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Original Research Article

# Reference intervals and gender variation in establishing blood parameters in dental institution: A retrospective study

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ABSTRACT

**Introduction:** Complete blood count (CBC) has been extensively used by general physicians to assess the status of sick and healthy people. The aim of the present study was to appraise the hematological changes in different blood parameters amongst males and females and also to set a reference range for general population.

**Materials and Methods:** The study was conducted on 361 patients of age 18-50 years comprising of 163 males (45.2%) and 198 females (54.8%). 20 $\mu$l sample of capillary blood was obtained through finger prick and subjected to complete blood count, including a white blood cell (WBC) differential and a reticulocyte count, by means of a fully automatic blood cell counter. The findings were tabulated and subjected to statistical analysis.

**Results:** The mean values of the RBC, Hb, HCT, MCV, MCH between males and females showed statistically highly significant reduction with $p < 0.001$, while MCHC was significantly reduced with $p < 0.05$ in females compared to males (Figure 1). Haematological parameters such as PLT, PCT were slightly higher in females compared to males showing statistically significant results with $p < 0.05$.

**Conclusion:** Blood parameters aid in diagnosis of various disorders and also serve to guide decision making for clinicians. Therefore, periodically establishing reference intervals is crucial for better correlation with health and disease conditions. Our findings would serve to update present available data pertaining to blood parameters, their reference range and gender variation on a regional level.



## 1. Introduction

Complete blood count (CBC) has been extensively used by general physicians to assess the status of sick and healthy people. CBC is routinely employed owing to its simplicity, low cost and easy availability, which enables an appropriate approach to investigate, diagnose and determine the prognosis of the conditions such as anemia, risk of infection and/or hematologic malignancies, coagulation disorders and screening of blood donors.[1]

Observed values from laboratory tests of a person or patient are compared with a reference interval for determination of accurate diagnosis, therapeutic management or other physiological assessment.[2] In general, nearly all the investigations in a clinical laboratory have a pre-determined reference interval.[3] The decision-making process of a clinician is guided by the difference between observed values and reference intervals.

Several studies has investigated hematologic parameters in different populations, racial, ethnic and gender subgroups, even in different seasons.[4,5] In most of these studies gender difference in hematological parameters

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were significant and therefore it is essential to establish this difference and also to estimate reference range for population. The present study was conducted to appraise the hematological changes in different blood parameters amongst males and females and also to set a reference range for general population.

## 2. Materials and Methods

### 2.1. Sample size and sample collection

The study was conducted on a total of 361 patients comprising of 163 males (45.2%) and 198 females (54.8%) in the institutional hematology laboratory. Patients of age 18 to 50 years reporting for dental treatment and did not have any systemic illness were included in the study by means of convenience sampling method. The study was conducted over a period of six months from May to December, 2019. 20 $\mu$l sample of capillary blood obtained through skin puncture over a finger which was added to 100 $\mu$l diluent containing sodium chloride, sodium sulfate, a phosphate buffer agent, ethylene diamine tetra-acetic acid (EDTA), 1-pyridone-2-sulphur and formaldehyde with pH value of the solution 6.5-7.4.

### 2.2. Haematology analyser

CBC, including a white blood cell (WBC) differential and a reticulocyte count, were performed using fully automatic blood cell counter PCE 210 [ERMA INC, Japan] by small volume of sample suction. Measurements of parameters and histograms were quickly processed within 60 sec. The main principle involved for counting cells is by electrical impedance, also known as the Coulter principle.[6] Whole blood is passed between two electrodes through an aperture where the area is so small that only one cell can pass at a time. Using hydrodynamic focusing, the cells are sent through an aperture one cell at a time. During this, a laser is directed at them, and the scattered light is measured at multiple angles. The absorbance is also recorded. The cell can be identified based on the intensity of the scattered light and the level of absorbance and haemoglobin measurement is done by colorimetric method.[6,7]

### 2.3. Observational parameters

The components of the CBC consisting of hemogram and differential WBC count. The hemogram includes enumeration of WBCs, red blood cells (RBCs), and platelets. It also allows evaluation of hemoglobin, hematocrit, and RBC indices. The differential WBC count enumerates the different WBC types. Together, hemogram and differential WBC count aid in determining diagnosis, treatment and prognosis of haematological disorders.[8]

Following parameters were assessed: (Table 1)[9,10]

### 2.4. Statistical analysis

Descriptive and inferential statistical analyses were carried out in the present study. Results on continuous measurements were presented on Mean ± SD and results on categorical measurement were presented in number (%). Level of significance was fixed at p=0.05 and any value less than or equal to 0.05 was considered to be statistically significant.

