

Assessment of Blood and Renal Function Profiles in Pregnant Women from Narowal, Pakistan

Fizza Azeem^{1,#}, Rabia Mehmood^{2,3,#}, Rahat Raza Rasheed^{4,#}, Amna Rehman^{3,#}, Muhammad Zohaib⁵, Suneela Amaan⁶, Zoya Asif¹, Tayyba Jan⁷, Hanan Afzal², Sadia Ahmad³, Kaleem Maqsood⁸, Ali Afzal⁹, Muddasir Hassan Abbasi^{4,*}, Nadeem Sheikh^{3,*}, Muhammad Babar Khawar^{1,4,*}

Abstract

Pregnancy induces significant physiological changes that can impact hematological and renal function parameters which makes them potential indicators of gestational status. The objective of this study was to evaluate whether selected haematological and renal function parameters can serve as potential indicators of pregnancy. It was hypothesized that pregnancy would significantly alter these parameters compared to non-pregnant controls. Blood samples of pregnant women and controls were analyzed for hematological and renal function parameters. Sampling was conducted in hospitals and laboratories in Narowal, Punjab, Pakistan, from February 2021 to June 2022. Blood samples from 160 pregnant women and 160 controls were collected in EDTA-coated vials for Complete Blood Count analysis. Additionally, blood samples from 51 pregnant women and 51 controls were collected in serum vials to assess renal function. A statistically significant difference was observed in haematological parameters, including haemoglobin (Hb) ($t = 4.87$, $p < 0.0001$), haematocrit (HCT) ($t = 3.92$, $p = 0.0001$), and white blood cells (WBC) ($t = 2.76$, $p = 0.0063$). Renal function analysis revealed distinctive differences, with blood urea showing a significant increase in pregnant women

¹Applied Molecular Biology and Biomedicine Lab, Department of Zoology, University of Narowal, Narowal, Pakistan.

²Molecular Medicine and Cancer Therapeutics Lab, Department of Zoology, Faculty of Science and Technology, University of Central Punjab, Lahore, Pakistan.

³Institute of Zoology, University of the Punjab, Lahore, Pakistan.

⁴Department of Zoology, University of Okara, Okara, Pakistan.

⁵Department of Zoology, Government College University, Lahore, Pakistan.

⁶University of Veterinary and Animal Sciences (UVAS), Lahore, Pakistan.

⁷Department of Zoology, Lahore College for Women University (LCWU), Lahore, Pakistan.

⁸Department of Biology, Lahore Garrison University, Lahore

⁹University of Chinese Academy of Sciences, Beijing, 100049, China.

¹⁰Institute of Translational Medicine, Medical College, Yangzhou University, Yangzhou, China.

*Corresponding author's E-mail: dr.muddasir@uo.edu.pk, nadeem.zool@pu.edu.pk, babarkhawar@yahoo.com

#These authors contributed equally to this study.

Received: 23 November 2024; Received in revised form: 07 July 2025; Accepted: 09 July 2025.

Available online: 28-07-2025

This is an open-access article.

DOI: <https://doi.org/10.24312/ucp-jst.02.02.428>

(mean \pm SD: 34.6 ± 8.2 mg/dL) compared to controls (28.1 ± 6.7 mg/dL), $t(100) = 3.76$, $p = 0.0002$, and serum creatinine levels also significantly higher in the pregnant group (0.89 ± 0.15 mg/dL) versus controls (0.76 ± 0.12 mg/dL), $t(100) = 3.70$, $p = 0.0003$. These findings suggest that specific hematological and renal function tests may serve as descriptive indicators of physiological changes during pregnancy. Further studies on larger populations are recommended to validate these results and provide more precise information.

Keywords: Pregnancy; Complete Blood Count; Hematology; Renal Functioning Test; Biomarker

1. Introduction

Pregnancy, also known as gravidity or gestation, is the period during which a woman's womb creates one or more babies (Chandra et al., 2012). Pregnancy is a period in which the fetus survives and grows in the body of the mother and the immune system of the mother body does not reject the implantation of a fetus (Mazza, 2007). This gestation period is alienated into 3 trimesters. The first trimester is of less than 14 complete weeks, the second trimester is 14 to 27 complete weeks, and the third trimester is of 28 complete weeks till delivery (Hurjui et al., 2017).

