Biochemical and Hematological Profile of Anemic and Non-Anemic Pregnant Women

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ABSTRACT

Objective: To determine the biochemical and hematological profile of pregnant women with and without anemia attending Sanderman Provincial Hospital, Quetta.

Methods: This descriptive cross-sectional study was conducted among pregnant women aged ranged from 18 to 48 years attending the antenatal clinic for the first-time during February 2019 to August 2019. Hematological parameters like white blood cells (WBC), hemoglobin (Hb), hematocrit (HCT), Red blood cell distribution width (RDW), mean corpuscular hemoglobin concentration (MCHC), and mean corpuscular volume (MCV) were collected. Furthermore, biochemical parameters like serum ferritin and iron level, total iron-binding capacity (TIBC), transferrin saturation (TfS), total bilirubin, direct bilirubin, and indirect bilirubin were also noted.

Results: Of 357 patients, the mean Hb level was 10.27 ±1.53 g/dl. Majority of the pregnant were anemic, i.e. 248 (69.5%). Of these 248 anemic patients, mild anemia was found in 135 (54.4%), moderate anemia in 100 (40.3%) and severe anemia in 13 (5.2%) patients. A significantly lower level of Hb (p-value <0.001), WBC (p-value), HCT (p-value <0.001), MCHC (p-value <0.001), MCV (p-value <0.001), and RDW (p-value <0.001) was observed in patients with anemia as compared to non-anemic patients. Moreover, serum ferritin level (p-value 0.002), serum iron (p-value <0.001), and total bilirubin level (p-value <0.001) were found to be significantly lower in anemic patients than non-anemic patients.

Conclusion: The anemia in pregnant women is associated with considerably low level of hematological and biochemical levels. Health providers should give attention to pregnant women with lower hematological and biochemical levels to prevent maternal and fetal complications.

Keywords: Biochemical, Hematology, Anemia, Pregnant Women

INTRODUCTION

Anemia is the most common nutritional deficiency in pregnant women. In developing nations such as Pakistan the most common cause is iron deficiency. It is estimated that 38% of pregnant women worldwide are affected by anemia. In particular, the reported prevalence of anemia among pregnant women in low- and middle-income countries is 56%. Iron deficiency anemia and the term anemia are often used interchangeably in pregnancy.

In normal physiology, pregnancy manifests several changes. The ones that require observation and intervention are those that change hematological serum levels. Most of these modifications lead to plasma expansion and hemodilution during pregnancy. Several hematological and other markers used to discern iron levels were used in the diagnosis of anemia during pregnancy. These include serum ferritin level, serum iron level, serum transferrin, transferrin receptor (TfR), total iron-binding capacity (TIBC), transferrin saturation, MCV, MCHC, and RDW.

Moreover, several biochemical changes are also reported in the literature with respect to anemia in pregnant women. Anemia during pregnancy has several well-known complications regarding fetal and maternal health including increased risk of preterm birth, perinatal mortality, and intrauterine growth retardation. Maternal stores of iron are depleted during pregnancy to supply the fetus for the formation of fetal hemoglobin.

We were unable to reach consensus cutoff values due to the varying results found across the literature as well as a lack of a specific population. Across the length of pregnancy, there are multiple assays repeated at
intervals to monitor all the parameters according to assure both maternal and fetal wellbeing. Despite anemia being a preventable condition in pregnancy, there is very limited local research work available on the hematological and biochemical difference in pregnant women with anemia.

**METHODS**

A descriptive cross-sectional study was conducted at Sanderman Provincial Hospital, Quetta from February 2019 to August 2019. The study was conducted after getting approval from the institutional committee. Moreover, informed consent was also obtained from all study participants. All pregnant women aged ranging from 18 to 48 years attending the antenatal clinic for the first time were enrolled. Expectant women in need of emergent care or possessing risk factors such as gestational diabetes, pre-eclampsia, or eclampsia, or confirmed case of HIV were excluded.

Open Epi sample size calculator was used for the estimation of sample size taking confidence interval 95%, margin of error 5%, reported prevalence of anemia 63.2%. The estimated sample size came out to be 357. Hemoglobin (Hb) cut off value adjusted to sea level altitude was used to define anemia on the basis of gestational age and to classify severity using criteria as devised by WHO. The presence of Hb value <11.0 g/dl at 1st and 3rd trimesters and <10.5 g/dl at 2nd trimester was classified as anemia. As far as the severity is concerned, 10-11g/dl Hb level at 1st and 3rd trimesters and 10-10.5 g/dl Hb level at 2nd trimester were classified as mild anemia. Pregnant women having a Hb value of 7-10g/dl was classified as moderate anemia, and Hb level of <7g/dl were classified as severe anemia.

