

Leukocyte and Thrombocyte Baseline Dynamics: Age and Gender-Related Physiological Variations in a Healthy Libyan Population

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ديناميكية الكريات البيضاء والصفائح الدموية: التغيرات الفسيولوجية المرتبطة بالعمر والجنس لدى عينة سليمة من سكان مدينة توبره، ليبيا

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Abstract:

Establishing region-specific baseline norms for circulating leukocytes (WBC) and thrombocytes (PLT) is basic to precise hematological evaluation and cell-mediated physiological profiling. Globally standardized clinical boundaries frequently overlook eco-geographic, environmental and regional genetic heterogeneity. This investigation aimed to define local reference standards and evaluate demographic-dependent cell kinetics (age and sex) across a cohort in Tobra, Eastern Libya. A cross-sectional study executed involving 101 healthy adult volunteers (50 males, 51 females; age range: 15–55 years). Confounding systemic influences eliminated through a stringent exclusion protocol (omitting pregnancy, lactation, vascular/renal pathologies, diabetes, tobacco use and pharmacological therapy) to isolate an uncompromised baseline. Hematological parameters profiled using automated analyzers, with statistical validation utilizing unpaired Student's t-test, One-Way ANOVA, Pearson correlation, and non-parametric 2.5th–97.5th percentile interval calculations. The cohort-wide mean concentration for leukocytes reached $7.28 \pm 2.03 \times 10^9/L$ (95%) Reference Interval [RI]: 3.70 - 11.62 $\times 10^9/L$, While, thrombocyte density averaged $248.32 \pm 52.74 \times 10^9/L$ (95%) RI: 163.50 - 361.00 $\times 10^9/L$. Sexual dimorphism was statistically absent between men and women for both leukocyte (7.25 ± 1.94 vs. $7.32 \pm 2.13 \times 10^9/L$, $P = 0.864$) and Thrombocyte lineage parameters (239.92 ± 49.95 vs. $256.55 \pm 54.58 \times 10^9/L$, $p = 0.114$). Age - stratified analysis (< 25, 25–39, 40–55 years) revealed a minor, non - big inverse path with advancing age in both cell populations ($r = -0.157$, $p = 0.118$ for WBC; $r = -0.155$, $p = 0.122$ for PLT). Conclusion: This research establishes the inaugural reference intervals for WBC and PLT profiles within the healthy adult population of Tobra, Eastern Libya. The findings highlight strong homeostatic stability across age brackets and gender divisions, providing an authentic regional baseline for diagnostic pathology and biomedical research.

Keywords: Leukocytes, Thrombocytes, Reference Intervals, Hematological Homeostasis, Tobra, Libya.

الملخص:

