ORIGINAL RESEARCH

Evaluation of Some Haematological Parameters Among Helicobacter pylori infected students in a Nigerian tertiary educational institution

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ABSTRACT
The study investigated some haematological parameters among Helicobacter pylori infected students of Madonna University, in Elele, River state, Nigeria. A total of 32 patients participated in this study. Of these, 17 were males while 15 were females. Twenty control subjects were established for each gender. Blood was collected from the participants and analyzed for haemoglobin concentration (Hb), packed cell volume (PCV) and erythrocyte sedimentation rate (ESR) following standard procedures. Result showed that test and control subjects had mean values of 12.21g/dl and 13.19g/dl respectively (haemoglobin) 27.76% and 39.70% respectively (PCV) and 24.88 mm/hour and 8.10mm/hour respectively (ESR) for females, and 12.39 g/dl and 13.29g/dl respectively (haemoglobin), 37.53% and 39.90% respectively (PCV) and 25.47 mm/hour and 7.80mm/hour respectively (ESR) for males. Statistically, there was significant difference (P<0.05) among the test and control subjects in the three parameters of study. PCV and haemoglobin were significantly lower while ESR was significantly higher in the test subjects when compared with the controls. The alteration in these parameters could predispose H. pylori patients to other health related effects. Therefore, proper diet must be maintained by the patients to avoid medical complications associated with anemia. Proper and effective management of the infection must be maintained.

KEY WORDS: Bacteria, Hematology, Human health, Ulcer

INTRODUCTION
Helicobacter pylori is a gram-negative, micro aerophilic bacterium (Yahya et al., 2017; Tamokou et al., 2017) that causes ulcers. Different types of ulcer are caused by H. pylori including duodenal ulcer (occurring in the lining of the upper part of the small intestine), stomach ulcer (occurring in the lining of the stomach), peptic ulcer disease and gastric malignancies (Yahya et al., 2017). It could cause gastritis (an inflammation of the stomach lining leading to nausea, vomiting or aching, burning stomach pain). Mucosa-associated-lymphoid-type lymphoma (a cancerous tumor that develops from white blood cells in the stomach lining) (Yahya et al., 2017; Hajimahmoodi et al., 2011) and non-ulcer dyspepsia (a stomach discomfort that is not caused by an ulcer) are other disease conditions that H. pylori can cause. H. pylori infection is usually higher in the elderly compared to children (Jin et al., 2007; Tamokou et al., 2017; Yahya et al., 2017). Furthermore, most individuals infected with H. pylori can leave normal live without knowing that they
harbor the bacterium. According to Kusters et al. (2006), individuals infected with H. pylori develop peptic ulcer (10 – 20% as lifetime risk) and stomach cancer (1 – 2% risk).

The prevalence of H. pylori shows large geographical variations. Socioeconomic and hygiene level also determine the prevalence of H. pylori infection in a given population. Authors have reported that H. pylori is one of the bacteria infection that frequently affects humans worldwide (Lv et al., 2015; Yahya et al., 2017). Globally, over 50% of world population harbours the bacterium in the upper gastrointestinal tract (Ananthakrishnan and Kate, 1998; Amieva et al., 2016). Of these, over 80% of the individuals affected are mostly in the developing nations.

Helicobacter pylori infection is mostly acquired via contaminated food, water and through person to person contact (Karen et al., 2007). Since the bacterium is present in saliva, gastric secretions, faeces, and dental plates, therefore person-to-person is the most likely route of transmission (Clemens et al., 1996; Raymond et al., 2004; Tamokou et al., 2017; Yahya et al., 2017) under suitable environmental conditions such as temperature, oxygen and nutrients.

Overall, patients infected with H. pylori are usually asymptomatic Yahya et al. (2017), MFMER (2017) summarized the signs and symptoms of H. pylori to include abdominal pain, diarrhea, nausea, halitosis, hunger in the morning, heartburn, vomiting, loss of appetite, frequent burping, bloating and unintentional weight loss. Bleeding can also occur and it could lead to weakness and fatigue when the bleeding is prolonged.

H. pylori infection weakens the protective mucous coating of the stomach and upper small intestine, allowing stomach acid to get through to the sensitive lining beneath. When stomach acid and the bacteria irritate the lining, they cause a sore, or stomach ulcer. Helicobacter pylori infection is a major gastric infection worldwide and has been associated with many hematological disorders (Humeida and Abdalla, 2017). Therefore, this study aimed at accessing packed cell volume, hemoglobin estimation and Erythrocyte Sedimentation rate among Helicobacter pylori infected patients in a Nigeria tertiary institution.