All the variables from the study were statistically analyzed using the statistical software IBM SPSS statistics 20.0 (IBM Corporation, Armonk, NY, USA).

## 3. Results

The mean values of haematological parameters obtain are enlisted in Table 2. The mean values of haematological parameters between males and females were compared, and it revealed statistically significant results with certain parameters using unpaired t test (Table 3). The reference range of parameters were established using median and SD (Table 4).

The mean values of the RBC, Hb, HCT, MCV, MCH between males and females showed statistically highly significant reduction with $p < 0.001$, while MCHC was significantly reduced with $p < 0.05$ in females compared to males (Figure 1). Haematological parameters such as PLT, PCT were slightly higher in females compared to males showing statistically significant results with $p < 0.05$ (Figure 2).

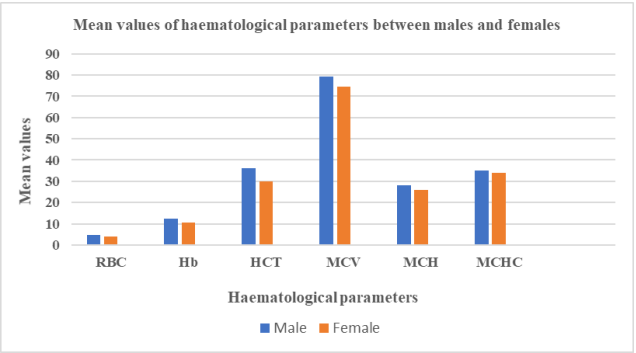


**Fig. 1:** Mean values of haematological parameters in males & females

## 4. Discussion

Having thorough knowledge of the reference values of RBCs and WBCs variables amongst males and females is profoundly important for proper interpretation of the results of CBC.[11] The gender differences seen in haematological profile is a well-documented fact that has been similarly reported in other studies, males having a higher RBC profile compared to females.[12,13] Females had a higher platelet

**Table 1:**

| S. No. | Parameters | Description | Normal value | |
|---|---|---|---|---|
| | | | Male | Female |
| 2. | White Blood Cell (WBC) | Immune system | 4,000-11,000 cells/ul | |
| 3. | Lymphocytes | Cell mediated and humoral immunity | 1,500-4,000 cells/ul | |
| 4. | Granulocyte | | | |
| | Neutrophils | Phagocytosis | 2,000-7, 500 cells/ul | |
| | Eosinophils | Ag-Ab reaction | 40-400 cells/ul | |
| | Basophils | Histamine release | 10-100 cells/ul | |
| 5. | Red Blood Cell (RBC) | Oxygen transport | 4.5-6.5 million/$mm^3$ | 4-5.5 million/$mm^3$ |
| 6. | Haemoglobin (Hb) | Oxygen transport | 13.5-18 g/dl | 11.5-16 g/dl |
| 7. | Haematocrit (HCT) | Ratio of the volume of RBC to the total volume of the blood | 40-52% | 37-47% |
| 8. | Mean Corpuscular Volume (MCV) | Average volume of red blood corpuscles | 77-93 femtoliter | |
| 9. | Mean Corpuscular Haemoglobin (MCH) | Average mass of Hb per RBC | 27-32 picogram | |
| 10. | Mean Corpuscular Haemoglobin Concentration (MCHC) | Average concentration of Hb in given volume of blood | 30-35 g/dl | |
| 11. | Red cell Distribution Width (RDW) | Amount of RBC variation in volume and size | 11.8-14.5% | 12.2-16.1% |
| 12. | Platelet (PLT) | Primary haemostasis | 1,50,000-4,00,000 cells/ul | |
| 13. | Plateletcrit (PCT) | Volume occupied by PLT in a blood as a percentage | 25-27% | |
| 14. | Mean platelet volume (MPV) | Average size of PLT | 8-12 femtoliter | |

