Gene Cell Tissue. 2015 July; 2(3): e27818. DOI: 10.17795/gct27818

Published online 2015 July 25. **Research Article**

Eradication of Gastric Infection by *Helicobacter pylori* and Chronic Idiopathic Thrombocytopenic Purpura Situation

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Received: March 1, 2015; **Revised:** May 20, 2015; **Accepted:** May 27, 2015

Background: Chronic idiopathic thrombocytopenic purpura (CITP) is an autoimmune disease with low platelet count and potentially bleeding tendency. Although autoimmune mechanisms play a crucial role in the disease yet in 20% of cases underlying diseases such as *Helicobacter pylori (HP)* infection can be involved.

Objectives: The aim of this study was to evaluate effects of *H. pylori* eradication therapy on platelet count in CITP patients with *H. pylori* infection.

Patients and Methods: During this clinical trial, performed from September 2011 to August 2012, twenty-three patients with CITP were divided in two groups, including one group with *H. pylori* infection and the other without infection. After determination of baseline platelet count in the two groups, patients with *H. pylori* infection underwent eradication therapy. Both groups were followed up for six months and platelet counts were checked at one-month intervals and finally, changes in platelet counts were statistically compared before and after treatment and also between the two groups.

Results: From a total of 23 patients with CITP, 43% (10) of the cases were *H. pylori* positive and after eradication therapy, eight patients (80%) were successfully treated. *Helicobacter pylori* eradication therapy causes a statistically significant difference between the mean platelet count before and after HP eradication therapy.

Conclusions: *Helicobacter pylori* infection has a significant effect on platelet count in patients with CITP and *H. pylori* eradication therapy should be considered as an important factor in platelet count recovery in CITP patients.

Keywords: Blood Platelets; Thrombocytopenia; *Helicobacter pylori*

1. Background

Chronic immune idiopathic thrombocytopenia (CITP) is an autoimmune disease that is characterized by shortened intravascular survival of platelets due to autoantibodies-mediated destruction. These antiplatelet autoantibodies that are formed as a result of immune system stimulation against platelet membrane proteins, are mostly directed against platelet glycoprotein (GP) IIb-IIIa and/or GPIb-IX complexes (1-3). These autoantibodies bind to platelets and megakaryocytes and result in platelet destruction by reticuloendotelial cells in the spleen and liver and to a lesser extend in bone marrow, and also impair the production of platelets by interference in megakaryopoiesis (1, 2). In addition to autoantibodies, complement mediated lysis and direct T-cell cytotoxicity are also involved in pathogenesis of CITP (2). The main clinical features of the disease are low platelet count and bleeding tendency (1, 3). Although idiopathic thrombocytopenia (ITP) results from an autoimmune mechanism yet in 20% of cases the underlying disorders play an important role in pathogenesis of the disease. Several studies have focused on the association between *H. pylori* infection, as one of these underlying disorders, and chronic ITP (4).

Helicobacter pylori is a gram-negative microaerophilic bacteria that colonizes the stomach of 50% to 80% of the world population (5). It is involved in the pathogenesis of various diseases such as gastroduodenal diseases, including gastric and duodenal ulcers, chronic active gastritis, and mucosa-associated lymphoid tissue (MALT) lymphoma. In addition, *H. pylori* is implicated in extragastric disorders such as cardiovascular, respiratory, neurological and autoimmune diseases including pernicious anemia, rheumatoid arthritis, autoimmune thyroiditis, idiopathic thrombocytopenic purpura etc. (6). Although *H. pylori* infection is a possible cause of CITP, yet the exact pathological mechanism is obscure and needs further research. It seems that *H. pylori* may trigger an autoimmune event (5). Some studies have shown that eradication of *H. pylori* infection leads to platelet recovery in CITP patients while others have failed to show the same beneficial effects (7, 8).

2. Objectives

The purpose of the present study was to determine the efficacy of *H. pylori* eradication on improvement of platelet counts in subjects with chronic ITP.

3. Patients and Methods

3.1. Patients

This study was conducted on 24 children under 20 years old with chronic ITP, admitted to the pediatric ward of Ali-ebn Abitaleb hospital from September 2011 to August 2012. Idiopathic Thrombocytopenia was defined based on platelet counts below 100×10^3 μL (lasting more than six months without identified causes) according to the American society of hematology (ASH) guidelines, and was confirmed by bone marrow examinations at the time of diagnosis (9). All patients received intravenous immunoglobulin (IVIG), yet none of them received anti-D immunoglobulin. Cases who had previously received *H. pylori* eradication treatment or were using corticosteroid at a dosage of more than 0.5 mg/kg/day at baseline, and during the study period, subjects with secondary ITP and patients with serious illness including cardiac, hepatic and renal diseases were excluded from the study. One out of 24 patients died due to cerebral hemorrhage during the study and therefore was excluded from the study. A written consent was obtained from all parents for participation of their child in this study. This study was confirmed by the children and adolescent health research center.

