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ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

Медицинские новости Грузии

საქართველოს სამედიცინო სიახლენი

GEORGIAN MEDICAL NEWS

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GMN: Georgian Medical News is peer-reviewed, published monthly journal committed to promoting the science and art of medicine and the betterment of public health, published by the GMN Editorial Board since 1994. GMN carries original scientific articles on medicine, biology and pharmacy, which are of experimental, theoretical and practical character; publishes original research, reviews, commentaries, editorials, essays, medical news, and correspondence in English and Russian.

GMN is indexed in MEDLINE, SCOPUS, PubMed and VINITI Russian Academy of Sciences. The full text content is available through EBSCO databases.

GMN: Медицинские новости Грузии - ежемесячный рецензируемый научный журнал, издаётся Редакционной коллегией с 1994 года на русском и английском языках в целях поддержки медицинской науки и улучшения здравоохранения. В журнале публикуются оригинальные научные статьи в области медицины, биологии и фармации, статьи обзорного характера, научные сообщения, новости медицины и здравоохранения. Журнал индексируется в MEDLINE, отражён в базе данных SCOPUS, PubMed и ВИНИТИ РАН. Полнотекстовые статьи журнала доступны через БД EBSCO.

GMN: Georgian Medical News – საქართველოს სამედიცინო სიახლენი – არის ყოველთვიური სამეცნიერო სამედიცინო რეცენზირებადი ჟურნალი, გამოიცემა 1994 წლიდან, წარმოადგენს სარედაქციო კოლეგიისა და აშშ-ის მეცნიერების, განათლების, ინდუსტრიის, ხელოვნებისა და ბუნებისმეტყველების საერთაშორისო აკადემიის ერთობლივ გამოცემას. GMN-ში რუსულ და ინგლისურ ენებზე ქვეყნდება ექსპერიმენტული, თეორიული და პრაქტიკული ხასიათის ორიგინალური სამეცნიერო სტატიები მედიცინის, ბიოლოგიისა და ფარმაციის სფეროში, მიმოხილვითი ხასიათის სტატიები.

ჟურნალი ინდექსირებულია MEDLINE-ის საერთაშორისო სისტემაში, ასახულია SCOPUS-ის, PubMed-ის და ВИНИТИ РАН-ის მონაცემთა ბაზებში. სტატიების სრული ტექსტი ხელმისაწვდომია EBSCO-ს მონაცემთა ბაზებიდან.

WEBSITE

www.geomednews.com

К СВЕДЕНИЮ АВТОРОВ!

При направлении статьи в редакцию необходимо соблюдать следующие правила:

1. Статья должна быть представлена в двух экземплярах, на русском или английском языках, напечатанная через **полтора интервала на одной стороне стандартного листа с шириной левого поля в три сантиметра**. Используемый компьютерный шрифт для текста на русском и английском языках - **Times New Roman** (**Кириллица**), для текста на грузинском языке следует использовать **AcadNusx.** Размер шрифта - **12**. К рукописи, напечатанной на компьютере, должен быть приложен CD со статьей.

2. Размер статьи должен быть не менее десяти и не более двадцати страниц машинописи, включая указатель литературы и резюме на английском, русском и грузинском языках.

3. В статье должны быть освещены актуальность данного материала, методы и результаты исследования и их обсуждение.

При представлении в печать научных экспериментальных работ авторы должны указывать вид и количество экспериментальных животных, применявшиеся методы обезболивания и усыпления (в ходе острых опытов).

4. К статье должны быть приложены краткое (на полстраницы) резюме на английском, русском и грузинском языках (включающее следующие разделы: цель исследования, материал и методы, результаты и заключение) и список ключевых слов (key words).

5. Таблицы необходимо представлять в печатной форме. Фотокопии не принимаются. **Все цифровые, итоговые и процентные данные в таблицах должны соответствовать таковым в тексте статьи**. Таблицы и графики должны быть озаглавлены.

6. Фотографии должны быть контрастными, фотокопии с рентгенограмм - в позитивном изображении. Рисунки, чертежи и диаграммы следует озаглавить, пронумеровать и вставить в соответствующее место текста **в tiff формате**.

