ORIGINAL ARTICLE

Effect of Helicobacter Pylori Infection on Hemoglobin, MCV, and Vitamin B_{12}

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ABSTRACT

Objective: The objective of this study was to elucidate effect of *H. pylori* infection on serum vitamin B_{12} levels and to find out association of this effect with changes in red blood indices in our local population.

Subjects and methods: A total number of 90 subjects were included in the study. They were divided into group A (30 subjects with gastric symptoms and *H. pylori* infection), group B (30 subjects with gastric symptoms but without *H. pylori* infection), and group C (30 normal healthy age and sex matched subjects). The subjects were between 15-60 years of age. *H. pylori* infection was considered positive on the basis of positive serology, rapid urease test and histopathological examination. Hemoglobin and MCV were estimated by automated hematology analyzer. Serum vitamin B_{12} levels were measured by chemilumisecne technique. Intrinsic factor antibodies were determined by ELISA.

Results: There was no significant difference (p value > 0.05) of hemoglobin concentration between groups A, B, and C. MCV values did not show significant difference (p value > 0.05) between groups A, B, and C. The difference of serum vitamin B_{12} among groups A, B, and C was also non significant (p value > 0.05).

Conclusion: *H. pylori* infection was not found to cause any significant changes in hemoglobin and MCV. This infection did not produce significant effect on vitamin B_{12} levels in the subjects studied.

Key words: MCV- Mean corpuscular volume, *H. pylori- Helicobacter pylori*

INTRODUCTION

Red blood cells are an important component of cellular portion of the blood. [1] Red blood cells do not contain usual cell organelles and are without nucleus, so they can not divide and synthesize structural proteins and enzymes. The energy demands are also very low. The red blood cells derive energy from anaerobic glycolysis and hexose monophosphate pathway. Due to these features, the life span of red blood is relatively short and is only 120 days[2].

Red blood cell synthesis is regulated by many factors, of which erythropoietin[3], vitamin B_{12}, folic acid, iron[1], and vitamin C[4] are especially important. Of these factors, vitamin B_{12} is especially important for maturation of red blood cells. Deficiency of vitamin B_{12} results in the production of immature red blood cells called macrocytes. This immature production of red blood cells results from defective DNA synthesis[1,3].

Malabsorption plays a major role in the development of different nutritional deficiencies. In this regard, stomach plays very important role especially with respect to vitamin B_{12}. Deficiency of vitamin B_{12} mostly results from gastrointestinal diseases and gastric surgery that impair secretion of intrinsic factor from parietal cells of the gastric glands. Major problems of the stomach include gastritis that leads to the development of peptic ulcers, gastric lymphomas, and gastric cancers. It has been reported that the most common cause of gastric problems is *Helicobacter pylori*. The human stomach is regarded as the principal host and reservoir of *Helicobacter pylori*[5,6] *Helicobacter pylori* usually get entry in the human stomach in early childhood. Upon exposure to appropriate host and environmental factor, *Helicobacter pylori* produce low grade inflammatory changes in the gastric mucosa that finally lead to either antral gastritis or pangastritis. These changes result in the development of duodenal and gastric ulcers. It is reported that *Helicobacter pylori* affects red blood cells by causing extragastric complications like vitamin B_{12} deficiency[5-8].

Helicobacter pylori is an organism that is reported to cause deficiency of vitamin B_{12} by producing gastritis and peptic disease which results in malabsorption of vitamin B_{12}.[9] The exact mechanism of *H. pylori* induced vitamin B_{12} deficiency is not known but the suggested mechanisms are that *H. pylori* first produces either achlorhydria or decreases the hydrochloric acid content of the gastric juice by diminishing secretion of

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hydrochloric acid (HCl) from the parietal cells of the gastric glands. [10] Secondly, *H. pylori* neutralizes gastric acidity by producing ammonia from the breakdown of urea that is required for separating vitamin B_{12} from the dietary sources. [11] Thirdly, *H. pylori* also cause decreased secretion of the pepsin (a proteolytic enzyme) from the chief (zymogenic) cells of the gastric glands. The above mentioned three factors are necessary for release of vitamin B_{12} from the food. [10, 11] Fourthly, *H. pylori* decreases the release of intrinsic factor from the parietal cells of the gastric glands by causing their destruction that is a necessary factor for the absorption of vitamin B_{12} from the small intestine[9,12,13]

Different results have been reported about the effects of *Helicobacter pylori* infection on vitamin B_{12}. The present study was planned to elucidate the effects of *H. pylori* infection on serum vitamin B_{12} deficiency and its associated megaloblastic anemia.