Certain physiological changes affect the alimentary canal, cardiovascular, pulmonary, and renal organs to provide nutrients and remove waste (Kaur et al., 2014). The fetus's blood volume increases to oxygenate vital organs (Osonuga et al., 2011), leading to hyperpigmentation specially in darker areas (Ahmed et al., 1993). Maternal blood pressure drops during the first and second trimesters, returning to baseline in the third trimester (Khellaf et al., 2012). During pregnancy, sodium retention increases due to factors like estrogens, cortisol, renin-angiotensin, aldosterone, and posture. Pregnancy affects potassium balance and excretion, with a less precise regulation mechanism than sodium (de Flamingh & van der Merwe, 1984). Estrogens and progesterone moderate potassium excretion, enhancing sodium reabsorption and acting as

mineralocorticoid antagonists (Lindheimer et al., 1987). Lactate dehydrogenase (LDH) levels may rise from pre-pregnancy levels in the first trimester to normal levels in the third trimester. Other authors claim that LDH has remained unchanged (Cattozzo et al., 2013). During the first trimester, significant variation in renal plasma flow and glomerular filtration rate were already reported (Teasdale & Morton, 2018).

There are many physiological, hematological and serological parameters that significantly fluctuate during pregnancy, which are otherwise an indication of diseased condition in non-pregnant women (Anwar et al., 2023; Chandra et al., 2012). Hematological parameters have been increasingly studied as disease-specific biomarkers across a variety of physiological and pathological conditions. Our previous investigations have demonstrated significant alterations in CBC profiles in patients with infectious diseases (Rasheed et al., 2022), inflammatory skin conditions (Nawaz et al., 2022), and renal pathologies (Butt et al., 2023). This suggests that the broad diagnostic potential of blood-based markers. Therefore, we aim to explore whether fluctuations in blood profiles and renal function tests may indicate pregnancy-related physiological changes. Pregnancy is related to hematological and renal alterations, such as changes in hemoglobin, white blood cells, and serum creatinine levels. While these parameters appeal promising biomarkers, further

validation in larger and more diverse populations is necessary.

2. Methodology

2.1. Subjects

Our case-control investigation involved 160 pregnant women and 160 healthy non-pregnant women as controls. An additional 102 blood samples were collected for renal function test (RFT) screening, comprising 51 samples from pregnant women and 51 from healthy controls. All participants provided written informed consent before enrollment. To minimize potential bias, all subjects were recruited exclusively from the same geographical region, specifically various cities in District Narowal, Pakistan. The study was approved by the Ethical Committee of the University of Narowal, Narowal, Pakistan.

2.2. Inclusion and exclusion criteria

Inclusion criteria for pregnant participants were: confirmed pregnancy by ultrasound, age between 18–40 years, and no known pre-existing chronic illnesses. Controls were age-matched, non-pregnant women with no history of chronic disease. Exclusion criteria for both groups included any known renal disease, hematological disorders, recent infections, or current use of medications affecting renal function or hematological parameters.

2.3. Sampling and Hematological Analyses

Three milliliters of venous blood were collected aseptically in EDTA-coated vials from all participants. Complete blood count (CBC) was performed on these samples using an automated hematology analyzer (XP-300, Sysmex Corporation, Japan). The analyzer was calibrated daily according to the instructions, and validation was performed using commercially available quality control materials to ensure accuracy and precision, as previously described (Butt et al., 2023).

2.4. Measurement of Serum Urea and Creatinine

Under aseptic conditions, hemolysis-free serum was separated. Ready-to-use kits (e.g., Pfizer) were used for estimation of serum urea and creatinine following the manufacturer's guidelines. All assays were performed in duplicate to confirm reproducibility. The analytical methods employed in this study were consistent with protocols established in our previous investigations involving CBC and renal function testing in clinical populations across Punjab (Afzal et al., 2024; Rasheed et al., 2022; Riasat et al., 2022).

2.5. Statistical Analysis

Data were analyzed using Prism GraphPad version 8. An unpaired t-test was applied to assess statistically significant differences between groups. A p-value less than 0.05 was considered statistically significant. In addition to p-values, 95% confidence intervals were calculated to evaluate the precision of estimated differences, and effect sizes (Cohen's d) were reported to indicate the magnitude of the observed effects.

3. Results

Investigation of complete blood count leads to the finding of certain significant changes. A statistically significant decrease was found in the levels of hemoglobin (Hb) and hematocrit (HCT) of pregnant women when compared with the non-pregnant women. Comparison by t test revealed a marked increase in count of white blood cells (WBCs) during pregnancy with a level of significance $p < 0.0001$ (Figure 1).