В подписях к микрофотографиям следует указывать степень увеличения через окуляр или объектив и метод окраски или импрегнации срезов.

7. Фамилии отечественных авторов приводятся в оригинальной транскрипции.

8. При оформлении и направлении статей в журнал МНГ просим авторов соблюдать правила, изложенные в «Единых требованиях к рукописям, представляемым в биомедицинские журналы», принятых Международным комитетом редакторов медицинских журналов - http://www.spinesurgery.ru/files/publish.pdf и http://www.nlm.nih.gov/bsd/uniform_requirements.html
В конце каждой оригинальной статьи приводится библиографический список. В список литературы включаются все материалы, на которые имеются ссылки в тексте. Список составляется в алфавитном порядке и нумеруется. Литературный источник приводится на языке оригинала. В списке литературы сначала приводятся работы, написанные знаками грузинского алфавита, затем кириллицей и латиницей. Ссылки на цитируемые работы в тексте статьи даются в квадратных скобках в виде номера, соответствующего номеру данной работы в списке литературы. Большинство цитированных источников должны быть за последние 5-7 лет.

9. Для получения права на публикацию статья должна иметь от руководителя работы или учреждения визу и сопроводительное отношение, написанные или напечатанные на бланке и заверенные подписью и печатью.

10. В конце статьи должны быть подписи всех авторов, полностью приведены их фамилии, имена и отчества, указаны служебный и домашний номера телефонов и адреса или иные координаты. Количество авторов (соавторов) не должно превышать пяти человек.

11. Редакция оставляет за собой право сокращать и исправлять статьи. Корректура авторам не высылается, вся работа и сверка проводится по авторскому оригиналу.

12. Недопустимо направление в редакцию работ, представленных к печати в иных издательствах или опубликованных в других изданиях.

При нарушении указанных правил статьи не рассматриваются.

REQUIREMENTS

Please note, materials submitted to the Editorial Office Staff are supposed to meet the following requirements:

1. Articles must be provided with a double copy, in English or Russian languages and typed or compu-ter-printed on a single side of standard typing paper, with the left margin of **3** centimeters width, and **1.5** spacing between the lines, typeface - **Times New Roman** (**Cyrillic**), print size - **12** (referring to Georgian and Russian materials). With computer-printed texts please enclose a CD carrying the same file titled with Latin symbols.

2. Size of the article, including index and resume in English, Russian and Georgian languages must be at least 10 pages and not exceed the limit of 20 pages of typed or computer-printed text.

3. Submitted material must include a coverage of a topical subject, research methods, results, and review.

Authors of the scientific-research works must indicate the number of experimental biological species drawn in, list the employed methods of anesthetization and soporific means used during acute tests.

4. Articles must have a short (half page) abstract in English, Russian and Georgian (including the following sections: aim of study, material and methods, results and conclusions) and a list of key words.

5. Tables must be presented in an original typed or computer-printed form, instead of a photocopied version. **Numbers, totals, percentile data on the tables must coincide with those in the texts of the articles.** Tables and graphs must be headed.

6. Photographs are required to be contrasted and must be submitted with doubles. Please number each photograph with a pencil on its back, indicate author's name, title of the article (short version), and mark out its top and bottom parts. Drawings must be accurate, drafts and diagrams drawn in Indian ink (or black ink). Photocopies of the X-ray photographs must be presented in a positive image in **tiff format**.

Accurately numbered subtitles for each illustration must be listed on a separate sheet of paper. In the subtitles for the microphotographs please indicate the ocular and objective lens magnification power, method of coloring or impregnation of the microscopic sections (preparations).

7. Please indicate last names, first and middle initials of the native authors, present names and initials of the foreign authors in the transcription of the original language, enclose in parenthesis corresponding number under which the author is listed in the reference materials.

8. Please follow guidance offered to authors by The International Committee of Medical Journal Editors guidance in its Uniform Requirements for Manuscripts Submitted to Biomedical Journals publication available online at: http://www.nlm.nih.gov/bsd/uniform_requirements.html
http://www.icmje.org/urm_full.pdf
In GMN style for each work cited in the text, a bibliographic reference is given, and this is located at the end of the article under the title "References". All references cited in the text must be listed. The list of references should be arranged alphabetically and then numbered. References are numbered in the text [numbers in square brackets] and in the reference list and numbers are repeated throughout the text as needed. The bibliographic description is given in the language of publication (citations in Georgian script are followed by Cyrillic and Latin).