يعد تحديد معايير مرجعية أساسية خاصة بالمنطقة لمستويات كريات الدم البيضاء (WBC) والصفائح الدموية (PLT) أمراً جوهرياً لإجراء تقييم دقيق للأمراض الدم وتكوين ملف فسيولوجي يعتمد على خصائص الخلايا؛ إذ غالباً ما تغفل الحدود السريرية الموحدة عالمياً التباينات البيئية والجغرافية، فضلاً عن التباين الوراثي الإقليمي. وقد هدفت هذه الدراسة إلى تحديد معايير مرجعية محلية وتقييم حركية الخلايا المرتبطة بالخصائص الديموغرافية (العمر والجنس) لدى مجموعة من المشاركين في مدينة توكرة بشرق ليبيا. أجريت دراسة مقطعية شملت 101 متطوع بالغ يتمتعون بصحة جيدة (50 ذكراً و51 أنثى؛ وتراوح أعمارهم بين 15 و55 عاماً)، حيث تم استبعاد العوامل الجهازية المؤثرة والمربكة للنتائج من خلال بروتوكول صارم للاستبعاد (يشمل حالات الحمل والرضاعة، وأمراض الأوعية الدموية والكلية، وداء السكري، وتعاطي التبغ، والعلاجات الدوائية) وذلك لضمان الحصول على قيم مرجعية أساسية دقيقة وغير متأثرة بعوامل خارجية. تم تحديد المؤشرات الدموية باستخدام أجهزة تحليل آلية، مع إجراء التحليل الإحصائي عبر اختبار "ت" للعينات المستقلة (Student's t-test)، وتحليل التباين أحادي الاتجاه (One-Way ANOVA)، ومعامل ارتباط بيرسون، وحسابات النطاق المئوي (من 2.5 إلى 97.5) باستخدام الطرق غير المعلمية. بلغ متوسط تركيز كريات الدم البيضاء لدى المجموعة بأكملها $7.28 \pm 2.03 \times 10^9$ /لتر (بفاصل مرجعي [RI] بنسبة 95% يتراوح بين 3.70 و 11.62×10^9 /لتر)، في حين بلغ متوسط كثافة الصفائح الدموية $248.32 \pm 52.74 \times 10^9$ /لتر (بفاصل مرجعي بنسبة 95% يتراوح بين 163.50 و 361.00×10^9 /لتر). ولم يظهر اختلاف إحصائي مرتبط بالجنس بين الرجال والنساء فيما يتعلق بمؤشرات كريات الدم البيضاء (1.94 ± 7.25 مقابل $2.13 \pm 7.32 \times 10^9$ /لتر؛ قيمة $P = 0.864$) أو مؤشرات الصفائح الدموية (239.92 ± 49.95 مقابل $256.55 \pm 54.58 \times 10^9$ /لتر؛ قيمة $P = 0.114$). كما كشف التحليل المصنّف حسب الفئات العمرية (أقل من 25 عاماً، 25–39 عاماً، 40–55 عاماً) عن وجود ارتباط عكسي طفيف وغير دال مرتبط بتقدم العمر في كلتا مجموعتي الخلايا (معامل الارتباط $r = -0.157$ ، وقيمة $P = 0.118$ لكريات الدم البيضاء؛ و $r = -0.155$ ، وقيمة $P = 0.122$ للصفائح الدموية). يرسخ هذا البحث النطاقات المرجعية الأولية لمؤشرات خلايا الدم البيضاء (WBC) والصفائح الدموية (PLT) لدى البالغين الأصحاء في مدينة توكرة بشرق ليبيا. وتُبرز النتائج استقراراً قوياً في التوازن الحيوي عبر مختلف الفئات العمرية والجنسين، مما يوفر مرجعاً إقليمياً موثقاً لأغراض التشخيص والبحوث الطبية الحيوية.

الكلمات المفتاحية: الكريات البيضاء، الصفائح الدموية، النطاقات المرجعية، التوازن الدموي، توكرة، ليبيا.

Introduction:

Leukocytes (white blood cells) and thrombocytes (platelets) stand for core cellular effectors within the circulation, orchestrating immune surveillance, inflammatory signaling cascades, as well as hemostatic maintenance (Bain, 2015; Hoffbrand & Steensma, 2019). While leukocyte lineages drive both innate and adaptive cell-mediated immunity, thrombocytes preserve vascular integrity, start primary clot nucleation, and help with tissue regeneration (Gharoor & El-Kewafi, 2021). Leukocyte density (WBC) and thrombocyte concentrations (PLT) remains a foundation of routine clinical diagnostics, providing essential physiological insight into infectious etiologies, oncological evaluations, autoimmune disorders, and perioperative stratifications (Clinical and Laboratory Standards Institute [CLSI], 2008).

Despite the routine clinical utilization of automated complete blood count (CBC) platforms, many diagnostic facilities across developing nations rely heavily on reference boundaries provided by Western medical hardware manufacturers or global literature (El-Nagger & Aburawi, 2019). However, advanced insights in cell biology and eco - genetics highlight large population - level divergence in baseline hematological parameters, driven by geographical elevation, ethnic genetic architecture, dietary profiles, and environmental exposures. In addition, local microenvironments (Bain, 2015) applying external, non - tailored reference thresholds create a big risk of misclassifying healthy people with benign leukopenia, physiological leukocytosis and borderline thrombocytopenia resulting in unwarranted clinical interventions or missed diagnostic windows.

Furthermore, dynamic physiological shifts associated with sex and age directly influence circulating leukocyte and thrombocyte kinetics. Although sex steroid hormones (estrogen and testosterone) heavily govern, erythroid lineages, their precise regulatory roles in granulopoiesis, lymphopoiesis, and megakaryopoiesis remain actively investigated within cell biology (Hoffbrand & Steensma, 2019). Concurrently, chronological aging introduces subtle bone marrow involution and immunosenescence, potentially altering steady-state cell clearance and peripheral cell counts in disease-free cohorts (El-Nagger & Aburawi, 2019).

