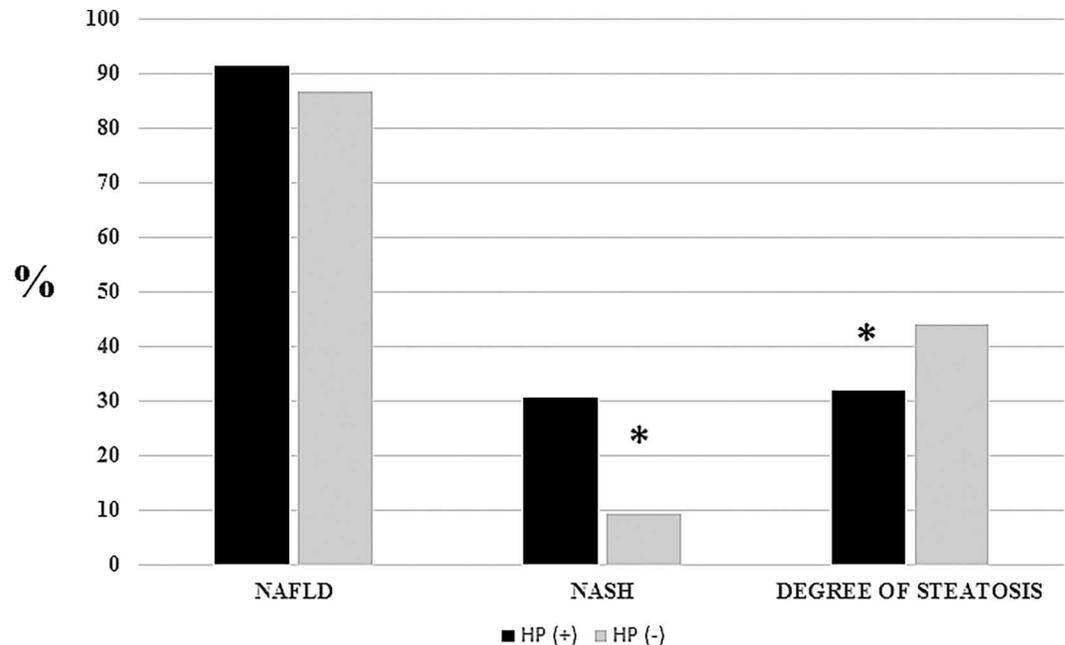
On the other hand, the prevalence of both IFG and T2D was similar when comparing subjects with and without HP infection (24.2% vs. 22.0%, p = 0.290 and 29.4% vs. 29.9%, p = 0.916, respectively). In addition, no differences between subjects HP + and HP- in their lipid profile, liver function test neither inflammatory parameters were found (Table 1).

When the subgroup of non-diabetic patients was evaluated (n = 293), HP infection was not associated with higher levels of insulin resistance (HOMA-IR: 5.6 ± 7.6 vs. 5.6 ± 3.4, p = 0.993) neither fasting plasma glucose (98.1 ± 14.6 vs. 95.0 ± 10.6 mg/dl, p = 0.079) or HbA1c (6.1 ± 6.7% vs. 5.8 ± 5.4%, p = 0.733).

Finally, the prevalence of NAFLD was similar between subjects with and without HP infection (91.4% vs 86.6%, p = 0.499). However, among those subjects with NAFLD, those without HP infection presented a lower percentage of NASH (9.3% vs. 30.7%, p = 0.023) as well as higher degree of steatosis (44.1 ± 26.4% vs. 32.0 ± 20.7, p = 0.038) (Fig 1).

## Discussion

We failed to observe any difference in the prevalence of carbohydrate abnormalities according to the presence of histologically proven HP infection in a subgroup of subjects with morbid obesity. In recent years HP infection has been associated with increased insulin resistance [21, 22], as well as with a greater incidence and prevalence of T2D and metabolic syndrome [4, 23, 24]. However, this link appears to be inconstant, mainly attributed to heterogeneous study



**Fig 1. Prevalence of non-alcoholic fatty liver disease, non-alcoholic steatohepatitis, and the degree of steatosis according to the presence of Helicobacter Pylori infection in the subgroup of 93 patients with both gastric and liver biopsies.** HP: Helicobacter Pylori; NAFLD: non-alcoholic fatty liver disease; NASH: non-alcoholic steatohepatitis.