9. To obtain the rights of publication articles must be accompanied by a visa from the project instructor or the establishment, where the work has been performed, and a reference letter, both written or typed on a special signed form, certified by a stamp or a seal.

10. Articles must be signed by all of the authors at the end, and they must be provided with a list of full names, office and home phone numbers and addresses or other non-office locations where the authors could be reached. The number of the authors (co-authors) must not exceed the limit of 5 people.

11. Editorial Staff reserves the rights to cut down in size and correct the articles. Proof-sheets are not sent out to the authors. The entire editorial and collation work is performed according to the author's original text.

12. Sending in the works that have already been assigned to the press by other Editorial Staffs or have been printed by other publishers is not permissible.

Articles that Fail to Meet the Aforementioned Requirements are not Assigned to be Reviewed.

ავტორთა საყურადღებოდ!

რედაქციაში სტატიის წარმოდგენისას საჭიროა დავიცვათ შემდეგი წესები:

1. სტატია უნდა წარმოადგინოთ 2 ცალად, რუსულ ან ინგლისურ ენებზე,დაბეჭდილი სტანდარტული ფურცლის 1 გვერდზე, 3 სმ სიგანის მარცხენა ველისა და სტრიქონებს შორის 1,5 ინტერვალის დაცვით. გამოყენებული კომპიუტერული შრიფტი რუსულ და ინგლისურენოვან ტექსტებში - **Times New Roman (Кириллица)**, ხოლო ქართულენოვან ტექსტში საჭიროა გამოვიყენოთ **AcadNusx.** შრიფტის ზომა – 12. სტატიას თან უნდა ახლდეს CD სტატიით.

2. სტატიის მოცულობა არ უნდა შეადგენდეს 10 გვერდზე ნაკლებს და 20 გვერდზე მეტს ლიტერატურის სიის და რეზიუმეების (ინგლისურ, რუსულ და ქართულ ენებზე) ჩათვლით.

3. სტატიაში საჭიროა გაშუქდეს: საკითხის აქტუალობა; კვლევის მიზანი; საკვლევი მასალა და გამოყენებული მეთოდები; მიღებული შედეგები და მათი განსჯა. ექსპერიმენტული ხასიათის სტატიების წარმოდგენისას ავტორებმა უნდა მიუთითონ საექსპერიმენტო ცხოველების სახეობა და რაოდენობა; გაუტკივარებისა და დაძინების მეთოდები (მწვავე ცდების პირობებში).

4. სტატიას თან უნდა ახლდეს რეზიუმე ინგლისურ, რუსულ და ქართულ ენებზე არანაკლებ ნახევარი გვერდის მოცულობისა (სათაურის, ავტორების, დაწესებულების მითითებით და უნდა შეიცავდეს შემდეგ განყოფილებებს: მიზანი, მასალა და მეთოდები, შედეგები და დასკვნები; ტექსტუალური ნაწილი არ უნდა იყოს 15 სტრიქონზე ნაკლები) და საკვანძო სიტყვების ჩამონათვალი (key words).

5. ცხრილები საჭიროა წარმოადგინოთ ნაბეჭდი სახით. ყველა ციფრული, შემაჯამებელი და პროცენტული მონაცემები უნდა შეესაბამებოდეს ტექსტში მოყვანილს.

6. ფოტოსურათები უნდა იყოს კონტრასტული; სურათები, ნახაზები, დიაგრამები - დასათაურებული, დანომრილი და სათანადო ადგილას ჩასმული. რენტგენოგრამების ფოტოასლები წარმოადგინეთ პოზიტიური გამოსახულებით **tiff** ფორმატში. მიკროფოტოსურათების წარწერებში საჭიროა მიუთითოთ ოკულარის ან ობიექტივის საშუალებით გადიდების ხარისხი, ანათალების შეღებვის ან იმპრეგნაციის მეთოდი და აღნიშნოთ სურათის ზედა და ქვედა ნაწილები.