MATERIALS AND METHODS

It was a cross sectional analytical study conducted at University of Health Sciences, Lahore. Subjects having *Helicobacter pylori* infection with gastric symptoms and subjects having gastric symptoms without this infection were selected from the Services Hospital, Lahore. Ninety subjects including both male and female were selected for the study. The subjects were between 15-60 years of age. They were divided into three groups. Each group comprised of thirty subjects. In group 1 subjects having history of gastric symptoms and positive *Helicobacter pylori* infection after determining serum *Helicobacter pylori* antibodies quantitatively and by carrying out biopsy based rapid urease test and histopathological examination were included in this group. Grroup 2. This group was composed of subjects with history of gastric symptoms without *Helicobacter pylori* infection. In group 3, age and sex matched healthy subjects without gastric symptoms and *Helicobacter pylori* infection were included.

Subjects having *gastric symptoms for more than one year and Helicobacter pylori* infection were included in the study. Subjects included were not receiving antacids for four weeks before the sample collection. Subjects of both sexes between ages 15 and 60 years were enrolled for the study.

Subjects with history of bleeding ulcer, menorrhagia, fever, blood transfusion, iron or vitamin B_{12} supplement intake, pregnancy and gastric surgery were excluded. Subjects having intrinsic factor antibodies in their serum were also excluded.

Methods: An informed consent and detailed history was taken from all the subjects after explaining the study purpose and procedure. Clinical examination was also carried out. Gastric biopsy samples were collected from the subjects having symptoms of gastric disease through gastric endoscopy for the confirmation of *Helicobacter pylori* infection on the basis of rapid urease test and histopathological examination. Six milliliters of blood was drawn from antecubital vein under aseptic measures. Two milliliter of blood was taken in an EDTA tube for hemoglobin and MCV. Remaining four milliliter of blood was taken in a plain tube and was centrifuged at 5000 rpm for 10 minutes. Serum was separated and stored at -80°C for estimation of serum *H. pylori* IgG antibodies, serum vitamin B_{12}, and intrinsic factor antibodies. Hemoglobin and MCV were estimated by automated hematology analyzer. Serum vitamin B_{12} levels were measured by chemilumisecne technique. Intrinsic factor antibodies were determined by ELISA.

Arithmetic mean and standard deviation of all the quantitative variables like serum vitamin B_{12}, hemoglobin and MCV was determined. One way ANOVA was applied to determine the significance of difference of variables between groups. p value of less than 0.05 was considered statistically significant.

RESULTS

The difference of Mean $\pm$ SD age in three groups was non-significant ($p > 0.05$). In group A, there were 18 males and 12 females while in group B, 16 males and 14 females. In group C, males and females were 14 and 16 respectively. (Table 1) Heaviness in epigartrum, pain epigartrum, retosternal distress and fullness after meal were common gastric symptoms in groups A and B. (Table 2). There was no significant difference ($p>0.05$) in hemoglobin concentration between groups A, B and C (Fig 10). The difference of MCV values between three groups was non significant (Table 3). There was no significant difference in serum vitamin B12 concentration (Fig. 2) between groups A, B and group C (Table 4).

Table 1 Age and sex distribution in groups A, B & C

Parameter	Group A (n = 30)	Group B (n = 30)	Group C (n = 30)
Age (Years)	32.87±12.31	33.27±11.64	33.60±11.12
Male Female	18 12	16 14	14 16

Table 2 Comparison of gastric symptoms in groups A & B

Symptoms	Group A	Group B
Pain epigastrium	24	26
Retrosternal discomfort	22	27
Epigastric heaviness	30	30
Bloating	17	12
Fullness after meals	21	25
Generalized Weakness	15	18
Vomiting	08	11

Table 3 Comparison of red blood cell indices between groups A, B and C

Parameter	Group A (n=30)	Group B (n=30)	Group C (n=30)	ANOVA p value
Hemoglobin (g/dl)	14.86±1.04	14.25±1.36	14.86±.974	.06*
MCV (fl)	86.45±6.47	88.86±6.25	87.14±6.46	.32*

Non- significant

Table 4 Comparison of serum B_{12} between groups A, B and C

Parameter	Group A n =30	Group B n =30	Group C n =30	ANOVA p value
Serum Vitamin B_{12} (pmol/l)	288.1±160.1	262.5±130.8	261.8±135.4	.71*