In Libya, epidemiological and cell-level physiological data detailing healthy reference boundaries remain remarkably limited, particularly across eastern coastal communities like Tobra. Existing regional reports frequently omit strict participant screening protocols or focus predominantly on erythrocytic parameters and hemoglobin disorders (Gharoor & El-Kewafi, 2021). To resolve this gap, the present

study evaluated baseline leukocyte and thrombocyte dynamics within a strictly selected, non-smoking, drug-free adult cohort in the city, assessing age- and sex-dependent physiological variations to establish regional, clinically valid reference intervals.

Materials and Methods:

Study Design and Inclusion Criteria:

A cross-sectional study was conducted in Tobra, a coastal city in eastern Libya. The study involved 101 healthy volunteers (50 males and 51 females; aged 15–55 years) randomly selected from the local community. Verbal consent was obtained from all participants. The inclusion and exclusion criteria were applied as follows:

- Gestational or lactating states in females.
- Clinically diagnosed chronic systemic pathologies (e.g., essential hypertension, diabetes mellitus, cardiovascular anomalies, renal insufficiency, or hepatic dysfunction).
- Concurrent acute infectious febrile or inflammatory syndromes during specimen acquisition.
- Current or previous history of tobacco consumption or smoking.
- Substance abuse or active administration of pharmacological agents known to alter bone marrow kinetics or hematopoiesis (e.g., systemic corticosteroids, anti-platelet therapies, or immunosuppressive drugs).

Specimen Acquisition and Laboratory Analysis:

Peripheral venous blood (3.0mL) collected under aseptic conditions via clean antecubital venipuncture. Blood specimens immediately transferred into tubes containing dipotassium ethylenediaminetetraacetic acid (K2-EDTA) anticoagulant. To prevent cellular micro - aggregation. Blood containers gently inverted and processed within a 2-4 hours window post - collection to preserve cell morphology and viability. Automated hematology platforms quantified leukocyte density (10⁹/L) and thrombocyte concentrations (10⁹/L).

Statistical Analysis:

Data processing and statistical validation conducted using SPSS (version 26.0). Distributional normality evaluated using the Shapiro-Wilk test. Continuous variables were expressed as mean ± standard deviation (SD), median values and central non-parametric 95% reference boundaries (2.5th to 97.5th percentiles). Gender-based differences evaluated via independent Student's t-tests and non-parametric Mann-Whitney U tests. Inter-group variability across three age cohorts (<25, 25–39, and 40–55 years) was assessed using One-Way Analysis of Variance (ANOVA). Linear relationships between age progression and cellular parameters calculated using Pearson's correlation coefficient (r). Statistical significance established at p-value < 0.05.

Results:

The mean age of the entire reference group was 29.06 ± 12.41 years (ranging from 15 to 55 years). Demographic assessment revealed no statistically significant difference in age between the male (30.70 ± 12.71 years) and female (27.45 ± 12.01 years) subgroups (p = 0.190). Distribution analysis using the Shapiro-Wilk test confirmed the validity of these data; the assumptions of normal (parametric) distribution were met for both circulating leukocyte counts (p = 0.137) and platelet density distributions (p = 0.144).

Table (1): Baseline Demographic and Hematological Characteristics of the Study Participants (N=101)

Characteristic	Total Sample (N=101)	Male (n=50)	Female (n=51)	p-value
Distribution by Gender, n (%)				—
Male	50 (49,5%)	50 (100%)	—	—
Female	51 (50,5%)	—	51 (100%)	—
Age (Years)				
Mean ± SD	29,06 ± 12,41	30,70 ± 12,71	27,45 ± 12,01	0.190
Range (Min–Max)	15 - 55	—	—	—
Normality Assessment (Shapiro-Wilk Test)				
Total Leukocyte Count (WBC)	p = 0.137 (Normally Distributed)	—	—	—
Platelet Count (PLT)	p = 0.144 (Normally Distributed)	—	—	—