7. სამამულო ავტორების გვარები სტატიაში აღინიშნება ინიციალების თანდართვით, უცხოურისა – უცხოური ტრანსკრიპციით.

8. სტატიას თან უნდა ახლდეს ავტორის მიერ გამოყენებული სამამულო და უცხოური შრომების ბიბლიოგრაფიული სია (ბოლო 5-8 წლის სიღრმით). ანბანური წყობით წარმოდგენილ ბიბლიოგრაფიულ სიაში მიუთითეთ ჯერ სამამულო, შემდეგ უცხოელი ავტორები (გვარი, ინიციალები, სტატიის სათაური, ჟურნალის დასახელება, გამოცემის ადგილი, წელი, ჟურნალის №, პირველი და ბოლო გვერდები). მონოგრაფიის შემთხვევაში მიუთითეთ გამოცემის წელი, ადგილი და გვერდების საერთო რაოდენობა. ტექსტში კვადრატულ ფჩხილებში უნდა მიუთითოთ ავტორის შესაბამისი N ლიტერატურის სიის მიხედვით. მიზანშეწონილია, რომ ციტირებული წყაროების უმეტესი ნაწილი იყოს 5-6 წლის სიღრმის.

9. სტატიას თან უნდა ახლდეს: ა) დაწესებულების ან სამეცნიერო ხელმძღვანელის წარდგინება, დამოწმებული ხელმოწერითა და ბეჭდით; ბ) დარგის სპეციალისტის დამოწმებული რეცენზია, რომელშიც მითითებული იქნება საკითხის აქტუალობა, მასალის საკმაობა, მეთოდის სანდოობა, შედეგების სამეცნიერო-პრაქტიკული მნიშვნელობა.

10. სტატიის ბოლოს საჭიროა ყველა ავტორის ხელმოწერა, რომელთა რაოდენობა არ უნდა აღემატებოდეს 5-ს.

11. რედაქცია იტოვებს უფლებას შეასწოროს სტატია. ტექსტზე მუშაობა და შეჯერება ხდება საავტორო ორიგინალის მიხედვით.

12. დაუშვებელია რედაქციაში ისეთი სტატიის წარდგენა, რომელიც დასაბეჭდად წარდგენილი იყო სხვა რედაქციაში ან გამოქვეყნებული იყო სხვა გამოცემებში.

აღნიშნული წესების დარღვევის შემთხვევაში სტატიები არ განიხილება.

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RETICULOCYTE SUBPOPULATION ANALYSIS AND ITS CORRELATION WITH IRON DEFICIENCY ANEMIA: A RETROSPECTIVE STUDY IN A PREDOMINANTLY FEMALE POPULATION

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Abstract.

Background: Serum ferritin and iron have high inter-individual variability and inflammatory sensitivity, limiting its diagnostic value in several clinical settings. The Reticulocyte Subpopulation Analysis may indicate iron availability during erythropoiesis in real time. **Aim:** This study investigated the associations between reticulocyte parameters and various hematological and iron-related biomarkers in Iron Deficiency Anemia. **Methods:** In this retrospective study, 150 patients with IDA (Iron Deficiency Anemia) were included (146 females, 97.3%; 23 pregnant, 15.8%; 12 on iron treatment, 8.0%), RET-He, LFR/MFR/HFR, and IRF were correlated with iron markers. Associations between anemia severity and treatment status were examined using Spearman correlation, Kruskal–Wallis, and Mann–Whitney U analyses. **Results:** While other reticulocyte indices exhibited minor connections, RET-He showed strong positive correlations with ferritin ($r=0.430$, $p<0.001$), serum iron ($r=0.364$, $p<0.001$), and transferrin saturation ($r=0.350$, $p<0.001$ A significant decrease in RET-He was seen in mild anemia (24.6 pg) and severe anemia (15.2 pg, $p<0.001$). RET-He was marginally reduced ($p=0.0750$) while serum iron and transferrin saturation were considerably lower in iron-treated subjects ($p=0.0023$). Reticulocyte and iron characteristics did not correlate with age ($p>0.05$). **Conclusion:** A sensitive, independent biomarker of iron status, RET-He measures functional iron availability during erythropoiesis. Strong correlation and severity-stratified discrimination support in populations where inflammation confounds ferritin interpretation, RET-He inclusion into iron deficient diagnostic methods. Prospective studies are needed to establish clinical thresholds and validate RET-He in chronic renal disease, inflammatory conditions, and pregnancy-related iron metabolism.