Non- significant

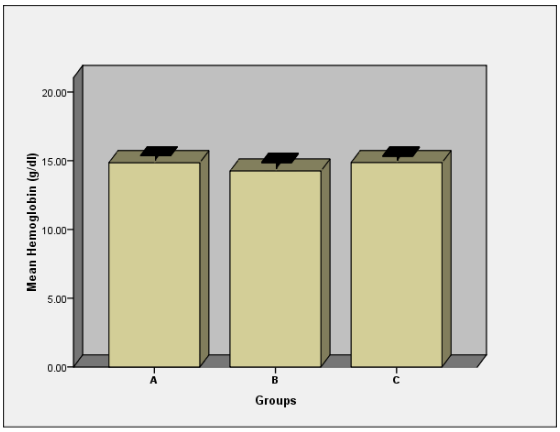

Fig.1 Hemoglobin concentration in groups A, B and C

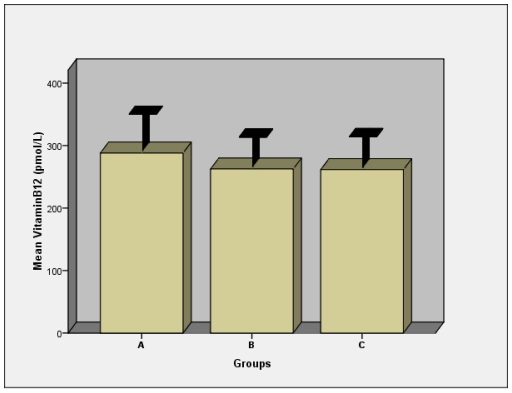

Fig.2 Serum Vitamin B_{12} in groups A, B and C

DISCUSSION

Helicobacter pylori infection is regarded as one of the most common infections all over the World and is labeled as the major cause of gastritis and peptic ulcer disease in the patients. It is also suggested to be associated with extra- gastric complications like iron deficiency, vitamin B_{12} deficiency, and ischemic heart disease. [11, 14] Red blood cells are among the most important cells of the body due to their potential role in oxygen and carbon dioxide transport. Vitamin B_{12} is necessary for the red blood cell maturation[2,15]. The deficiency of vitamin B_{12} results in megaloblastic anemia due to defective DNA synthesis[1,16].

Helicobacter pylori cause deficiency of vitamin B_{12} by producing gastritis and peptic disease which results in malabsorption of vitamin $B_{12.}$ [9] *H. pylori* does this by producing either achlorhydria or decreasing the hydrochloric acid content of the gastric juice[10] and by neutralizing the gastric acidity by producing ammonia from the breakdown of urea that is required for separating vitamin B_{12} from the dietary sources[14]. *H. pylori* also decreases the release of intrinsic factor from the parietal cells of the gastric glands by causing their destruction that is a necessary for the absorption of vitamin B_{12} from the small intestine[9,12,13].

Controversial picture is also present in literature about the vitamin B_{12} status in *Helicobacter pylori* infected cases. Kaptan *et al*., (2000) reported low vitamin B_{12} levels in patients having helicobacter pylori infection[17]. Similarly, Serin *et al*., (2002) found that *Helicobacter pylori* infection had significant effect on vitamin B_{12} level[18]. Akcam *et al*., (2007) reported that effect of *Helicobacter pylori* infection was not significant on both vitamin B_{12} levels and serum ferritin levels[19].

Some studies documented no association between *Helicobacter pylori* infection and vitamin B_{12} deficiency in elderly people[20,22]. In patients having microcytic anemia, no difference was observed in red blood cell parameters of patients with or without *Helicobacter pylori* infection[23]. Many case control reports also documented no relationship between B_{12} deficiency and *Helicobacter pylori* infection[24,25] *Helicobacter pylori* infection has not been suggested as the cause of B_{12} deficiency in alcoholic patients also[26]. No relationship was observed by the Rogers *et al*, (2003) between *Helicobacter pylori*, inflammatory changes, degree of gastritis and deficiency of vitamin B_{12} levels[27]. The present study revealed no significant difference of vitamin B_{12} levels among the three groups. No significant effect of *Helicobacter pylori* infection on serum vitamin B_{12} levels was observed in the patients. Similarly, *Helicobacter pylori* was not found to affect hemoglobin and MCV.

CONCLUSION

It is concluded that *Helicobacter pylori* has no significant effect on vitamin B_{12} level and *H. pylori* infection is not the cause of vitamin B_{12} deficiency. Moreover, there is no significant change in

hemoglobin concentration and MCV levels in patients having *Helicobacter pylori* infection.

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