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populations, with or without gastrointestinal symptoms, and the diversity of the diagnostic techniques (histology vs. serology) to confirm HP infection [25].

Regarding insulin resistance, a recent systematic review confirmed the positive epidemiological association with HP infection [21]. Similarly, among non-diabetic subjects, those with HOMA-IR  $\geq 2.5$  showed a significantly higher rate of HP seropositivity compared with non-insulin resistant subjects [22]. These data contrast with our results in morbidly obese subjects, in whom high degrees of insulin resistance were observed independently of HP status. Discordant results also emerge when the effect on HOMA-IR is evaluated after HP eradication [3, 26, 27, 28].

On the other hand, when data from participants included in the NHANES III and NHANES 1999–2000 was evaluated, there were no association between HP seropositivity and history of self-reported diabetes. Only after excluding individuals with history of diabetes and controlling for potential confounder factors, HP seropositivity was positively associated with higher mean HbA1c levels [29]. In addition, a recent meta-analysis that included more than 14,000 patients from forty-one observational studies communicated an OR for HP infection of 1.33 (95% CI: 1.08–1.64;  $p = 0.008$ ) among patients with diabetes [30]. Nevertheless, our results do not confirm these data, as no differences in HbA1c were detected among morbidly obese patients without DM according to HP infection, as well as no differences appeared in the prevalence of T2D when the whole population was evaluated.

The evaluation by indirect methods like anti-HP titer has the disadvantage that the serological positivity can persist despite the bacterial eradication, resulting in subject misclassification [31]. Therefore, in a recent study of 5,889 subjects, the association between metabolic syndrome and HP infection seemed to be higher with histologic positivity for HP [adjusted odds ratio = 1.26; 95% CI 1.08–1.48] than serologic positivity (adjusted odds ratio = 1.12; 95% CI 0.95–1.32), after adjusting for age, sex, smoking status, alcohol consumption, and economic

status [23]. In our study histologic positivity for HP was mandatory, reinforcing the lack of relationship between HP infection and abnormal carbohydrate metabolism in obese patients awaiting for bariatric surgery.

The prevalence of HP infection in patients with morbid obesity is highly variable, ranging between 11 and 85%. However, a higher prevalence of HP infection among bariatric patients compared to the general population [12, 32] and similar non-obese groups [33–35] has been reported. For example, the prevalence rate of 48% HP seropositivity in the general population from United States increased significantly to 61% in morbidly obese patients [12]. Also in a study conducted with the NHANES population (13,489 participants) it was observed that the effect of HP on impaired glucose tolerance could be enhanced by a high BMI [29]. Similarly, the rate of HP infection significantly increased from 60.0% in patients with a BMI < 40 kg/m<sup>2</sup> to 71.7% when the BMI was equal or higher than 40.0 kg/m<sup>2</sup>.

The contributing effect of HP infection to the development of NAFLD has also been suggested, as well as for the progression from hepatic steatosis to NASH [13, 36–38]. In addition to insulin resistance, invasion of HP in the small bowel mucosa might increase intestinal permeability and facilitate the passage of bacterial endotoxins via the portal vein to the liver [13]. In this way, the severity of steatosis in humans has been correlated with the disruption of intercellular tight junctions in the gut, increasing intestinal permeability, and the intestinal bacterial overgrowth in the small bowel microbiota [39]. In a recent work, the prevalence of NASH, the total NAFLD activity score, and the grade of hepatocyte ballooning were significantly higher in patients positive for anti-HP immunoglobulin G than in those negative [36]. Similarly, higher rates of anti-HP IgG were detected in patients with biopsy-proven NAFLD compared to control group [38]. Despite this, a similar prevalence of NAFLD among patients with and without HP infection was detected in our population of morbidly obese subjects. In addition, an unexpected finding was observed in those individuals with NAFLD: obese patients with histologic positivity for HP showed higher degrees of liver steatosis and lower prevalence of NASH than subjects without HP infection. Our results are in concordance with those observed in a large-scale cross-sectional study performed with Japanese adults in whom body mass index, platelet count, and serum ALT, but not HP infection, were positively associated with NAFLD diagnosed based on ultrasound methodology [40]. As imaging has limited diagnostic value for NAFLD, the prediction of fatty liver has been investigated through various surrogate scores, including the fatty liver index (FLI), and the hepatic steatosis index (HSI) in general population [41, 42]. However, those scores fail to be a valid diagnostic tool for non-alcoholic fatty liver disease (NAFLD) in our population of morbidly obese subjects [43].