Key words. Biomarker, erythropoiesis, iron deficiency anemia, iron metabolism and reticulocyte hemoglobin content.

Introduction.

Iron deficiency anemias (IDA) is the most prevalent form of anemias globally, affecting approximately 25% of the world's population, with higher prevalence among women of reproductive age, children, and individuals in low-resource settings [1]. It is primarily caused by an inadequate supply of iron to meet the body's physiological demands, leading to impaired hemoglobin synthesis and reduced oxygen-carrying capacity of red blood cells the condition can result from dietary deficiency [2], chronic blood loss, malabsorption syndromes, or increased physiological demands during periods such as pregnancy and growth.

Clinically, IDA manifests with symptoms such as fatigue, pallor, general weakness, and decreased physical performance. In severe (chronic) cases, it may cause cognitive impairment, weakened immunity, and delayed psychomotor development, particularly in children. Laboratory findings typically include low hemoglobin (Hb) [3], reduced mean corpuscular volume (MCV), low serum ferritin, and elevated total iron-binding capacity (TIBC). Early detection and appropriate management of IDA are essential to prevent long-term complications [3].

The majority of the patients regardless of their complaints in hospitals are tested for CBC. After the CBC is done, the hemoglobin is going to show low for patients with anemia hemoglobin of under 12.1 g/dL for females and a hemoglobin of under 13.8 g/dL for males is considered anemic [2]. Establishing that the type of anemia is distinguished through MCV. MCV of lower than 80 femtolitre (fL) is considered as microcytic anemia. An iron profile is done to confirm the IDA knowing that anemia of chronic disease thalassemia are similar in laboratory findings with distinguishable tests between them [3].

Reticulocyte analysis has gained importance in evaluating bone marrow response to anemias and monitoring the effectiveness of iron therapy. Reticulocytes are immature RBCs that still contain residual RNA, and their presence in peripheral blood indicates active erythropoiesis [4]. With the advent of

modern automated hematology analyzers, it is now possible to classify reticulocytes into three subpopulations based on their RNA content: high fluorescence reticulocytes (HFR), medium fluorescence reticulocytes (MFR), and low fluorescence reticulocytes (LFR) [5].

In the context of IDA, these subpopulations provide valuable information about the kinetics of erythropoiesis and the body's response to iron availability [5]. HFR, which represent the youngest reticulocytes, may increase shortly after the initiation of iron therapy, serving as an early marker of bone marrow recovery. Studies have shown that reticulocyte indices, including reticulocyte hemoglobin content (CHr) [5], may reflect iron status more rapidly than traditional parameters like Hb or ferritin [5].

Therefore, analyzing the distribution of reticulocyte subpopulations in IDA patients not only enhances diagnostic accuracy but also offers a real-time indicator of therapeutic efficacy. This study aims to investigate the pattern of reticulocyte subpopulations in individuals with IDA and explore potential correlations with conventional iron markers, thereby contributing to more precise and timely patient care [4].

Iron deficiency anemia diagnosis and management depend heavily on understanding iron dynamics and erythropoiesis [6]. Reticulocyte subpopulation analysis, including immature and mature reticulocytes, offers a valuable tool in evaluating bone marrow activity and the body's response to iron availability. However, little is known about the specific correlation between reticulocyte subpopulation dynamics and markers of iron status, such as serum ferritin and serum iron levels.

This study aimed to assess whether reticulocyte-derived measures offer clinically relevant insights into iron status and functional iron availability during erythropoiesis in Iron Deficiency Anemia. The secondary objectives included examining the variability of these associations across different categories of anemia severity and evaluating the impact of iron supplementation on the reticulocyte-iron biomarker.

Materials and Methods.