RESULTS AND DISCUSSION

The effects of Helicobacter pylori infection on some hematological parameters were presented in Table 1 and 2 for males and females respectively. In the test and control subjects, the mean values were 12.21g/dl and 13.19g/dl respectively (hemoglobin), 27.76% and 39.70% respectively (packed cell volume) and 24.88 mm/hour and 8.10mm/hour respectively (erythrocyte sedimentation rate) for females (Table 1). In females, results showed that H. pylori infection leads to decline in hemoglobin and packed cell volume and an increase in erythrocyte sedimentation rate. Statistically, there was significant difference (P<0.001) among the test and control subjects in the three parameters analyzed.

In males, the test and control subjects had mean values of 12.39 g/dl and 13.29g/dl respectively (hemoglobin), 37.53% and 39.90% respectively (packed cell volume) and 25.47 mm/hour and 7.80mm/hour respectively (erythrocyte and sedimentation rate) (Table 2). There was significant difference (P<0.05) among the test and control subjects in the three parameters of study. The values in males showed similar trend it showed in females, with a decline in haemoglobin and packed cell volume and an increase in erythrocyte sedimentation rate in H. pylori infected patients.

Based on sex distribution of H. pylori on hemoglobin, packed cell volume and erythrocyte sedimentation rate, the effect is highly similar (Figure 1-3). This is an indication that H. pylori do not affect a particular sex more among patients within a given geographical area.

**Figure 1:** Sex based distribution of haemoglobin concentration on H. pylori infected patients
Table 1: Effect of *H. pylori* on hemoglobin, packed cell volume and erythrocyte sedimentation rate in females

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean± standard error</th>
<th>t-value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subjects (n=17)</td>
<td>Control (n=20)</td>
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<td></td>
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<tr>
<td>Hemoglobin, g/dl</td>
<td>12.21±0.19</td>
<td>13.19±0.14</td>
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<tr>
<td>Packed cell volume, %</td>
<td>35.76±0.82</td>
<td>39.70±0.45</td>
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<tr>
<td>Erythrocyte sedimentation rate, mm/hour</td>
<td>24.88±0.70</td>
<td>8.10±0.58</td>
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</tbody>
</table>

Table 2: Effect of *H. pylori* on haemoglobin, packed cell volume and erythrocyte sedimentation rate in males

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean± standard error</th>
<th>t-value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subjects (n=15)</td>
<td>Control (n=20)</td>
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<tr>
<td>Hemoglobin, g/dl</td>
<td>12.39±0.22</td>
<td>13.29±0.18</td>
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<tr>
<td>Packed cell volume, %</td>
<td>37.53±0.99</td>
<td>39.90±0.54</td>
<td></td>
</tr>
<tr>
<td>Erythrocyte sedimentation rate, mm/hour</td>
<td>25.47±0.63</td>
<td>7.80±0.57</td>
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</table>

Packed cell volume is the portion of whole blood volume covered by erythrocytes. The packed cell volume recorded in test subjects were significantly lower compared to the control. This is an indication of an aberration in the body. Typically, some health conditions are known to decrease packed cell volume. For instance, Eledo *et al.* (2015b) reported that menopause among women lead to decline in packed cell volume level. Pregnancy conditions also lead to decline in packed cell volume (Eledo *et al.*, 2017).

Erythrocyte sedimentation rate is one of the parameters used in assessing possible inflammation in the body. Furthermore, a normal result does not rule out inflammation or disease. Slight elevation in erythrocyte sedimentation rate generally occurs due to health condition such as ageing and menopause in women (Eledo *et al.*, 2017), anemia, infection, pregnancy, etc. However, in this study, elevation in erythrocyte sedimentation rate could be due to *H. pylori* infection which predisposes the patients to anemia.

Hemoglobin aids in the transportation of oxygen to the various part of the body. Oxygen is required by body cells to function effectively. When hemoglobin levels are low, cells, tissues, and organs are starved of required oxygen. Among the *H. pylori* patients, the hemoglobin content decreased significantly. Generally, a slight decline in hemoglobin is compensated for by the body and has no major adverse effects.
effects. Despite the significant difference that exits in both sexes, the test subject hemoglobin level is near the permissible level. This suggests that *H. pylori* infection leads to slight decline in hemoglobin level. But the variation is not much to cause a major adverse effect on the patients. However, caution should be exercised by the subjects when they have symptoms of low hemoglobin concentration. Typically, health conditions are known to affect hemoglobin concentration. For instance, Eledo et al. (2017) reported that menopause among women leads to decline in hemoglobin level. Furthermore, pregnancy could also reduce hemoglobin level (Eledo et al., 2015a).

**CONCLUSION**

This study assessed erythrocyte sedimentation rate, packed cell volume and haemoglobin concentration among *H. pylori* infected patients in Elele, River state, Nigeria. The findings of this study showed that *H. pylori* infection leads to a slight decline in packed cell volume and hemoglobin concentration and an increase in erythrocyte sedimentation rate. In all cases, there was significant variation (P<0.05) between the test subjects and control. The variations in the parameters compared to the control suggest possible inflammation due to *H. pylori* bacterium. Therefore, caution should be exercised by *H. pylori* infected patients in managing their health status so as to avoid adverse health conditions. Therefore, proper diet must be maintained by the patients to avoid medical complications associated with anemia. Proper and effective management of the infection must be maintained.

