ORIGINAL ARTICLE

Comparison of Platelet Counts between H. Pylori infected and non-infected individuals

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ABSTRACT

Background: Immune Thrombocytopenic Purpura (ITP) may be associated with Helicobacter Pylori (H. Pylori) infection of stomach, in which case its eradication often, but not invariably, leads to improvement in platelets counts. There is relative dearth of studies comparing platelets counts in H. Pylori infected and non-infected individuals.

Methods: We compared platelet counts in 200 H. Pylori infected and 200 non-infected individuals during upper oesophago-gastroduodenoscopy in patients presenting to GI clinic with Dyspepsia. H. Pylori infection was documented with histopathological examination of stomach biopsies.

Results: H. Pylori positive and negative patients were comparable in age (35.89±10.73 and 34.93±11.31 respectively, P value: 0.3845). Twenty (10%) of infected patients had platelet count below $1.5 \times 10^6/mm^3$ and 35(17.5%) had counts between 1.5 and $2 \times 10^6/mm^3$ (Vs 1% and 5% respectively in non-infected group, P Value: 0.0001). Moreover, all 20 patients with thrombocytopenia were above 45 years of age and those with lowest counts were the oldest (in 6^{th} or 7^{th} decade).

Conclusion: Patients with H. pylori infection of stomach have lower platelet counts when compared with non-infected individuals after exclusion of secondary causes of thrombocytopenia. Most of patients with lower platelets counts are in their 5^{th} to 7^{th} decades.

Keywords: Helicobacter Pylori, Platelets, Thrombocytopenia, ITP, Older-age

INTRODUCTION

Chronicity of many bacterial or viral infections is associated with development of many a misdirected immune phenomenon (e.g., HIV, Tuberculosis, Sub-acute bacterial endocarditis)[1]. It would be no surprise if high prevalence and chronicity of H. Pylori infection, especially in developing countries, may be implicated in many such immune diseases[2]. A higher incidence of Helicobacter Pylori (H. Pylori) infection of gastric mucosa in patients with Immune Thrombocytopenic Purpura (ITP) was first reported by Gasbarrini A et al in 1988[3] They reported that platelet counts increased in all of 8 H. Pylori-infected patients with ITP who were treated with a regimen to eradicate H. Pylori, while the platelet counts were unchanged in 3 H. Pylori infected patients who did not receive the regimen. Since then, many studies across globe have reported improvement of platelet counts in ITP patients with the treatment H. Pylori, including those with refractory[4,5,6] while in certain studies, no favorable effect was observed[7,8]. The contrariety may be due to different strains of H. Pylori in different geographic regions.[9]Many factors related to infective organisms (e.g., Cag A protein), variable distributions of these factors in different geographical locations, chronicity of infection and host factors, have been proposed to account for both the variations in incidence of chronic ITP in infected host and its response to eradication of infection. The question as to if any favorable response of platelets in chronic ITP is due to H. Pylori eradication itself or the effects of drugs given for eradication, has been addressed in a metanalysis of studies from various geographical locations[9].

It has been observed that Patients with ITP and concurrent H. Pylori infection were older than non-infected ITP patients[4]. It possibly may take years for the misdirected immune reaction to lead to ITP. The answer to this question would be a long follow-up study of infected patients for the development of ITP. Such a prolonged follow-up study in infected patients may raise ethical question even though H. Pylori eradication therapy may not be indicated in asymptomatic infected patients. We looked for a comparison of platelet count in infected and non-infected patients with H. Pylori coming to the gastrointestinal (GI) endoscopy suite for upper GI endoscopy.We also looked for prevalence of asymptomatic thrombocytopenia in infected patients.

METHODS

Study population: Between March 2014 and December 2015, two hundred consecutive patients for each group (infected and non-infected with H.

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Pylori) presenting to endoscopy suite of our gastroenterology unit for upper GI endoscopy were recruited for this study. Primary complaint of all the patient was dyspepsia. Patient could be of any age beyond 12 years and of either gender. Exclusion criteria were; 1) gastrointestinal bleeding, 2) gastric or duodenal ulcers, 3) gastrointestinal malignancy, 4) portal hypertension, 5) chronic liver disease, 6) chronic kidney disease, 7) patients on nonsteroidal anti-inflammatory drugs, proton pump inhibitors or any cytotoxic treatment and 8) patient previously treated for H. Pylori infection. The study was conducted in accordance with good clinical practice guidelines and was approved by local Ethical Review Committee. All patients signed informed consent before being included in study.