This retrospective study was performed from 2024 to 2025 at Burjeel Royal Hospital in Al Ain, United Arab Emirates. The study comprised 150 adult patients diagnosed with iron deficiency anemia (IDA), aged 18 to 65 years.

Iron deficiency anemia was defined according to established laboratory criteria employed in clinical practice, including hemoglobin levels below 12.0 g/dL in women and below 13.0 g/dL in men, combined with biochemical indicators of iron deficiency comprising serum ferritin below 15 µg/L and/or transferrin saturation below 16%. Patients who had undergone blood transfusion in the prior three months, as well as those with malignancies, significant systemic comorbidities, myelodysplastic syndromes, or bone marrow diseases, were excluded.

SYSMEX XN-1000 was utilized for CBC and reticulocyte parameters (Low, Medium, and High Fluorescence Ratios; LFR, MFR, and HFR), and the Cobas e411-Cobas Integra, Cobas Pure were utilized for the quantification of iron, ferritin, UIBC, and TIBC.

The hematological and biochemical analyzers in the lab were calibrated in accordance with the manufacturer's instructions and the Burjeel Royal and Burjeel Royal Asharej hospitals' quality control standards.

Descriptive statistics were computed using STATA's summarize command. Simple linear regressions assessed the relationship between reticulocyte counts and individual iron indices. A p-value < 0.05 was considered statistically significant. To show variable relationships, scatter plots with regression lines and confidence intervals were created. Spearman's rank correlation coefficient, a nonparametric measure, analyzed statistical relationships because the data did not match normalcy criteria. The regression lines in the illustrations are for visual aid only and do not imply parametric linear regression assumptions were used.

Institutional review board in Gulf medical university (IRB: COHS-STD-77-Oct-2024) approval and Burjeel Institutional Review Board (IRB) were obtained before starting the study. The privacy of the patients was fully protected. Personal and medical information was anonymized (with numbers) and treated confidentially, ensuring that no identifying details are exposed throughout the research process.

Results.

The study comprised 150 patients with IDA, exhibiting a notable female predominance (146 females, 97.3%). The largest proportion was found in the reproductive and early middle-aged categories, especially in the 30-44 age group (44.7%), followed by the 18-29 age group (36.7%). Of the participants, 23 individuals (15.8%) were undergoing treatment, while only 12 participants (8.0%) were receiving iron treatment; 138 participants (92.0%) were untreated.

Discussion.

This retrospective study involving 150 patients with IDA (Iron Deficiency Anemia) identified significant associations between reticulocyte parameters and iron homeostasis, with reticulocyte hemoglobin content (RET-He) identified as the primary marker associated with functional iron status. The strong positive correlations identified between RET-He and serum ferritin ($r=0.430$, $p<0.001$), serum iron ($r=0.364$, $p<0.001$), and transferrin saturation ($r=0.350$, $p<0.001$) align with previous studies that designate RET-He as a sensitive, real-time biomarker of iron availability in erythropoiesis [7]. RET-He serves as an indicator of hemoglobin incorporation into newly released reticulocytes from the bone marrow, offering a 3–4-day physiological window for evaluating acute changes in iron metabolism. This presents a temporal advantage compared to traditional static markers like serum ferritin, which is characterized by significant inter-individual variability and sensitivity to inflammation [8].

The lack of substantial correlations between various reticulocyte fluorescence ratios (low fluorescence reticulocytes [LFR], medium fluorescence reticulocytes [MFR], immature reticulocyte fraction [IRF]) and iron parameters stands in contrast to the strong associations observed with RET-He, indicating differing biological relevance among reticulocyte subpopulations. This finding is consistent with recent mechanistic studies indicating that RET-He captures iron-

Table 1. *Baseline demographic and clinical characteristics of the study population.*

Characteristic	Category	N	%
Gender	*Male*	*4*	*2.7*
	Female	*146*	*97.3*
Age group (years)	*<18*	*3*	*2.0*
	18-29	*55*	*36.7*
	30-44	*67*	*44.7*
	45-59	*23*	*15.3*
	≥60	*2*	*1.3*
Treatment status	*On iron treatment*	*12*	*8.0*
	Not on iron treatment	*138*	*92.0*
Pregnancy status	*Non-pregnant*	*123*	*84.2*
	Pregnant	*23*	*15.8*
Total	*All patients*	*150*	*100.0*