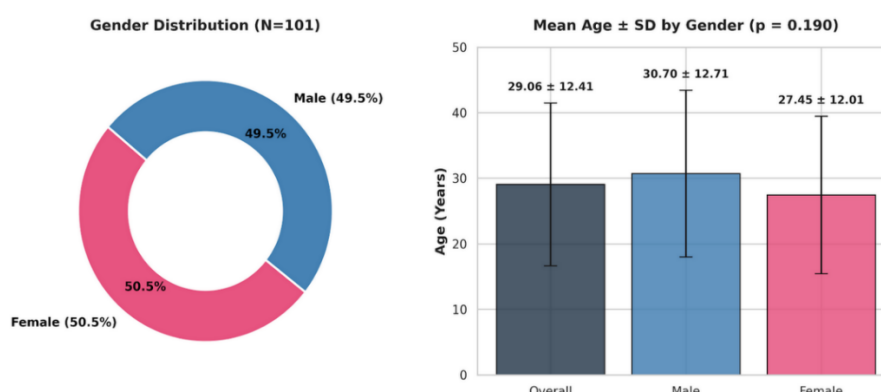


Figure (1): Shows Baseline Demographic and Hematological Characteristics

For the complete cohort, total leukocyte density averaged $7.28 \pm 2.03 \times 10^9/L$ (median: $7.10 \times 10^9/L$), establishing a 95% parametric reference boundary (2.5th–97.5th percentiles) of $3.70 - 11.62 \times 10^9/L$. Concurrently, circulating thrombocyte concentration demonstrated an overall mean of $248.32 \pm 52.74 \times 10^9/L$ (median: $243.00 \times 10^9/L$). Defining a localized reference window spanning $163.50 - 361.00 \times 10^9/L$.

A comparison of hematological parameters between the sexes revealed no statistically significant difference in white blood cell levels between male and female participants (7.25 ± 1.94 vs. $7.32 \pm 2.13 \times 10^9/L$; $p = 0.864$). Similarly, platelet counts were comparable between the two groups, with females showing a slightly higher mean count than males (256.55 ± 54.58 vs. $239.92 \pm 49.95 \times 10^9/L$); however, this difference was not statistically significant ($p = 0.114$).

Table (2): Descriptive Statistics and Reference Intervals of WBC and PLT Counts Disaggregated by Gender

Parameter	Total Cohort (N=101)	Males (n=50)	Females (n=51)	p-value*
Age (years)	29.06 ± 12.41	30.70 ± 12.71	27.45 ± 12.01	0.190
WBC Count ($10^9/L$)				
Mean $\pm$ SD	7.28 ± 2.03	7.25 ± 1.94	7.32 ± 2.13	0.864
Median	7.10	6.95	7.20	-
95% Reference Interval	3.70 - 11.62	4.41 - 11.33	3.65 - 11.38	-
Min – Max	3.50 - 13.82	3.50 - 13.82	3.60 - 12.50	-
Platelet Count ($10^9/L$)				
Mean $\pm$ SD	248.32 ± 52.74	239.92 ± 49.95	256.55 ± 54.58	0.114
Median	243.00	239.50	254.00	-
95% Reference Interval	163.50 - 361.00	161.57 - 339.85	170.25 - 373.50	-
Min – Max	140.00 - 386.00	140.00 - 368.00	151.00 - 386.00	-

*Independent Samples t-test.