Diagnosis of Secondary Thrombocytopenia: Potential etiologies to be considered when a low platelet count is found are numerous and there is no confirmatory test for its attribution to H. Pylori infection, if present. Any lower platelet counts in an H. Pylori infected patient, therefore, largely remains a diagnosis of exclusion. A detailed history, physical examination and investigations indicated thereafter, were carried out for this purpose on all patients of both groups to look for potential factors affecting platelet counts. Presence of liver disease, abnormal liver function tests and portal hypertension (clinical and ultrasonographic presence of ascites, portal vein diameter and presence of esophageal varices), renal impairment, presence of sepsis and fever, fibrinogen degradation products (if indicated), prothrombin time, unexplained proteinuria, Antinuclear Antibodies and Anti-double stranded Antibodies (if indicated), HIV status (if indicated), were few of the important pre-induction evaluations. Potentially any drug may be implicated in thrombocytopenia. We did not exclude patient taking commonly prescribed drugs e.g. anti-hypertensive and anti-diabetic medications. All the drugs being taken currently or within last month, were evaluated for their potential to induce thrombocytopenia by literature search. Most of the drug induced thrombocytopenia are acute and particular attention was paid to medications started or taken during last month.

Diagnosis of H. Pylori: Two antral biopsy specimen were taken and collected in formalin for histological examination, stained with Giemsa, andevaluated by two pathologists who checked gastric biopsies and resected surgical specimens of stomach on daily basis. The sensitivity and specificity of antral specimens stained with Giemsa is 91% and 98% respectively, although admittedly, results are operator dependent. [10]Whole blood counts were determined by Sysmex and Beckman Coulter LH 780 autoanaylzers. Blood samples are collected in EDTA containing tubes for whole blood count and it was ensured that they reach laboratory in 150 minutes. All the blood samples with platelet counts below 150×10^9/L were confirmed in citrated blood samples.

RESULTS

Table 1 shows comparisons of ages between H. Pylori positive and negative patients and between male patients (H. Pylori positive and negative) and female patients (H. Pylori positive and negative). None of these comparison shows a significant age difference. Table 2 shows number and percentages of H. Pylori positive and negative patients in various ranges of platelet counts. As can be seen, number of infected patients in lower ranges of platelet counts is more in comparison to non-infected patients and the difference is statistically significant. Graph 1 depicts differences in mean platelet counts between H. Pylori positive and negative patients divided into age groups of 5 years. The differences in platelet counts become wider with every 5 years of increasing age.

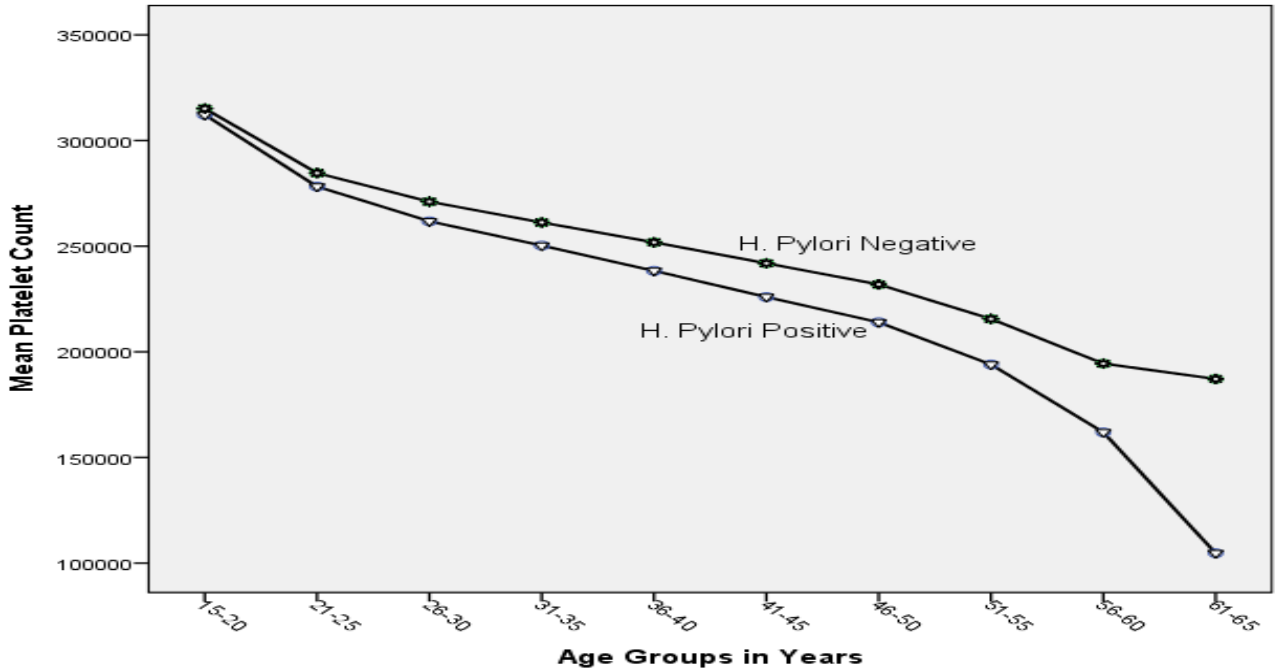
Table 1: Age comparison two group of patients; H. Pylori positive and H. Pylori negative

Characteristic	H. Pylori Positive	H. Pylori Negative	P Value
Age in years	35.89±10.73	34.93±11.31	0.3845
Age (males)	35.74±10.63	34.78±11.38	0.54
Age (females)	36.05±10.89	35.07±11.30	0.535