Table 2. *Spearman correlations between reticulocyte indices and iron markers.*

Reticulocyte Index	Iron Marker	n	Spearman rho	p-value
HFR (%)	FERR (ug/L)	147	-0.048	0.56
	IRON (ug/dL)	148	-0.155	0.06
	TIBC (ug/dL)	114	0.155	0.10
	TRANSFERRIN SATURATION (%)	114	-0.229	0.014
	UIBC (ug/dL)	120	0.200	0.028
IRF (%)	FERR (ug/L)	147	0.044	0.59
	IRON (ug/dL)	148	-0.045	0.58
	TIBC (ug/dL)	114	0.110	0.24
	TRANSFERRIN SATURATION (%)	114	-0.107	0.25
	UIBC (ug/dL)	120	0.117	0.20
LFR (%)	FERR (ug/L)	147	-0.009	0.91
	IRON (ug/dL)	148	0.073	0.37
	TIBC (ug/dL)	114	-0.115	0.22
	TRANSFERRIN SATURATION (%)	114	0.146	0.12
	UIBC (ug/dL)	120	-0.135	0.14
MFR (%)	FERR (ug/L)	147	0.028	0.73
	IRON (ug/dL)	148	-0.011	0.89
	TIBC (ug/dL)	114	0.050	0.59
	TRANSFERRIN SATURATION (%)	114	-0.054	0.57
	UIBC (ug/dL)	120	0.053	0.56
RET-He (Pg)	FERR (ug/L)	147	0.430	< 0.001
	IRON (ug/dL)	148	0.364	< 0.001
	TIBC (ug/dL)	114	-0.108	0.2540
	TRANSFERRIN SATURATION (%)	114	0.350	< 0.001
	UIBC (ug/dL)	120	-0.223	0.0143

dependent hemoglobin synthesis with greater specificity than morphological reticulocyte classifications alone [9]. The observed negative correlation between high fluorescence reticulocytes (HFR) and transferrin saturation (r=-0.229, p=0.0143) indicates an inverse relationship that may suggest changes in reticulocyte maturation kinetics in iron-replete conditions or compensatory erythropoietic responses to fluctuations in iron availability.

Anemia Severity Stratification and Reticulocyte Phenotyping.

The notable reduction in RET-He from mild to severe anemia (24.6 pg [IQR 23.75-26.35] compared to 15.2 pg [IQR 14.07-18.23], p<0.001) indicates a progressively severe deficiency in hemoglobin content within reticulocytes, aligning with iron-restricted erythropoiesis. This gradient effect aligns with findings from hemolytic anemias and chronic kidney disease cohorts, where RET-He depression is associated with functional iron deficiency, despite variations in conventional iron parameters [10]. The differential expression of HFR across severity categories (p=0.0023), with higher proportions in moderate and severe anemia, may indicate a paradoxical retention of immature

Table 3. *Median (IQR) of reticulocyte indices by anemia severity with Kruskal–Wallis tests.*

Variable	Severity	n	Median (IQR)	p-value
LFR (%)	Mild	27	87.10 (79.85–89.70)	0.0146
	Moderate	111	80.70 (75.60–85.70)	
	Severe	12	81.05 (75.25–84.70)	
MFR (%)	Mild	27	10.50 (8.85–15.75)	0.0965
	Moderate	111	13.30 (11.25–15.60)	
	Severe	12	13.50 (11.05–15.55)	
HFR (%)	Mild	27	2.10 (1.20–6.45)	0.0023
	Moderate	111	4.90 (2.70–8.85)	
	Severe	12	5.35 (1.95–9.28)	
RET-He (Pg)	Mild	27	24.60 (23.75–26.35)	<0.001
	Moderate	111	22.30 (19.90–24.35)	
	Severe	12	15.20 (14.07–18.23)	
IRF (%)	Mild	27	12.60 (10.25–19.65)	0.0034
	Moderate	111	19.10 (14.10–24.20)	
	Severe	12	18.95 (15.30–24.75)	