The mechanism implicated in the metabolic effects related with HP infection seems to be independent of the clinical manifestations and related with a systemic low-grade inflammatory process initiated at the gastric level [1, 44]. In this way, HP has been correlated with an increased release of pro-inflammatory cytokines and acute phase proteins, such as CRP, IL-6, and TNF- $\alpha$  [45–47] which have not been confirmed in our study with morbidly obese subjects. Similarly, in a 10-years prospective cohort study, among 782 diabetes-free elderly Latino subjects, baseline serological HP infection, but no inflammatory cytokines (i.e., CRP and IL-6), was associated with a 2.7-fold increase in risk to develop diabetes compared with individuals without the infection [4].

Some limitations have to be considered when analyzing our results. First, we have not considered the presence of gastrointestinal symptoms in HP infected patients, what has been considered as a confounding factor. Second, our study has evaluated not only very obese but also young subjects. Despite the possibility that a longer period with an active HP infection could enhance its systemic effects cannot be discharged, we believe this is not a determinant factor in our results since HP infection is acquired early in life.

## Conclusions

In conclusion, in view of the results of our study, we can suggest that data observed in normo-weight and overweight population does not seem to be true in patients with morbidly obesity, in whom HP infection does not seem to be associated with a higher prevalence of abnormal carbohydrate metabolism. It is possible that low-grade inflammation that accompanies obesity mitigates the diabetogenic effect of HP in this population, so the presence of obese patients should be considered in studies that seek to evaluate the HP metabolic effect.

## Author Contributions

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## References

1. Cover TL, Blaser MJ. Helicobacter pylori infection and disease. *Gastroenterology* 2009; 136: 1863–1873. doi: [10.1053/j.gastro.2009.01.073](https://doi.org/10.1053/j.gastro.2009.01.073) PMID: [19457415](https://pubmed.ncbi.nlm.nih.gov/19457415/)
2. Brown LM. Helicobacter pylori: epidemiology and routes of transmission. *Epidemiol Rev.* 2000; 22: 283–297. PMID: [11218379](https://pubmed.ncbi.nlm.nih.gov/11218379/)
3. Buzas GM. Metabolic consequences of Helicobacter pylori infection and eradication. *World J Gastroenterol.* 2014; 20: 5226–5234. doi: [10.3748/wjg.v20.i18.5226](https://doi.org/10.3748/wjg.v20.i18.5226) PMID: [24833852](https://pubmed.ncbi.nlm.nih.gov/24833852/)
4. Jeon CH, Haan MN, Cheng C, Clayton ER, Mayeda ER, Miller JW et al. Helicobacter pylori infection is associated with an increased rate of diabetes. *Diabetes Care.* 2012; 35: 520–525. doi: [10.2337/dc11-1043](https://doi.org/10.2337/dc11-1043) PMID: [22279028](https://pubmed.ncbi.nlm.nih.gov/22279028/)
5. Gasbarrini A, Ojetti V, Pitocco D, De Luca A, Franceschi F, Candelli M et al. Helicobacter pylori infection in patients affected by insulin-dependent diabetes mellitus. *Eur J Gastroenterol Hepatol.* 1998; 10: 469–472. PMID: [9855061](https://pubmed.ncbi.nlm.nih.gov/9855061/)
6. So WY, Tong PC, Ko GT, Ma RC, Ozaki R, Kong AP et al. Low plasma adiponectin level, white blood cell count and Helicobacter pylori titre independently predict abnormal pancreatic beta-cell function. *Diabetes Res Clin Pract.* 2009; 86: 89–95. doi: [10.1016/j.diabres.2009.08.010](https://doi.org/10.1016/j.diabres.2009.08.010) PMID: [19747747](https://pubmed.ncbi.nlm.nih.gov/19747747/)
7. Lutsey PL, Pankow JS, Bertoni AG, Szklo M, Folsom AR. Serological evidence of infections and type 2 diabetes: the Multi-Ethnic Study of Atherosclerosis. *Diabet Med.* 2009; 26: 149–152. doi: [10.1111/j.1464-5491.2008.02632.x](https://doi.org/10.1111/j.1464-5491.2008.02632.x) PMID: [19236617](https://pubmed.ncbi.nlm.nih.gov/19236617/)
8. Xia HH, Talley NJ, Kam EP, Young LJ, Hammer J, Horowitz M. Helicobacter pylori infection is not associated with diabetes mellitus, nor with upper gastrointestinal symptoms in diabetes mellitus. *Am J Gastroenterol.* 2001; 96: 1039–1046. doi: [10.1111/j.1572-0241.2001.03604.x](https://doi.org/10.1111/j.1572-0241.2001.03604.x) PMID: [11316144](https://pubmed.ncbi.nlm.nih.gov/11316144/)