3.2. Diagnosis and Treatment

At the baseline, complete blood count (CBC) was measured in all 23 patients. Analysis of *H. pylori* infection was carried out by the *H. pylori* stool antigen (HpSA) using the enzyme-linked immunosorbent assay (ELISA) and 13 C-urea breath test (UBT). Thirteen C-urea breath test (UBT) was performed as following: After an overnight fast, breath samples were collected before and 30 minute after ingestion of 13 C-urea dissolved in 100 mL water. Doses of 13 C-urea were 50 mg for children under six years old and 75 mg for older children. Detecting of 13C in breath CO_2 was determined by Isotope Ratio Mass Spectrometry. A delta-over-baseline value of > 4% was considered positive (10). Four weeks before the test the use of antibiotics and proton pomp inhibitors were discontinued. The HPSA test was performed in all patients and those with positive HPSA test were positive for 13 C-UBT too.

In study, patients with confirmed *H. pylori* infection was treated with triple eradication therapy including clarithromycin 500 mg twice daily, amoxicillin 1000 mg twice daily and proton pump inhibitors (PPI) 40 mg twice daily for one or two weeks (7). Eradication was assessed by UBT more than four weeks after the eradication therapy.

3.3. Assessment of Response and Follow-Up

Platelet count was monitored once a month until six months after the end of eradication therapy and was compared with baseline counts. Some patients were checked more frequently depending on their platelet counts and clinical situation. Response to treatment was defined as a complete response (CR) if the platelet count was above 100×10^3 μL or when it reached the normal range (150×103 μL), and partial response (PR) was when the increase in platelet count was between 50 and 100×10^3 μL. Non-responders were those with an increase in platelet count of less than 50×10^3 μL or no increase in platelet count following eradication. In addition to *H. pylori* positive patients, the platelet counts were monitored at the mentioned intervals for uninfected individuals. The platelet count changes were compared for the following three groups; *H. pylori* infected patients who were successfully treated, *H. pylori* infected patients who were not successfully treated and uninfected individuals who had not received therapy. Other parameters including age, sex, baseline platelet count, previous splenectomy and present corticosteroid therapy were also compared between these groups.

3.4. Statistical Analysis

The data was analyzed using the SPSS software. T-student test was used for comparisons of mean values between the two groups and in cases with non-normal distribution, Mann-Whitney test was used. P values of < 0.05 were considered statistically significant. Kolmogorov-Smirnov Test and χ^2 test were also used in this study.

4. Results

Out of the 24 patients with CITP, 14 (58%) were boys and 10 (42%) were girls, with the mean age of 10 ± 0.5 years (age range between 3.5 and 20 years). One boy died due to cerebral hemorrhage and was excluded from the study. Of the 23 remaining subjects, *H. pylori* infection was detected in 10 (43.5%) patients with mean age (mean ± standard deviation (SD)) of 9.5 ± 4.7 and 13 subjects were uninfected (mean ± SD: 10.2 ± 5). The comparison between the baseline clinical characteristics of *H. pylori* positive and negative CITP patients is shown in Table 1.

Successful eradication was achieved in eight out of ten *H. pylori* infected patients and also two out of eight patients (25%) achieved a partial response. There was a significant increase in platelet count during the six months follow-up ($P < 0.001$) compared with the baseline platelet count. The difference between the mean platelet count ± SD before and after HP eradication therapy was statistically significant ($56.2 \pm 22.2 \times 10^3$ μL versus $233 \pm 85.6 \times 10^3$ μL). These patients also had a significant increase in their platelet counts compared with those in whom *H. pylori* was not successfully eradicated ($P < 0.05$) or untreated patients ($P < 0.05$) (Table 2).

Table 1. Comparison Between Some Characteristics of *Helicobacter pylori* Infected and Non-Infected Patients With Chronic Idiopathic Thrombocytopenic Purpura

Characteristics	*H. pylori* Infected Patients (n = 10)	*H. pylori*-Non Infected Patients (n = 13)	df	Statistical Value	P Value
Age, y [a]	9.5 ± 4.7	10.2 ± 5	21	T = -0.337	0.74
Gender			1	$\chi^2 = 0.87$	0.7
Female	4	6			
Male	6	7			
Platelet count [a]**, × 103 μL**	54.9 ± 22.2	56 ± 32.2			0.93
Prednisolone therapy [b]	5 (50)	6 (46.2)	1	$\chi^2 = 0.034$	0.86
Previous splenectomy [b]	1 (10)	1 (7.7)	19	T = 0.014	0.85

[a] The values are presented as mean ± SD.
[b] The values are presented as No. (%).

Table 2. Comparison Between Baselines and Secondary (After Six Months) Platelet Counts in Three Groups of Successfully Treated Patients With Chronic Idiopathic Thrombocytopenic Purpura (CITP), Unsuccessfully Treated Patients and Untreated CITP Patients

Groups	Baseline Platelet [a]	Secondary Platelet [a]	df	Statistical Value	P Value
***H. pylori* infected patients with response (n = 8)** [b]	56.2 ± 2.2	233 ± 85.6	7	T = -6.1	< 0.001
***H. pylori* infected patients no response (n = 2)** [b]	50 ± 49.5	52 ± 50.9	1	T = -2.2	0.276
Untreated patients (n = 13) [b]	56 ± 32.2	56 ± 27.8	12	T = -0.023	< 0.98
P values					
P1	0.82	< 0.05			
P2	0.98	= < 0.05			

[a] Count × 10^3 μL.
[b] The values are presented as mean ± SD.