A venous blood sample of 5 milliliters was drawn for hematological and biochemical analysis in between 8 am and 9 am. Hematological parameters like white blood cells (WBC), Hb, hematocrit (HCT), Red blood cell distribution width (RDW), mean corpuscular hemoglobin concentration (MCHC), and mean corpuscular volume (MCV) were collected. Furthermore, biochemical parameters like serum ferritin level, serum iron level, total iron-binding capacity (TIBC), Transferrin saturation (TfS), total bilirubin, direct bilirubin, and indirect bilirubin was also noted. Statistical analysis was performed using SPSS version 24. Quantitative variables like age, hemoglobin level, WBC, Hb, HCT, MCHC, MCV, RDW, serum ferritin level, serum iron level, TIBC, TfS, total bilirubin, direct bilirubin, and indirect bilirubin were calculated using mean and standard deviation. Frequency and percentages were calculated for variables like types of anemia and severity of anemia. The mean difference of hematological and biochemical parameters was compared with respect to the status of anemia. An independent t-test was applied. A comparison was also done to see the association of anemia status with serum ferritin level and severity of anemia. Chi-square test was applied. p-value <0.05 taken as significant.

**RESULTS**

Of 357 patients, the mean age of the patients was 27.29 ±2.68 years. The mean Hb level was 10.27 ±1.53 g/dl. There were 248 (69.5%) anemic and 109 (30.5%) non-anemic patients. Of these 248 anemic patients, mild anemia was found in 135 (54.4%), moderate anemia in 100 (40.3%) and severe anemia was observed in 13 (5.2%) patients.

The hematological analysis showed a mean serum Hb level was found to be 9.89 ±1.42 g/dl, WBC 7.84 ±1.85 x 10^3/µL, HCT 31.46 ±2.57 %, MCHC 32.03 ±1.29g/dl, MCV 80.32 ±3.71 fl, and RDW 15.83 ±1.37 %. A significantly lower level of Hb (p-value <0.001), WBC (p-value), HCT (p-value <0.001), MCHC (p-value <0.001), MCV (p-value <0.001), and RDW (p-value <0.001) was observed in patients with anemia as compared to non-anemic patients. (Table 1)

The biochemical analysis revealed that mean Serum Ferritin level was 35.99 ±25.53 ng/l, Serum Iron level 116.51 ±37.65 µg/dl, TIBC 307.54 ±74.47 µg/dl, TfS 20.88 ±6.70 %, Total Bilirubin 18.05 ±1.76 µmol/l, Direct Bilirubin 5.57 ±0.71 µmol/l, and Indirect Bilirubin 12.32 ±0.92 µmol/l. Except TfS (p-value 0.419), direct bilirubin level (p-value 0.645), and indirect bilirubin (p-value 0.778), all other biochemical parameters like serum ferritin level (p-value 0.002), serum iron (p-value <0.001), and total bilirubin level (p-value <0.001) were found to be significantly lower in anemic patients as compared to non-anemic patients. (Table 2)

Serum ferritin level of ≤30ng/l was significantly higher (n=159, 64.1%) among anemic patients as compared to non-anemia (n=44, 40.4%) (p-value <0.001). (Figure 1) Similarly, serum ferritin level of ≤30ng/l was significantly higher (n=13, 100%) in patients with severe anemia as compared to mild (n=86, 63.7%) and moderate anemia (n=60, 60%) (p-value 0.018). (Figure 2)
The majority of pregnant women, the overall anemia prevalence was predominantly higher in pregnant women, i.e. 69.5%. Comparing to current literature, a study conducted by Baig-Ansari et al in an urban city of Pakistan, the anemia prevalence in pregnant women is reported as 90%. Similar to our study, in their study the

### DISCUSSION

This study has reported a significantly lower threshold of both hematological and biochemical parameters in pregnant women with anemia as compared to non-anemia. A more detailed analysis of our results showed that though mild and moderate anemia was noted in the majority of the pregnant women, the overall anemia prevalence was predominantly higher in pregnant women, i.e. 69.5%. Comparing to current literature, a study conducted by Baig-Ansari et al in an urban city of Pakistan, the anemia prevalence in pregnant women is reported as 90%. Similar to our study, in their study the

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**Table 1: Mean difference of hematological characteristics with respect to anemic status of the patients (n=357)**