9. Osawa H, Nakazato M, Date Y, Kita H, Ohnishi H, Ueno H et al. Impaired production of gastric ghrelin in chronic gastritis associated with Helicobacter pylori. *J Clin Endocrinol Metab.* 2005; 90: 10–6. doi: [10.1210/jc.2004-1330](https://doi.org/10.1210/jc.2004-1330) PMID: [15483107](https://pubmed.ncbi.nlm.nih.gov/15483107/)
10. Francesco F, Tortora A, Di Rienzo T, D'Angelo G, Ianiro G, Scafaderi F et al. Role of Helicobacter pylori infection on nutrition and metabolism. *World J Gastroenterol.* 2014; 20: 12809–12817. doi: [10.3748/wjg.v20.i36.12809](https://doi.org/10.3748/wjg.v20.i36.12809) PMID: [25278679](https://pubmed.ncbi.nlm.nih.gov/25278679/)
11. Kamada T, Hata J, Kusunoki H, Ito M, Tanaka S, Kawamura Y et al. Eradication of Helicobacter pylori increases the incidence of hyperlipidaemia and obesity in peptic ulcer patients. *Dig Liver Dis.* 2005; 37: 39–43. doi: [10.1016/j.dld.2004.07.017](https://doi.org/10.1016/j.dld.2004.07.017) PMID: [15702858](https://pubmed.ncbi.nlm.nih.gov/15702858/)
12. Erim T, Cruz-Correa MR, Szomstein S, Velis E, Rosenthal R. Prevalence of Helicobacter pylori seropositivity among patients undergoing bariatric surgery: a preliminary study. *World J Surg.* 2008; 32: 2021–2025. doi: [10.1007/s00268-008-9608-7](https://doi.org/10.1007/s00268-008-9608-7) PMID: [18581170](https://pubmed.ncbi.nlm.nih.gov/18581170/)
13. Waluga M, Kukla M, Zorniak M, Bacik A, Koluiski R. From the stomach to other organs: Helicobacter pylori and the liver. *World J Hepatol.* 2015; 7: 2136–2146. doi: [10.4254/wjh.v7.i18.2136](https://doi.org/10.4254/wjh.v7.i18.2136) PMID: [26328025](https://pubmed.ncbi.nlm.nih.gov/26328025/)
14. Goo MJ, Ki MR, Lee HR, Yang HJ, Yuan DW, Hong IH et al. Helicobacter pylori promotes hepatic fibrosis in the animal model. *Lab Invest.* 2009; 89: 1291–1303. doi: [10.1038/abinvest.2009.90](https://doi.org/10.1038/abinvest.2009.90) PMID: [19736546](https://pubmed.ncbi.nlm.nih.gov/19736546/)
15. Fort JM, Vilallonga R, Lecube A, Gonzalez O, Caubet E, Mesa J et al. Bariatric surgery outcomes in a European Centre of Excellence (CoE). *Obes Surg.* 2013; 23: 1324–1332. doi: [10.1007/s11695-013-0980-5](https://doi.org/10.1007/s11695-013-0980-5) PMID: [23645480](https://pubmed.ncbi.nlm.nih.gov/23645480/)
16. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia.* 1985; 28: 412–419. PMID: [3899825](https://pubmed.ncbi.nlm.nih.gov/3899825/)
17. American Diabetes Association. Diagnosis and Classification of Diabetes Mellitus. *Diabetes Care* 2005; 28: S37–S42. American Diabetes Association. Diagnosis and Classification of Diabetes Mellitus. *Diabetes Care* 2005; 28: S37–S42. PMID: [15618111](https://pubmed.ncbi.nlm.nih.gov/15618111/)
18. Brunt EM, Janney CG, Di Bisceglie AM, Neuschwander-Tetri BA, Bacon BR. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. *Am J Gastroenterol.* 1999; 94: 2467–2474. doi: [10.1111/j.1572-0241.1999.01377.x](https://doi.org/10.1111/j.1572-0241.1999.01377.x) PMID: [10484010](https://pubmed.ncbi.nlm.nih.gov/10484010/)
19. Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, Ferrell LD, Liu YC, Torbenson MS, Unalp-Arida A, Yeh M, McCullough AJ, Sanyal AJ. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. *Hepatology.* 2005; 41: 1313–1321 doi: [10.1002/hep.20701](https://doi.org/10.1002/hep.20701) PMID: [15915461](https://pubmed.ncbi.nlm.nih.gov/15915461/)
20. Neuschwander-Tetri BA, Caldwell SH. Nonalcoholic steatohepatitis: summary of an AASLD Single Topic Conference. *Hepatology.* 2003; 37: 1202–1219. doi: [10.1053/jhep.2003.50193](https://doi.org/10.1053/jhep.2003.50193) PMID: [12717402](https://pubmed.ncbi.nlm.nih.gov/12717402/)
21. Polyzos SA, Kountouras J, Zavos C, Deretzi GI. The association between Helicobacter pylori infection and insulin resistance: a systematic review. *Helicobacter.* 2011; 16: 79–88. doi: [10.1111/j.1523-5378.2011.00822.x](https://doi.org/10.1111/j.1523-5378.2011.00822.x) PMID: [21435084](https://pubmed.ncbi.nlm.nih.gov/21435084/)
22. Gunji T, Matsuhashi N, Sato H, Fujibayashi K, Okumura M, Sasabe N et al. Helicobacter pylori infection significantly increases insulin resistance in the asymptomatic Japanese population. *Helicobacter.* 2009; 14: 144–150. doi: [10.1111/j.1523-5378.2009.00705.x](https://doi.org/10.1111/j.1523-5378.2009.00705.x) PMID: [19751440](https://pubmed.ncbi.nlm.nih.gov/19751440/)
23. Marrollo M, Latella G, Melideo D, Storelli E, Iannarelli R, Stornelli P et al. Increased prevalence of Helicobacter pylori in patients with diabetes mellitus. *Dig Liver Dis.* 2001; 33: 21–29. PMID: [11303971](https://pubmed.ncbi.nlm.nih.gov/11303971/)
24. Talebi-Taher M, Mashayekhi M, Hashemi MH, Bahrani V. Helicobacter pylori in diabetic and non-diabetic patients with dyspepsia. *Acta Med Iran.* 2012; 50: 315–318. PMID: [22837084](https://pubmed.ncbi.nlm.nih.gov/22837084/)
25. Shin DW, Kwon HT, Kang JM, Park JH, Choi HC, Park MS et al. Association between metabolic syndrome and Helicobacter pylori infection diagnosed by histologic status and serological status. *J Clin Gastroenterol.* 2012; 46: 840–845. doi: [10.1097/MCG.0b013e3182522477](https://doi.org/10.1097/MCG.0b013e3182522477) PMID: [23064216](https://pubmed.ncbi.nlm.nih.gov/23064216/)
26. Park SH, Jeon WK, Kim SH, Kim HJ, Park DI, Cho YK, Sung IK, Sohn CI, Kim BI, Keum DK. Helicobacter pylori eradication has no effect on metabolic and inflammatory parameters. *J Natl Med Assoc.* 2005; 97: 508–513. PMID: [15868771](https://pubmed.ncbi.nlm.nih.gov/15868771/)
27. Gen R, Demir M, Ataseven H. Effect of Helicobacter pylori eradication on insulin resistance, serum lipids and low-grade inflammation. *South Med J.* 2010; 103: 190–196. doi: [10.1097/SMJ.0b013e3181cf373f](https://doi.org/10.1097/SMJ.0b013e3181cf373f) PMID: [20134372](https://pubmed.ncbi.nlm.nih.gov/20134372/)
28. Abenavoli L, Milic N, Masarone M, Persico M. Association between non-alcoholic fatty liver disease, insulin resistance and Helicobacter pylori. *Med Hypotheses.* 2013; 81: 913–915 doi: [10.1016/j.mehy.2013.08.011](https://doi.org/10.1016/j.mehy.2013.08.011) PMID: [24011768](https://pubmed.ncbi.nlm.nih.gov/24011768/)