Table 2: Number of H. Pylori Positive and Negative patients in various ranges of platelets counts

Range of Platelet Count	H. Pylori Positive (n=200)	H. Pylori Negative (n=200)	p Value
Below 150×10^9/L	20(10%)	2(1%)	0.0001
150 - 200×10^9/L	35 (17.5%)	10 (5%)	
200 - 300×10^9/L	131 (65.5%)	171 (85.5%)	
Above 300×10^9/L	14(7%)	17 (8.5%)	

Graph 1 Comparison of mean platelet counts between H. Pylori positive and negative patients divided in age groups of 5 years



DISCUSSION

The study shows that blood platelets, in general, are lower in number in patients with H. Pylori infection of stomach compared to matched controls with significantly higher number of infected patients having platelets counts below the normal lower limit of 1.5×10^9/L.

Studies connecting H. Pylori infection with thrombocytopenia have mostly been done in patients with diagnosed ITP; Prevalence of H. Pylori in ITP, comparing this prevalence with that of general population in same area, response of platelets after H. Pylori eradicative therapy and host and bacterial factors possibly implicated in thrombocytopenia in ITP. There is dearth of studies looking at the prevalence of thrombocytopenia in H. Pylori infected patients

A literature search between 1950 and 2008, looking for effect of Anti-H. Pylori therapy trials in ITP patients given to both H. Pylori positive and negative patients, yielded 7 eligible studies (n = 222)[11]. A platelet count response was achieved following H. Pylori therapy in 65 (49.6%) of 131 infected patients, and none of the 44 uninfected patients. However, differing definitions of “success” limit the strength of this conclusion, and most studies were from Japan, where the prevalence of H. Pylori infection is high.

We find that after excluding secondary causes of thrombocytopenia, age is a major determinant of platelet count. Patients with ITP and concurrent H. Pylori infection have been found to be older than patients not infected.[4] It has been argued by the authors that incidence of H. Pylori infection increases with increasing age in H. Pylori prevalent regions. We also find that platelet counts in the lowest range were mostly in older population infected with H. Pylori. Another possible factor could be lower platelet counts in healthy older adults. We find a mean difference in platelet count of approximately 100×10^9/L between individuals in second and sixth decade. Analysis of 12,517 inhabitants of Sardinian geographic isolates found that a 10-year increase in age corresponded to a 9×10^9/L decrease in platelet count[12]. Very similar results were obtained in 7266 inhabitants of five additional geographic isolates located in different Italian areas[13] and in the cohort of the Moli-Sani Project including 24,318 subjects from 30 Molise cities and villages[14]. Finally, a recent study put together all data of subjects enrolled in the three population-based studies referenced above and concluded that age-related changes were actually very large: platelet count in old age was reduced by 35% in men and by 25% in women with respect to early infancy[15]. Thus, there is no longer any doubt that age is a major determinant of platelet count in healthy people.

The study most relevant to our study was by H Umit and EG Umit.[16] They compared mean platelet counts and mean platelet volume in H. Pylori positive

and negative patients. The data were collected in a retrospective manner in 4823 patients (H. Pylori positive = 1701, H. Pylori Negative=3122) with dyspeptic complaints who underwent an upper gastrointestinal endoscopy. They found that mean platelet count in H. Pylori positive and negative patients were 246381± 92225/mm^3 and 258135±89912/mm^3, respectively ($p<0.001$). Mean Platelet Volume (MPV) was higher in H. Pylori positive group (8.9±1.3 vs. 8.23±0.94, $p<0.001$). This difference was observed in both genders. They concluded that in patients with H. Pylori infection and normal platelet counts, it may be speculated that an ongoing and compensated platelet destruction-production process may be responsible for the increase in MPV. There are many causes of increased MPV e.g. younger and larger platelets (e.g., in ITP), possible congenital condition, role of platelets as cells of immunity like polymorph nuclear leukocytes, impairment of tissue oxygenation and nutrition, diabetes mellitus, myocardial infarct, ischemic shock, renal artery stenosis and hypertrophic dilated cardiomyopathy etc., as accepted by authors themselves. After ruling out presence of above causes the authors, however, considered increase in MPV in patients infected with H. Pylori to be due to younger larger platelets owing to on-gonging compensated platelet destruction and argued that increased MPV may possibly be forerunner of overt thrombocytopenia later. The authors, however, did not compared the platelet counts in younger and older patients nor gave any further details of platelet counts in the two groups, as done in our study. Our findings of lower platelet counts in H. Pylori infected patients in later age groups may suggest ITP to be a culminant of chronic, than acute,H. Pylori infection and possibly lends support to the assertion by H Umit, EG Umit that higher MPV in infected patient may ultimately lead to ITP, at least in a subset of patients.

We conclude that patients infected with H. Pylori have lower platelets counts when compared to non-infected age matched controls, with significantly higher number of infected patients having platelet count below the normal range. The difference in platelet counts between the two groups is more prominent with increasing age.

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