The results implies that the renal function test of the control and pregnant women samples showed a distinctive difference with the high variance in serum urea and creatinine. The levels of both markers i.e., serum urea and serum creatinine decreased significantly in pregnant women with a level of

significance $p=0.0002$ and $p=0.0003$ respectively (Figure 2).

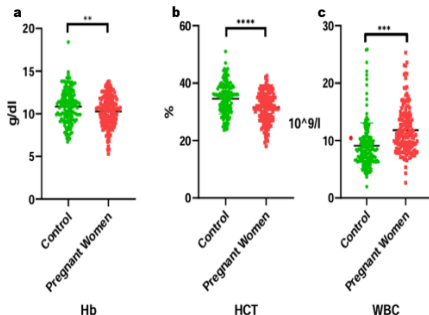


Figure 1 Hematological parameters (Hb, HCT, WBC) in pregnant women (n=160) vs controls (n=160). Data are presented in mean \pm S.E.M.; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$ (unpaired t-test).

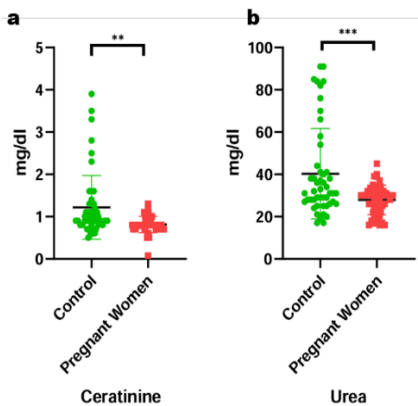


Figure 2 Serum creatinine and urea levels in pregnant women (n=51) and controls (n=51). Mean \pm S.E.M.; * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ indicate statistical significance.

4. Discussion

We observed notable differences in CBC parameters and RFTs between pregnant and non-pregnant women. Our results showed that hemoglobin and hematocrit levels were lower, while white blood cell counts were elevated during pregnancy. These findings align with previous reports indicating that pregnancy-related anemia is among the most prevalent hematologic issues faced by clinicians. The reduction in hemoglobin is likely due to increased iron requirements to support fetal growth and maternal blood volume expansion, which are often unmet

by dietary intake alone (Li et al., 2017). The current findings showing significant changes in hemoglobin, hematocrit, and WBC counts during pregnancy are consistent with our earlier work where similar CBC parameters were found to differentiate patients with hepatitis B and psoriasis from healthy individuals (Nawaz et al., 2022; Rasheed et al., 2022). The observed elevation in blood urea and serum creatinine among pregnant participants is aligned with previous findings from our lab, where such renal function markers potentially distinguish hemodialysis patients from healthy controls (Riasat et al., 2022).

The observed leukocytosis is consistent with established evidence showing a physiological rise in total WBC counts during pregnancy. Studies have demonstrated that neutrophil counts, in particular, increase steadily between 8 and 40 weeks of gestation, reflecting a response to physiological stress and hormonal changes (Chandra et al., 2012) (Dockree et al., 2021). Leukocytosis, a significant change in hematological profile during pregnancy, is caused by physiological stress and increases in white blood cell (WBC) count (Lurie et al., 2008). Moreover, leukocytosis has been reported to persist postpartum before gradually returning to pre-pregnancy levels over several weeks (Wadsworth, 2002) (Kaur et al., 2014). Our findings further support the concept that elevated neutrophil counts are a normal adaptive response during pregnancy, with counts potentially reaching up to twice those observed after delivery (Paidas & Hossain, 2010).

Regarding renal function, we found that serum urea and creatinine levels were significantly lower in pregnant women compared to controls. This is in agreement with earlier studies reporting decreased creatinine concentrations as a normal physiological change during pregnancy due to increased glomerular filtration rate (Girling, 2000). However, it is important

to interpret these reductions cautiously, as elevated levels can indicate underlying pathology. For instance, higher serum creatinine and blood urea nitrogen levels have been documented in pregnant women with pregnancy-induced hypertension (Tewabe & Wolde, 2020), while creatinine concentrations above 77 $\mu\text{mol/l}$ (0.87 mg/dl) may suggest acute kidney injury or previously unrecognized chronic kidney disease (Wiles et al., 2019). Lower urea and creatinine levels likely reflect increased glomerular filtration and renal clearance during pregnancy. These physiological adaptations help manage metabolic demands of the developing fetus.