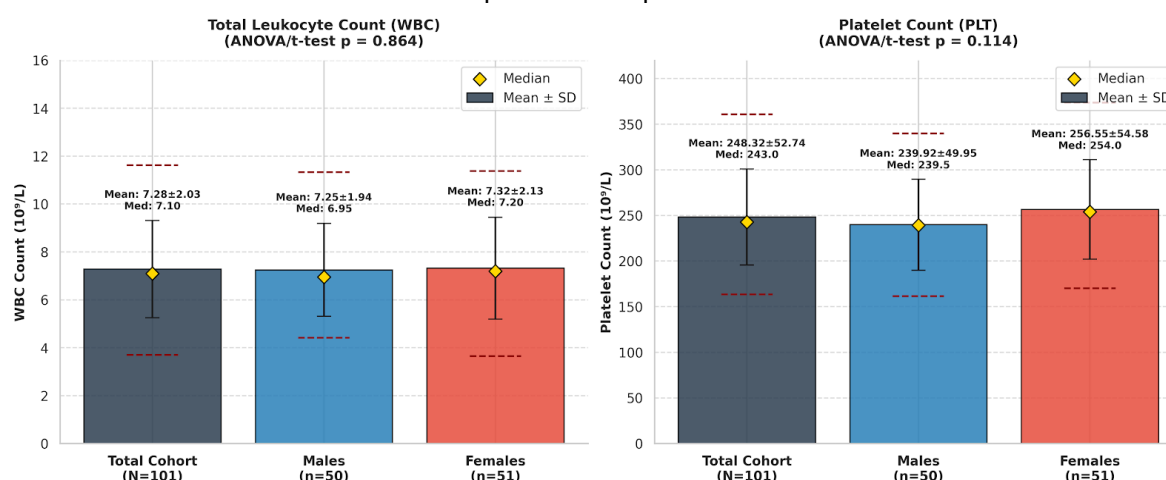


Figure (2): Shows the Demographic and Baseline Parameters (N = 101)

The study population was stratified into three age brackets: under 25 years (n = 52), 25–39 years (n = 21), and 40–55 years (n = 28). A one - way ANOVA revealed that neither white blood cell levels (F = 1.234, p = 0.296) nor platelet counts (F = 1.018, p = 0.365) varied a lot among the age categories. In addition, Pearson correlation testing demonstrated a slight, non-big negative association of age with both WBC (r = -0.157, p = 0.118) and PLT (r = -0.155, p = 0.122).

Table (3): WBC and Platelet Counts According to Age Stratification

Age Group (years)	Sample Size (n)	WBC Count (10 ⁹ /L) (Mean ± SD)	WBC Median	PLT Count (10 ⁹ /L) (Mean ± SD)	PLT Median
< 25	52	7.47 ± 2.11	7.35	254.42 ± 53.71	248.00
25 - 39	21	7.50 ± 1.92	7.40	248.57 ± 44.08	246.00
40-55	28	6.77 ± 1.92	6.45	236.79 ± 56.62	238.50
ANOVA (F-statistic)	-	F = 1.234	-	F = 1.018	-
p-value	-	0.296	-	0.365	-

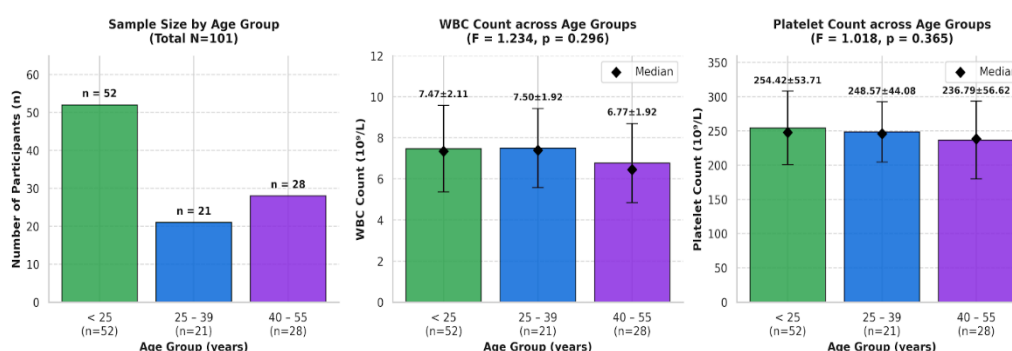


Figure (3): Shows the Hematological Parameters across Age Groups

Discussion:

Establishing accurate, region-specific reference baselines for complete blood count (CBC) parameters is crucial for distinguishing between normal physiological variation and silent (asymptomatic) diseases. The total leukocyte range determined in this study (3.70 – 11.62 X 10⁹/L) shows high concordance with benchmark standards, including Clinical and Laboratory Standards Institute (CLSI) guidelines (4.0 – 11.0 X 10⁹/L) and established hematological references (Bain, 2015; Hoffbrand & Steensma, 2019). Likewise, the derived platelet reference interval (163.50 – 361.00 X 10⁹/L) remains fully consistent with the broad physiological baseline (150 – 450 X 10⁹/L) recognized internationally.