**Table 2:** Mean values of haematological parameters

| Variable | WBC | LYM | GRA | RBC | Hb | HCT | MCV | MCH | MCHC | RDW | PLT | PCT | MPV |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Mean** | 7.6632 | 2.7136 | 4.0468 | 4.3355 | 11.318 | 32.827 | 76.745 | 26.974 | 34.499 | 15.922 | 251.47 | 0.2472 | 9.442 |
| **SD** | 2.6109 | 0.9822 | 1.7467 | 1.9620 | 2.920 | 7.707 | 11.386 | 4.335 | 2.986 | 2.129 | 93.553 | 0.1132 | 0.7362 |

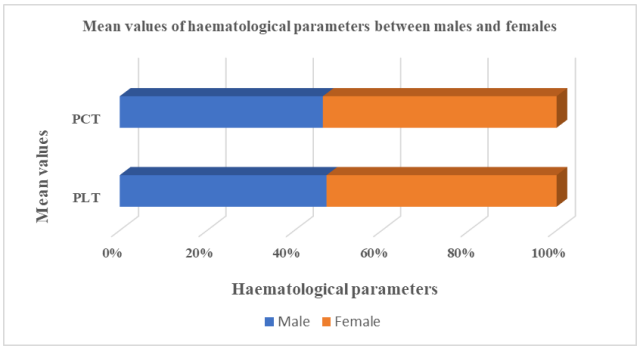


**Fig. 2:** Mean values of PLT, PCT in males & females

count compared to males, comparable to a study which looked at cultural and sex differences in WBCs and PLTs counts.[14]

The sex differences in Hb level in adults are well known, and the underlying mechanisms is probably a direct effect of sex hormones, both estrogen and androgens on erythropoiesis.[15] There is no evidence showing reduced cellular mechanisms for heme synthesis in females, and there is no difference in the iron absorption between females and males.[16] In pre-pubertal humans, no major differences can be found between the genders in RBC count or Hb and serum ferritin concentrations. The difference in haematological variables between genders emerges after onset of menstruation and persist until 10 year after the menopause.[17] The two main reasons for lower values of Hb, iron and RBC count of females compared to males are menstruation and nutritional intake.[18] The reduced level of RBC and Hb in females are responsible for subsequent reduction in HCT, MCV, MCH and MCHC as these parameters are dependent on the values of RBC and Hb.

The results in our study coincided with studies conducted by Pekelharing et al,[19] Wakeman et al,[20] in which they showed statistically significant reduction of reference intervals of Hb, RBC, HCT, MCH, MCHC while increase in PLT and PCT in females compared to males. No significant difference in WBC parameters were observed amongst males and females. Study by Rui Qiao[21] in 1259 Han individuals from North China also confirmed the mean values of RBC, Hb, HCT, MCV, MCH, and MCHC are higher in males; whereas, the PLTs are higher in females.

**Table 3:** Comparison of different parameters amongst males and females

| Variables | Gender | n (%) | Mean | SD | P value |
|---|---|---|---|---|---|
| **WBC** | Male | 163(45.2) | 7.5693 | 2.41623 | 0.536 |
| | Female | 198(54.8) | 7.7404 | 2.76464 | |
| **LYM** | Male | 163(45.2) | 2.6871 | 0.97513 | 0.643 |
| | Female | 198(54.8) | 2.7354 | 0.98998 | |
| **GRA** | Male | 163(45.2) | 3.9571 | 1.71702 | 0.376 |
| | Female | 198(54.8) | 4.1207 | 1.77178 | |
| **RBC** | Male | 163(45.2) | 4.7306 | 2.68114 | <0.001** |
| | Female | 198(54.8) | 4.0103 | 0.94242 | |
| **Hb** | Male | 163(45.2) | 12.3933 | 2.62328 | <0.001** |
| | Female | 198(54.8) | 10.4334 | 2.86172 | |
| **HCT** | Male | 163(45.2) | 36.2049 | 7.45234 | <0.001** |
| | Female | 198(54.8) | 30.0465 | 6.75814 | |
| **MCV** | Male | 163(45.2) | 79.2301 | 11.98352 | <0.001** |
| | Female | 198(54.8) | 74.7005 | 10.46623 | |
| **MCH** | Male | 163(45.2) | 28.2442 | 4.04556 | <0.001** |
| | Female | 198(54.8) | 25.9293 | 4.29646 | |
| **MCHC** | Male | 163(45.2) | 34.9920 | 3.31046 | 0.004* |
| | Female | 198(54.8) | 34.0939 | 2.63112 | |
| **RDW** | Male | 163(45.2) | 15.8362 | 2.33869 | 0.486 |
| | Female | 198(54.8) | 15.9935 | 1.94334 | |
| **PLT** | Male | 163(45.2) | 236.87 | 94.308 | 0.007* |
| | Female | 198(54.8) | 263.49 | 91.424 | |
| **PCT** | Male | 163(45.2) | .22834 | 0.105365 | 0.004* |
| | Female | 198(54.8) | .26273 | 0.117285 | |
| **MPV** | Male | 163(45.2) | 9.4663 | 0.73612 | 0.605 |
| | Female | 198(54.8) | 9.4260 | 0.73776 | |