Current study builds upon a body of work conducted by our research group, which has consistently shown that CBC and serum biomarkers offer valuable insight into physiological stress and systemic changes associated with various clinical conditions (Afzal et al., 2024; Butt et al., 2023; Nawaz et al., 2022; Rasheed et al., 2022; Riasat et al., 2022). Specifically, our earlier study on kidney stone patients emphasized how hematological profiles, including neutrophil-to-lymphocyte ratio (NLR), can serve as early indicators of renal complications (Butt et al., 2023), reinforcing the current study's use of such parameters to evaluate pregnancy-related renal shifts. Moreover, the use of CBC markers to detect immune or inflammatory imbalance, as in psoriasis and hepatitis B patients (Nawaz et al., 2022; Rasheed et al., 2022), supports the present analysis of immune-related hematological changes in pregnancy. These collective findings contribute to our ongoing effort to identify low-cost, non-invasive markers that may be useful for early screening or monitoring of various conditions in resource-limited settings. Although current study provides insightful results, there are certain limitations. Our study was limited by its relatively small sample size for renal function analysis, which may affect the generalizability of the findings.

Additionally, potential confounding factors such as dietary intake, hydration status, and underlying subclinical conditions were not controlled, which could influence hematological and renal parameters during pregnancy.

5. Conclusion

Our investigation gives proof of the differentiation between pregnant and normal women because of the CBC and renal profile results. The results of the CBC and renal functioning profile trial of normal women were altogether not quite the same as cases of pregnant women. As convenient biomarkers, hemoglobin, hematocrit, white blood cell count, serum urea, and serum creatinine can serve as useful indicators for evaluating physiological changes in pregnant women. These findings are important for future research and their potential application in clinical practice.

6. Declarations

6.1. Conflict of interest

The authors declare no potential conflicts of interest.

6.2. Acknowledgments

The authors would like to acknowledge the Biorender that aided in the creation of figure.

6.3. Funding statement

No funding was received for this study.

6.4. AI Tool Declaration

During the preparation of this work the authors used ChatGPT in order to simplify the content. After using this tool, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

Reference

Afzal, A., Kausar, R., Aslam, S., Shahid, N., Aman, S., Zohaib, M., Rehman, A., Afzal, N., Abbasi, M. H., & Sheikh, N. (2024). Clinical Biomarkers of Dyslipidemia in Diabetes Mellitus Type

- II Patients. *BioScientific Review*, 6(2), 8-19.
- Ahmed, Y., van Iddekinge, B., Paul, C., Sullivan, M. H., & Elder, M. G. (1993). Retrospective analysis of platelet numbers and volumes in normal pregnancy and in pre-eclampsia. *BJOG: An International Journal of Obstetrics & Gynaecology*, 100(3), 216-220.
- Anwar, S., Khawar, M. B., Ovais, M., Afzal, A., & Zhang, X. (2023). Gold nanocubes based optical detection of HIV-1 DNA via surface enhanced Raman spectroscopy. *Journal of Pharmaceutical and Biomedical Analysis*, 226, 115242. <https://doi.org/https://doi.org/10.1016/j.jpba.2023.115242>
- Butt, A. J., Khawar, M. B., Afzal, A., Bhalli, A. u., Hashmi, M. A. T. S., Afzal, N., Hamid, S. E., Shahzaman, S., Habiba, U., & Shah, S. S. (2023). Neutrophil-to-Lymphocyte ratio and Monocyte-to-HDL ratio as a Biomarker of Urolithiasis. *Comparative Clinical Pathology*, 32(5), 783-788. <https://doi.org/10.1007/s00580-023-03488-9>
- Cattozzo, G., Calonaci, A., Albeni, C., Guerra, E., Franzini, M., Ghezzi, F., & Ceriotti, F. (2013). Reference values for alanine aminotransferase, α -amylase, aspartate aminotransferase, γ -glutamyltransferase and lactate dehydrogenase measured according to the IFCC standardization during uncomplicated pregnancy. *Clinical Chemistry and Laboratory Medicine (CCLM)*, 51(10), e239-e241.
- Chandra, S., Tripathi, A. K., Mishra, S., Amzarul, M., & Vaish, A. K. (2012). Physiological changes in hematological parameters during pregnancy. *Indian journal of hematology and blood transfusion*, 28, 144-146.
- de Flamingh, J., & van der Merwe, J. (1984). A serum biochemical profile of normal pregnancy. *South African Medical Journal*, 65(14), 552-555.
- Dockree, S., Shine, B., Pavord, S., Impey, L., & Vatish, M. (2021). White blood cells in pregnancy: reference intervals for before and after delivery. *EBioMedicine*, 74.
- Girling, J. C. (2000). Re-evaluation of plasma creatinine concentration in normal pregnancy. *Journal of Obstetrics and Gynaecology*, 20(2), 128-131.
- Hurjui, L., Hurjui, I., Moscu, M., Popovici, D., & Gradinaru, I. (2017). Updates in relation between oral health and physiological changes in pregnancy. *Romanian Journal of Oral Rehabilitation*, 9(4), 18-24.
- Kaur, S., Khan, S., & Nigam, A. (2014). Hematological profile and pregnancy: a review. *Int J Adv Med*, 1(2), 68-70.
- Khellaf, M., Loustau, V., Bierling, P., Michel, M., & Godeau, B. (2012). Thrombocytopenia and pregnancy. *La Revue de Medecine Interne*, 33(8), 446-452.
- Li, A., Yang, S., Zhang, J., & Qiao, R. (2017). Establishment of reference intervals for complete blood count parameters during normal pregnancy in Beijing. *Journal of clinical laboratory analysis*, 31(6), e22150.
- Lindheimer, M., Richardson, D., Ehrlich, E., & Katz, A. (1987). Potassium homeostasis in pregnancy. *The Journal of reproductive medicine*, 32(7), 517-522.
- Lurie, S., Rahamim, E., Piper, I., Golan, A., & Sadan, O. (2008). Total and differential leukocyte counts percentiles in normal pregnancy. *European Journal of Obstetrics & Gynecology and Reproductive Biology*, 136(1), 16-19.
- Mazza, J. J. (2007). Insights into Global Nutritional Anemia Nutritional Anemia. Kraemer K, Zimmermann M, eds. 399 pp. \$35 donation to the UN World Food Programme "school feeding" is requested. ISBN 978-3-906412-33-4. Basel, Switzerland: Sight and Life Press, 2007; The Guidebook: Nutritional Anemia. Badham J, Zimmermann MB,