29. Chen Y, Blaser MJ. Association between gastric Helicobacter pylori colonization and glycated hemoglobin levels. *J Infect Dis.* 2012; 205: 1195–1202. doi: [10.1093/infdis/jis106](https://doi.org/10.1093/infdis/jis106) PMID: [22427676](https://pubmed.ncbi.nlm.nih.gov/22427676/)
30. Zhou X, Zhang C, Wu J, Zhang G. Association between Helicobacter pylori infection and diabetes mellitus: a meta-analysis of observational studies. *Diabetes Res Clin Pract.* 2013; 99: 200–208. doi: [10.1016/j.diabres.2012.11.012](https://doi.org/10.1016/j.diabres.2012.11.012) PMID: [23395214](https://pubmed.ncbi.nlm.nih.gov/23395214/)
31. McColl KE. Clinical practice. Helicobacter pylori infection. *N Engl J Med.* 2010; 362: 1597–1604. doi: [10.1056/NEJMcp1001110](https://doi.org/10.1056/NEJMcp1001110) PMID: [20427808](https://pubmed.ncbi.nlm.nih.gov/20427808/)
32. Al-Akwaa AM. Prevalence of Helicobacter pylori infection in a group of morbidly obese Saudi patients undergoing bariatric surgery: a preliminary report. *Saudi J Gastroenterol.* 2010; 16: 264–267. doi: [10.4103/1319-3767.70610](https://doi.org/10.4103/1319-3767.70610) PMID: [20871190](https://pubmed.ncbi.nlm.nih.gov/20871190/)
33. Renshaw AA, Rabaza JR, Gonzalez AM, Verdeja JC. Helicobacter pylori infection in patients undergoing gastric bypass surgery for morbid obesity. *Obes Surg.* 2001; 11: 281–283. doi: [10.1381/096089201321336601](https://doi.org/10.1381/096089201321336601) PMID: [11433901](https://pubmed.ncbi.nlm.nih.gov/11433901/)
34. Ramaswamy A, Lin E, Ramsaw BJ, Smith CD. Early effects of Helicobacter pylori infection in patients undergoing bariatric surgery. *Arch Surg.* 2004; 139: 1094–1096. doi: [10.1001/archsurg.139.10.1094](https://doi.org/10.1001/archsurg.139.10.1094) PMID: [15492150](https://pubmed.ncbi.nlm.nih.gov/15492150/)
35. Papasavas PK, Gange DJ, Donnelly PE, Salgado J, Urbandt JE, Burton KK et al. Prevalence of Helicobacter pylori infection and value of preoperative testing and treatment in patients undergoing laparoscopic Roux-en-Y gastric bypass. *Surg Obes Relat Dis.* 2008; 4: 383–388. doi: [10.1016/j.soard.2007.08.014](https://doi.org/10.1016/j.soard.2007.08.014) PMID: [17974495](https://pubmed.ncbi.nlm.nih.gov/17974495/)
36. Sumida Y, Kanemasa K, Imai S, Mori K, Tanaka S, Shimokobe H et al. Helicobacter pylori infection might have a potential role in hepatocyte ballooning in nonalcoholic fatty liver disease. *J Gastroenterol.* 2015; 50: 996–1004.
37. Li M, Shen Z, Li YM. Potential role of Helicobacter pylori infection in nonalcoholic fatty liver disease. *World J Gastroenterol.* 2013; 19: 7024–7031. doi: [10.3748/wjg.v19.i41.7024](https://doi.org/10.3748/wjg.v19.i41.7024) PMID: [24222944](https://pubmed.ncbi.nlm.nih.gov/24222944/)
38. Polyzos SA, Kountouras J, Papatheodorou A, Patsiaoura K, Katsiki E, Zafeiriadou E et al. Helicobacter pylori infection in patients with nonalcoholic fatty liver disease. *Metabolism* 2013; 62: 121–126. doi: [10.1016/j.metabol.2012.06.007](https://doi.org/10.1016/j.metabol.2012.06.007) PMID: [22841522](https://pubmed.ncbi.nlm.nih.gov/22841522/)