$p < 0.05$ - Significant*, $p < 0.001$ - Highly significant**

**Table 4:** Reference range of haematological parameters

| Variables | Median | SD | Reference range |
|---|---|---|---|
| WBC | 7.4000 | 2.61094 | 4.79-10.01 |
| LYM | 2.6000 | .98224 | 1.62-3.58 |
| GRA | 3.8000 | 1.74676 | 2.06-5.54 |
| RBC | 4.1300 | 1.96207 | 2.17-6.09 |
| Hb | 10.9000 | 2.92096 | 7.98—13.82 |
| HCT | 31.3000 | 7.70736 | 23.6-39.0 |
| MCV | 78.0000 | 11.38688 | 66.62-89.38 |
| MCH | 27.2000 | 4.33557 | 22.87-31.53 |
| MCHC | 34.5000 | 2.98667 | 31.52-37.48 |
| RDW | 15.4000 | 2.12933 | 13.28-17.52 |
| PLT | 245.00 | 93.553 | 151.45-338.55 |
| PCT | .22900 | .113212 | 0.11-0.33 |
| MPV | 9.4000 | .73627 | 8.67-10.13 |

According to Ginevra Biino et al.,[22] PLT count was similar in males and females until the age of 14 years, but as age advances females had steadily more PLT than males.

Greater understanding about normal and altered levels of blood parameters is essential as different parameters can be used as independent biomarker to investigate the prognosis of patient with various pathologic conditions. Elevated levels of RBC and HCT are associated with increased risk of coronary heart disease. MPV is an independent predictor of peripheral arterial disease, and could be used as a marker of inflammation in patients with peripheral arterial disease.[23] RDW, PLT count have diagnostic value in distinguishing patients with gastric carcinoma from those with intestinal metaplasia.[24] MCV and RDW are bio-markers that can be used as the predictive marker in patients with endometrial carcinoma.[25]

In literature, not many studies have been done on relation between blood parameters and oral lesions. Study by Ramesh el al, in 40 samples of OSMF patients showed significantly lower levels of Hb, RBC and PLT and

increased levels of erythrocyte sedimentation rate (ESR) and WBC than in controls.[26] So, it is put forward to subject the patients to hematological investigations as it may aid in early diagnosis and prognosis of the diseases.

To the best of our knowledge, less information is available on alteration of blood parameters in oral lesions, it is necessary to carry out such investigations, so that clinician will be updated regarding geographic, ethnic, gender, age wise variation of blood parameters in different oral lesions. Present study was done to analyze the complete hemogram amongst males and females and also to set a reference interval for the same. To employ the results the test should be performed on larger sample to allow a more precise estimate of the test so that it will be easier to assess the representativeness of the sample and to generalize the results.

## 5. Conclusion

The CBC is a rich collection of information about each blood cell rather than mere cell counts. Now a days, with new diagnostic tests and prognostic tools based on molecular analysis, it is important to not overlook the value of the tests clinicians have been ordering for generations. These blood parameters will not likely provide confirmative diagnostic or prognostic information, but when understood and used properly, they provide readily available, cost-effective, and useful data that can supplement and guide clinical decision making. It is important to periodically update reference intervals for better correlation with health and disease conditions. Our findings would serve to update present data available data pertaining to blood parameters, their reference range and gender variation on a regional level.

## 6. Source of Funding

None.

## 7. Conflict of Interest

The authors declare no conflict of interest.

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