Concerning sex-based variations, leukocyte and thrombocyte concentrations showed no statistically big divergence between male and female subjects (7.25 ± 1.94 vs. 7.32 ± 2.13 X 10⁹/L, p = 0.864 and 239.92 ± 49.95 vs. 256.55 ± 54.58 X 10⁹/L, p = 0.114, respectively). Mechanistically, sex hormones like testosterone mainly improve erythropoiesis through erythropoietin regulation, while, their modulating mess with on megakaryopoiesis, lymphopoiesis, and granulopoiesis is markedly less pronounced (Hoffbrand & Steensma, 2019). The slight, non - significant increase in female platelet levels aligns with earlier reports from Western Libya (El - Nagger & Aburawi, 2019) and Global cohorts (Biino et al., 2013). This mild elevation frequently linked to fluctuating estrogen levels and reduced systemic iron reserves in childbearing-aged females, of which both can even upregulate thrombopoietin pathways and drive megakaryocytic activity.

About age-dependent dynamics, variance analysis across the three age cohorts (<25, 25–39 — and 40–55 years) revealed a mild, non-big downward trend in both hematological lineages with increasing age (r = -0.157, p = 0.118 for WBCs; r = -0.155, p = 0.122 for PLTs). The reality is, at the cellular and histological level, the modest reduction of circulating mature elements in subjects above 40 years (6.77 X 10⁹/L for leukocytes and 236.79 X 10⁹/L for thrombocytes) reflects age-related bone marrow involution, in which functional red marrow is progressively replaced by fatty yellow marrow (Bain, 2015). In addition, immunosenescence alters the self-renewal capacity of hematopoietic stem cells, leading to a subtle decline in peripheral blood cell production while maintaining essential physiological homeostasis in healthy cohorts (Pantic et al., 2016).

Evaluating our findings against previous regional Libyan data (El-Nagger & Aburawi, 2019; Gharoor & El-Kewafi, 2021) shows the critical necessity of stringent cohort selection. Earlier investigations that recorded broader reference limits or elevated upper bounds frequently included active smokers or subjects with subclinical metabolic disorders. It is worth noting that cigarette smoke is a recognized

driver of systemic low-grade inflammation, leukocyte elevation, and vascular endothelial activation (Bain, 2015). By rigorously screening out these confounding variables, our established parameters offer a dependable diagnostic baseline for local practice, helping clinicians avoid mistaking normal physiological variants for pathological states such as leukopenia or thrombocytopenia within the Libyan population.

Conclusion and Recommendations:

Conclusion:

In summary, this investigation successfully defined population-specific baseline reference intervals for leukocytes ($3.70 - 11.62 \times 10^9/L$) and thrombocytes ($163.50 - 361.00 \times 10^9/L$) within a rigorously screened adult cohort in Tocra, Eastern Libya ($N = 101$). For the most part, enforcing strict exclusion criteria, omitting confounders like tobacco consumption, pregnancy, or even systemic pathology, ensured that these values stand for an unconfounded physiological benchmark. Statistical evaluations indicated no big disparities across age groups or sexes, despite minor physiological patterns, such as a slight female-predominant platelet trend and an age-associated decrease in cell lines. Implementing these local reference parameters in clinical settings will improve diagnostic precision and minimize false-positive classifications of leukopenia or thrombocytopenia in the regional population.

Recommendations:

Based on the outcome of this investigation, **several clinical and research directions are proposed:**

- **Clinical Integration:** Diagnostic laboratories and regional healthcare providers in Tocra are encouraged to adopt these localized reference parameters instead than relying solely on commercial kit inserts or global benchmarks.
- **Multi-Center Expansion:** Prospective investigations should increase sample size ($N > 500$) across diverse Libyan geographical regions to account for possible micro-environmental and demographic change.
- **Broadening Hematological Metrics:** Sub-fractional leukocyte analysis (neutrophils, lymphocytes, monocytes, eosinophils, basophils) and functional platelet parameters, such as mean platelet volume (MPV) and platelet distribution width (PDW), should be included in future cohort studies.
- **Environmental and Lifestyle Assessment:** Systematic epidemiological research warranted to assess how regional dietary patterns, altitude variations, and environmental exposures influence peripheral blood parameters in North African populations.

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