<table>
<thead>
<tr>
<th>Variables</th>
<th>Overall</th>
<th>Anemic (n=248)</th>
<th>Non-Anemic (n=108)</th>
<th>p-value</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemoglobin, g/dl</td>
<td>9.89 ±1.42</td>
<td>9.04 ±0.63</td>
<td>11.84 ±0.59</td>
<td>&lt;0.001</td>
<td>-2.94 to -2.65</td>
</tr>
<tr>
<td>WBC, x 10^3/µL</td>
<td>7.84 ±1.85</td>
<td>6.65 ±0.41</td>
<td>10.57 ±0.33</td>
<td>&lt;0.001</td>
<td>-4.01 to -3.84</td>
</tr>
<tr>
<td>HCT, %</td>
<td>31.46 ±2.57</td>
<td>29.93 ±0.81</td>
<td>34.98 ±1.59</td>
<td>&lt;0.001</td>
<td>-5.30 to -4.79</td>
</tr>
<tr>
<td>MCHC, g/dl</td>
<td>32.03 ±1.29</td>
<td>31.25 ±0.56</td>
<td>33.81 ±0.51</td>
<td>&lt;0.001</td>
<td>-2.67 to -2.43</td>
</tr>
<tr>
<td>MCV, fl</td>
<td>80.32 ±3.71</td>
<td>78.13 ±0.83</td>
<td>85.53 ±2.71</td>
<td>&lt;0.001</td>
<td>-7.59 to -6.85</td>
</tr>
<tr>
<td>RDW, %</td>
<td>15.83 ±1.37</td>
<td>16.35 ±1.26</td>
<td>14.62 ±0.63</td>
<td>&lt;0.001</td>
<td>1.48 to 1.98</td>
</tr>
</tbody>
</table>

WBC: White blood cells, HCT: Hematocrit, MCHC: Mean corpuscular hemoglobin concentration, MCV: Mean corpuscular volume, RDW: Red cell distribution width.

All data presented as mean ±SD.

Independent t-test applied, p-value <0.05 taken as significant.

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**Table 2: Mean difference of biochemical characteristics with respect to anemic status of the patients (n=357)**

<table>
<thead>
<tr>
<th>Variables</th>
<th>Overall</th>
<th>Anemic (n=248)</th>
<th>Non-Anemic (n=108)</th>
<th>p-value</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum Ferritin, ng/l</td>
<td>35.99 ±25.53</td>
<td>33.24 ±24.82</td>
<td>42.43 ±26.20</td>
<td>0.002</td>
<td>-14.90 to -3.45</td>
</tr>
<tr>
<td>Serum Iron, µg/dl</td>
<td>116.51 ±37.65</td>
<td>99.01 ±29.27</td>
<td>156.06 ±20.24</td>
<td>&lt;0.001</td>
<td>-63.14 to -50.95</td>
</tr>
<tr>
<td>TIBC, µg/dl</td>
<td>307.54 ±74.47</td>
<td>309.19 ±71.58</td>
<td>304.33 ±80.07</td>
<td>0.573</td>
<td>-12.04 to 21.77</td>
</tr>
<tr>
<td>TFS, %</td>
<td>20.88 ±6.70</td>
<td>20.69 ±6.84</td>
<td>21.32 ±6.44</td>
<td>0.419</td>
<td>-2.14 to 0.89</td>
</tr>
<tr>
<td>Total Bilirubin, µmol/l</td>
<td>18.05 ±1.76</td>
<td>18.25 ±1.81</td>
<td>17.59 ±1.55</td>
<td>&lt;0.001</td>
<td>0.25 to 1.04</td>
</tr>
<tr>
<td>Direct Bilirubin, µmol/l</td>
<td>5.57 ±0.71</td>
<td>5.56 ±1.81</td>
<td>5.60 ±0.69</td>
<td>0.645</td>
<td>-0.20 to 1.24</td>
</tr>
<tr>
<td>Indirect Bilirubin, µmol/l</td>
<td>12.32 ±0.92</td>
<td>12.30 ±0.89</td>
<td>12.33 ±0.95</td>
<td>0.778</td>
<td>-0.23 to 0.18</td>
</tr>
</tbody>
</table>

TIBC: Total Iron Binding Capacity, TFS: Transferrin saturation.

All data presented as mean ±SD.

Independent t-test applied, p-value <0.05 taken as significant.