- Kraemer, K, eds. 50 pp. Free of charge. ISBN 978-3-906412-35-1. Basel, Switzerland: Sight and Life Press, 2007. *Clinical Medicine & Research*, 5(4), 209-209.
- Nawaz, B., Khawar, M. B., Afzal, A., Aman, S., Hamid, S. E., Abbasi, M. H., Sheikh, N., Shah, S. S., Rafiq, M., & Azam, F. (2022). Neutrophil-to-lymphocyte ratio as a promising biomarker for the diagnosis of psoriasis in Pakistan. *Biomedical Research and Therapy*, 9(9), 5306-5312.
- Osonuga, I., Osonuga, O., Onadeko, A., Osonuga, A., & Osonuga, A. (2011). Hematological profile of pregnant women in southwest of Nigeria. *Asian Pacific Journal of Tropical Disease*, 1(3), 232-234.
- Paidas, M. J., & Hossain, N. (2010). Hematologic changes in pregnancy. *Hemostasis and thrombosis in obstetrics & gynecology*, 1-11.
- Rasheed, H., Khawar, M. B., Habiba, U., Aman, S., Shah, S. S., Afzal, A., Hamid, S. E., Abbasi, M. H., Sheikh, N., & Rafiq, M. (2022). Variations in Peripheral Hematological Parameters as a Diagnostic Biomarker of HBV Infection. *Asian Journal of Health Sciences*, 8(2), ID45-ID45.
- Riasat, M., Khawar, M. B., Habiba, U., Afzal, A., Shahzaman, S., Hamid, S. E., Aman, S., Abbasi, M. H., Sheikh, N., & Shah, S. S. (2022). Evaluation of trace elements in hemodialysis patients in Pakistan. *Asian Journal of Health Sciences*, 8(2), ID48-ID48.
- Teasdale, S., & Morton, A. (2018). Changes in biochemical tests in pregnancy and their clinical significance. *Obstetric medicine*, 11(4), 160-170.
- Tewabe, H., & Wolde, M. (2020). Assessment of Renal Function Tests and Serum Total Protein among Pregnant Women with Pregnancy-Induced Hypertension Attending in Asrade Zewudie Memorable Hospital (ASZMPH), Gojjam, Ethiopia: Case-Control Study. *J Clin Med Res*, 1(3), 1-19.
- Wadsworth, G. (2002). Blood-volume: a commentary. *Singapore medical journal*, 43(8), 426-431.
- Wiles, K., Bramham, K., Seed, P. T., Nelson-Piercy, C., Lightstone, L., & Chappell, L. C. (2019). Serum creatinine in pregnancy: a systematic review. *Kidney international reports*, 4(3), 408-419.