39. Miele L, Valenza V, La Torre G, Montalto M, Cammarota G, Ricci R et al. Increased intestinal permeability and tight junction alterations in nonalcoholic fatty liver disease. *Hepatology.* 2009; 49: 1877–1887. doi: [10.1002/hep.22848](https://doi.org/10.1002/hep.22848) PMID: [19291785](https://pubmed.ncbi.nlm.nih.gov/19291785/)
40. Okushin K, Takahashi Y, Yamamichi N, Shimamoto T, Enooku K, Fujinaga H et al. Helicobacter pylori infection is not associated with fatty liver disease including non-alcoholic fatty liver disease: a large-scale cross sectional study in Japan. *BMC Gastroenterol.* 2015; 15: 25. doi: [10.1186/s12876-015-0247-9](https://doi.org/10.1186/s12876-015-0247-9) PMID: [25880912](https://pubmed.ncbi.nlm.nih.gov/25880912/)
41. Bedogni G, Bellentani S, Miglioli L, Masutti F, Passalacqua M, Castiglione A et al. The Fatty Liver Index: a simple and accurate predictor of hepatic steatosis in the general population. *BMC Gastroenterol.* 2006; 6: 33. doi: [10.1186/1471-230X-6-33](https://doi.org/10.1186/1471-230X-6-33) PMID: [17081293](https://pubmed.ncbi.nlm.nih.gov/17081293/)
42. Lee JH, Kim D, Kim HJ, Lee CH, Yang JI, Kim W et al. Hepatic steatosis index: a simple screening tool reflecting nonalcoholic fatty liver disease. *Dig Liver Dis.* 2010; 42: 503–508. doi: [10.1016/j.dld.2009.08.002](https://doi.org/10.1016/j.dld.2009.08.002) PMID: [19766548](https://pubmed.ncbi.nlm.nih.gov/19766548/)
43. Estep J, Michael et al. Assessment of Fatty Liver Index (FLI) for the Diagnosis of Non-Alcoholic Fatty Liver Disease (NAFLD) in Morbidly Obese Patients. *Gastroenterology.* 2013; 144: S–125.
44. Hansen PS, Go MF, Varming K, Andersen LP, Genta RM, Graham DY et al. Proinflammatory activation of neutrophils and monocytes by Helicobacter pylori in patients with different clinical presentations. *Infect Immun.* 1999; 67: 3171–3174. PMID: [10338540](https://pubmed.ncbi.nlm.nih.gov/10338540/)
45. Aslan M, Horoz M, Nazligul Y, Bolukbas C, Bolukbas FF, Selek S et al. Insulin resistance in Helicobacter pylori infection and its association with oxidative stress. *World J Gastroenterol.* 2006; 12: 6865–6868. doi: [10.3748/wjg.v12.i42.6865](https://doi.org/10.3748/wjg.v12.i42.6865) PMID: [17106938](https://pubmed.ncbi.nlm.nih.gov/17106938/)
46. Hamed SA, Amine NF, Galal GM, Helal SR, Tag El-Din LM, Shawky OA et al. Vascular risks and complications in diabetes mellitus: the role of helicobacter pylori infection. *J Stroke Cerebrovasc Dis.* 2008; 17: 86–94. doi: [10.1016/j.jstrokecerebrovasdis.2007.10.006](https://doi.org/10.1016/j.jstrokecerebrovasdis.2007.10.006) PMID: [18346651](https://pubmed.ncbi.nlm.nih.gov/18346651/)
47. Kountouras J, Polyzos SA, Deretzi G, Katsinelos P, Kyriakou P. Helicobacter pylori infection and the risk for cardiovascular disease. *Eur J Intern Med.* 2011; 22: e146–47. doi: [10.1016/j.ejim.2011.07.013](https://doi.org/10.1016/j.ejim.2011.07.013) PMID: [22075304](https://pubmed.ncbi.nlm.nih.gov/22075304/)