In patients with successful eradication of *H. pylori*, during the first three months of eradication therapy, platelet counts gradually increased until no increase in platelet counts was observed. On the other hand, non-responder patients to *H. pylori* eradication therapy did not show statistically significant increase in platelet counts during the entire follow up period of six months.

5. Discussion

Chronic Idiopathic thrombocytopenic purpura is an autoimmune disease characterized by low platelet count, due to destruction and impairment in the production of platelets mediated by auto-antibodies (1-3). In addition to the autoimmune phenomena, infectious agents such as *H. pylori* are involved in the pathogenesis of CITP, yet the exact role of these agents remains controversial (2-5). This study hypothesized that in HP infected chronic ITP patients, *H. pylori* eradication therapy may be associated with platelet recovery. The results of this study partly confirmed this theory since from ten HP infected patients (43.5%), HP was eradicated in eight (80%) and two (20%) were non-responder. We also found a significant increase in platelet count of eight patients during six months of follow-up (P < 0.001) compared with the baseline platelet count. This finding was consistent with the findings of other studies (11, 12). In the study of Ferrara et al. (11) on 24 patients with CITP, eight individuals (33.3%) were infected with *H. pylori* and eradication treatment was successfully achieved in six out of eight patients with significant increases in platelet count in all six cases. Our findings as well as that of Ferrara et al. (11) highlight the effect of *H. pylori* infection on platelet count in CITP as a basic cause or exacerbation factor. Another study by Sato et al. in Japan was performed on 53 patients with CITP (12). In this study prevalence of *H. pylori* infection among CITP patients was 73% (39 patients); eradication of HP in 32 cases led to suitable response in 27 patients (12). Although the above articles are in line with our study, some reports did not show any platelet recovery in the *H. pylori* treatment group compared to the control group (13, 14). The findings of these studies that were not consistent with our findings, suggest that eradication of co-existing *H. pylori* infection is not associated with platelet recovery in childhood chronic ITP (13, 14). More research with more subjects and long-term follow up in different geographical regions is highly recommended. Although a considerable number of studies were conducted investigating the effect of *H. pylori* infection treatment on platelet count in CITP patients yet the effect of ethnicity on treatment should al-

ways be considered when comparing the results of different studies. This study was performed on Iranian patients, which obviously have their own genetic properties that can affect treatment results. Prevalence of *H. pylori* infection in our study was 43.5%. Previous studies on patients with CITP have demonstrated a different prevalence rate of *H. pylori* infection among different countries: 74% in Japan (12), 12.9% in Iran (15), 20% in another study from Japan (16) and no cases in Finland (17). This variation in prevalence rates among subjects might be due to differences in acquisition of *H. pylori* infection in the general population in different countries; as repots are higher in low socioeconomic countries compared to developed countries.

There was no significant change in platelet count in our uninfected patients who had not received HP eradication therapy (P = 0.98). Six out of eight patients (75%) achieved a good response that was demonstrated by the considerable and statistically significant increase in platelet counts after eradication therapy compared with baseline platelet counts (P < 0.001). We also observed 2.8 patients (25%) with partial response after treatment. This finding is similar to the study of Tag et al. Their study performed in Korea during 2009 revealed that from 23 patients with *H. pylori* infection, 11 (44%) exhibited a complete response (CR) to *H. pylori* eradication therapy, six (24%) partial response (PR), and eight (32%) were nonresponsive (NR) (7). Russo et al. (8) in Italy in 2010 also described the efficacy of *H. pylori* eradication in increasing platelet count in CITP patients. Their study was done on 244 CITP patients and revealed that from 50 (20%) patients with *H. pylori* infection, 37 received eradication therapy and were completely followed-up for six months. In 89% of these cases, eradication therapy was successful and platelet recovery was demonstrated in 39% of patients after eradication (8). In the present study responders who were defined as patients with successful treatment of *H. pylori* showed a significant platelet recovery within six months after the eradication therapy. Although longer follow up duration of platelet count was better for good judgment, yet it seems that six months of monitoring was adequate to find the association. According to the results of this study and other similar studies, *H. pylori* infection should be considered as a potential cause or an aggravating factor of thrombocytopenia in chronic ITP; eradication therapy in positive cases of *H. pylori* might have a beneficial effect on platelet recovery. However, a larger scale of randomized controlled trials is required for confirming our findings.

Acknowledgements

We acknowledge the Zahedan University of Medical Sciences and the children and adolescent health research center for their assistance in this study.

Authors' Contributions

Majid Naderi and Turan Shahraki performed the clinical assessment while Akbar Dorgalaleh and Shadi Tabibian confirmed the laboratory findings as well as the correlation between data.

Funding/Support

This study was supported by the Zahedan University of Medical Sciences, Zahedan, IR Iran.

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