**MATERIALS AND METHODS**

**Study Area**

The study was carried out among students of Madonna University, Elele in Rivers State Nigeria. Elele town is located in the tropics of southern part of Nigeria. Elele is located within Latitude N 05°05’06”-06’ Longitude E 006°45’-49’. The climatic condition of the area is similar to other Niger Delta regions as have been previously described by authors (Ben-Eledo et al., 2017a;b; Seiyaboh et al., 2017a-c; Seiyaboh and Izah, 2017a;b; Ogamba et al., 2015a,b,c; 2017a,b). There are other towns and villages that surround Elele town, they include; Isiokpo town, Ndoni, Omagwa, Ahaoda, Omoku, among others. Farming and other businesses are the major occupation of the Elele people. Palm oil and cassava cultivation and processing are some of the useful economic crops that serve as means of livelihood to the residents of the area.

**Selection Criteria for Subjects**

Inclusion Criteria: Subjects for this research were students of Madonna University, Elele. Only those who tested positive for *H. pylori* using one step anti-HP rapid screen test kit (Lot Number: 20161115) were selected for the study. A total of 17 female *Helicobacter pylori* infected patients and 15 male *Helicobacter pylori* infected patients within the age of 18 – 32 years participated in this study. Another 20 age-matched control subjects who were negative for *H. pylori* were selected for each.

Exclusion criteria: Pregnant women, lactating mothers, and individuals with known cases of HIV/AIDS, hepatitis, tuberculosis, diabetics and cardiovascular diseases.

**Blood Collection**

A standard venepuncture technique as described by Bain et al. (2008), was employed. A sterile dry plastic syringe of 5ml capacity together with a 21g size needle was used for the collection of blood.

A soft tubing tourniquet was applied to the upper arm of the patient to enable the veins to be seen and felt. The patient was asked to make a tight fist which made the veins more prominent. A suitable vein (cubital vein) was then selected for venepuncture. The puncture site was sterilized with 70% ethanol and allowed to dry.

Approximately 4mls of blood was collected from each subject and dispensed immediately into a tube containing Ethylene diamine tetra acetic acid (1.5mg/ml of blood) and mixed properly. All samples were analyzed within six hours from time of collection.

**Laboratory Analysis**

**Packed cell volume**

Packed cell volume was carried out using capillary tube with 75mm length and 1mm diameter. Well mixed anticoagulated blood was allowed to enter the capillary tubes leaving about 15mm unfilled. The capillary tubes were sealed with plasticine and centrifuged in a microhaematocrit centrifuge at a speed of 12000g for 5 minutes. At the end of the centrifugation, the tubes were brought out and readings were made using a micro-haematocrit reader and the packed cell volume was expressed as percentage.

**Hemoglobin Estimation**

The hemoglobin was estimated using Drabkin’s solution methods. Approximately 0.02ml of whole blood was added to 4ml of Drabkin’s solution to make a solution of 1:200 dilutions. The tubes were then well stoppered and several inversions made. The solution was allowed to stand for 5 minutes at room temperature. The absorbance of the solution was then read on a spectrophotometer at
540nm using Drabkin’s solution as blank. The absorbance of the cyanmethaemoglobin standard was read also against the blank. The hemoglobin concentration was calculated as follows: 

Hemoglobin estimation = Absorbance of test X Concentration of standard X Dilution factor / Absorbance of standard X 1000

Erythrocyte Sedimentation Rate
The Erythrocytes sedimentation rate was estimated using westergren method. EDTA anticoagulant venous blood was diluted in a teat with the diluent (trisodium citrate) in the proportion of 1 volume of trisodium citrate to 4 volumes of blood. The blood sample was then well mixed and drawn up into clear and dry westergren tube up to the 0mm mark from the 150mm mark by means of a teat. The tube was then placed vertically in a rack and left to stand undisturbed for one hour away from direct sunlight and vibrations. At the end of the 1 hour period, readings were then made on the rate of fall of red cells by taking the distance of the clear plasma above the upper limit of the column of sedimenting cells and expressed as mm/hour.

Statistical analysis
Statistical analysis was carried out using SPSS software version 20. Data were presented as mean ± standard error. The mean of Erythrocyte sedimentation rate, packed cell volume and hemoglobin estimation for the H. pylori infected patients and control subjects were subjected to “t” test. Significance level was determined at P-value < 0.05.

ETHICAL CONSIDERATION
Permission was obtained from the ethics committees of the Medical Laboratory Science Department of Madonna University, Elele, Nigeria. Informed consent was obtained from the patients prior to sample collections.

REFERENCES