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**Figure 1: Frequency of anemia status by categories of serum ferritin (n=248)**

**Figure 2: Comparison of types of anemia with serum ferritin level (n=248)**

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**DISCUSSION**

This study has reported a significantly lower threshold of both hematological and biochemical parameters in pregnant women with anemia as compared to non-anemia. A more detailed analysis of our results showed that though mild and moderate anemia was noted in
prevalence of mild anemia was found to be highest followed by moderate anemia, and then severe anemia. Similarly, a recent study from Lahore, Pakistan has also reported more than half, i.e. 57.7% pregnant women with anemia.\(^6\) Similar to the study of Baig-Ansari et al,\(^7\) a relatively higher proportion of anemia, i.e. 64% and 68% was reported in a recent studies conducted in India.\(^8\)\(^9\) These findings are surprisingly higher compared to the findings reported in a low-income country like Ethiopia in which the anemia in pregnant women ranges from 16.88% to 52%.\(^10\)\(^11\) In a recent study conducted in Ghana, amongst 400 pregnant women, 50.8% of pregnant women had anemia.\(^12\) The variability in the prevalence of the disease can be attributed to the variability of socioeconomic standards across Pakistan. Lower-income households often have fewer means to correct nutritional deficiencies even if they are aware of them. The same subset is also more likely to have consecutive pregnancies, leading to a further detriment in the problem.

Our study also found a significantly lower Hb, and WBC levels in pregnant women with anemia than that of non-anemic pregnant women, along with lower HCT, MCV, MCHC, and RDW values. These findings are consistent with the current literature. Various studies including studies done outside of Pakistan such as Ahenkorah et al, James et al and Anchang-Kimbi et al report similarly decreased levels of hemoglobin and white blood cells.\(^6\)\(^20\)\(^21\) Since HCT expresses the concentration of Hb in blood, the reduced Hb in the blood give a lower HCT Value.\(^22\) Additionally, a physiologic change in pregnancy is the increase in plasma volume and hormonal changes, which may give a lower HCT due to hemodilution and fluid retention.\(^22\) A low mean corpuscular volume (MCV) is a marker of microcytic anemia, most commonly iron deficiency anemia, which is expected and often prophylactically treated in pregnancy.

In our study, a significantly lower prevalence of serum ferritin was also reported in anemic patients. In addition, a significantly higher proportion of low serum ferritin was also reported in patients with severe anemia. Among anemic pregnant women, the presence of noticeably low levels of serum iron, ferritin, and transferrin saturation are supportive of findings in recent studies including the finding by Ahenkorah B et al\(^6\) and Raza et al\(^14\). Decreased values for serum iron, ferritin, and transferrin on blood assays are suggestive of iron deficiency anemia. As our study found there was a higher incidence of pregnant anemic women with low iron, among anemic pregnant women. This supports our previous stipulation of the presence of iron deficiency anemia.\(^25\) This belief is compounded by the fact that our study found a higher mean TIBC level in pregnant women as compared to those who were not. A higher TIBC is also an indication of iron deficiency. The findings of the current study could be highlighted in the light of various limitations including the fact that biochemical and hematological profiles for each trimester of the pregnancy are not reported in the current study. It is expected that serum levels of iron and hemoglobin may shift during the curse of pregnancy and so, the stage of pregnancy is a variable that would have provided additional clarity in our work. Moreover, other effect modifiers like residence, educational status, occupational status, and socioeconomic status are also not reported in this study. As we have noted, such qualifiers affect the maternal health of women in many ways. In addition, the dietary habits of women in our study with and without anemia is also an important factor which needs to be explored. An important strength of our work is the combination of measurement of biochemical and hematological parameters, we have been able to accomplish despite an operation in a low resource setting. This is unlike many other studies, where the two types of tests have been reported using separate samples, which may be a cause for variability. These lab test samples were drawn from the same patient, synchronously, while in other studies, different subjects were used. This helps keep the data consistently uniform and removes a source of uncertainty.

**CONCLUSION**

Maternal health and the morbidity and mortality associated with it is an important source of concern in Pakistan. The anemia in pregnant women is associated with a considerably low level of hematological and biochemical levels. It is recommended that health providers should give attention to pregnant women with lower hematological and biochemical levels to prevent maternal and fetal complications. More resources need to be targeted toward educating women in how pregnancy affects their nutritional needs, as well as consideration for prophylactic supplement use during pregnancy. More studies need to be done on the ways these changes affect women across various economic classes in Pakistan and how availability and adherence to education regarding these subjects can improve the quality of healthcare they receive.

**ETHICAL APPROVAL:** The study was approved by the Sanderman Provincial Hospital, Quetta.
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AUTHORS’ CONTRIBUTION: ZA, JL conceived and designed study, AK, RU, ML, JL, ZA did data collection, ZA did analysis, ZA, TG, RN did manuscript writing, ZA did manuscript editing. All authors gave final approval of manuscript.

CONFLICT OF INTEREST: The authors have no conflict of interest to declare.

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