Table 4. *Spearman correlations between age and reticulocyte/iron indices.*

Variable	n	Spearman rho	p-value
LFR (%)	150	-0.090	0.2754
MFR (%)	150	0.030	0.7129
HFR (%)	150	0.098	0.2339
RET-He (Pg)	150	-0.109	0.1828
IRF (%)	150	0.151	0.0654
IRON (ug/dL)	148	0.041	0.6167
FERR (ug/L)	147	-0.029	0.7305
UIBC (ug/dL)	120	0.083	0.3676
TIBC (ug/dL)	114	0.013	0.8936
TRANSFERRIN SATURATION (%)	114	0.063	0.5076

Table 5. *Mann–Whitney U tests for differences by treatment status.*

Variable	n1 (treated)	n2 (untreated)	U statistic	p-value
LFR (%)	12	138	541.0	0.0472
MFR (%)	12	138	1026.0	0.1712
HFR (%)	12	138	1124.5	0.0403
RET-He (Pg)	12	138	1085.5	0.0750
IRF (%)	12	138	1150.0	0.0259
RET-He (Pg)	12	138	1085.5	0.0750
IRON (ug/dL)	11	137	1171.0	0.0023
FERR (ug/L)	12	135	1064.0	0.0727
UIBC (ug/dL)	11	109	584.5	0.8951
TIBC (ug/dL)	9	105	627.0	0.1056
TRANSFERRIN SATURATION (%)	9	105	709.0	0.0131

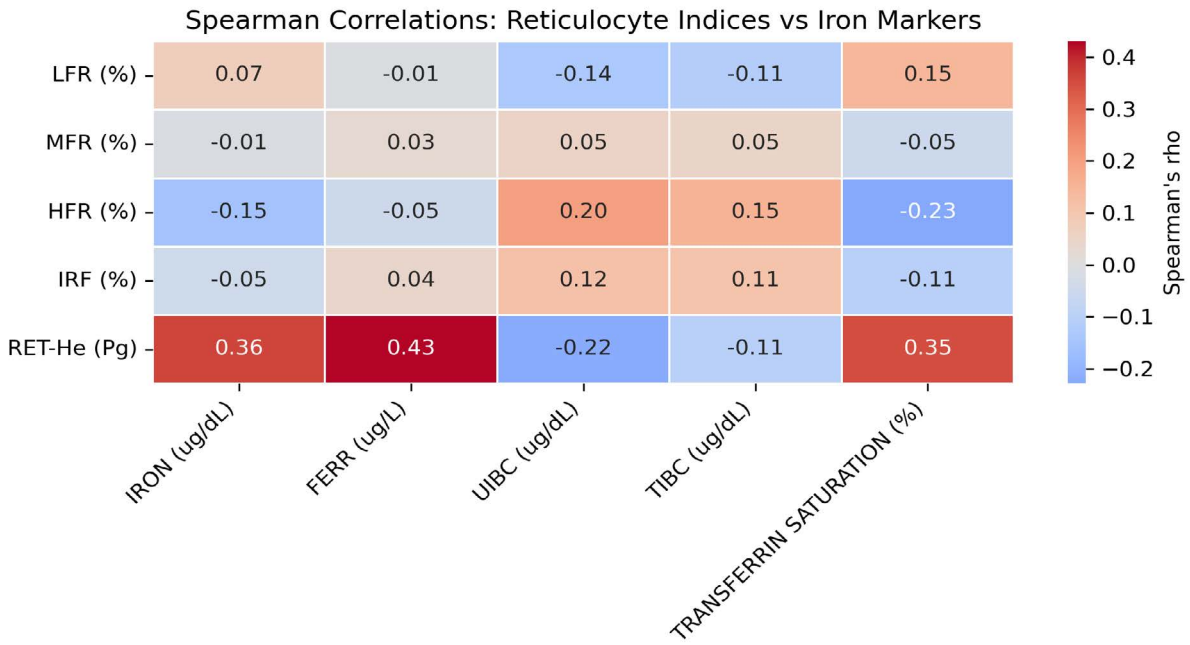

Figure 1. *Spearman correlations between reticulocyte indices and iron markers.*

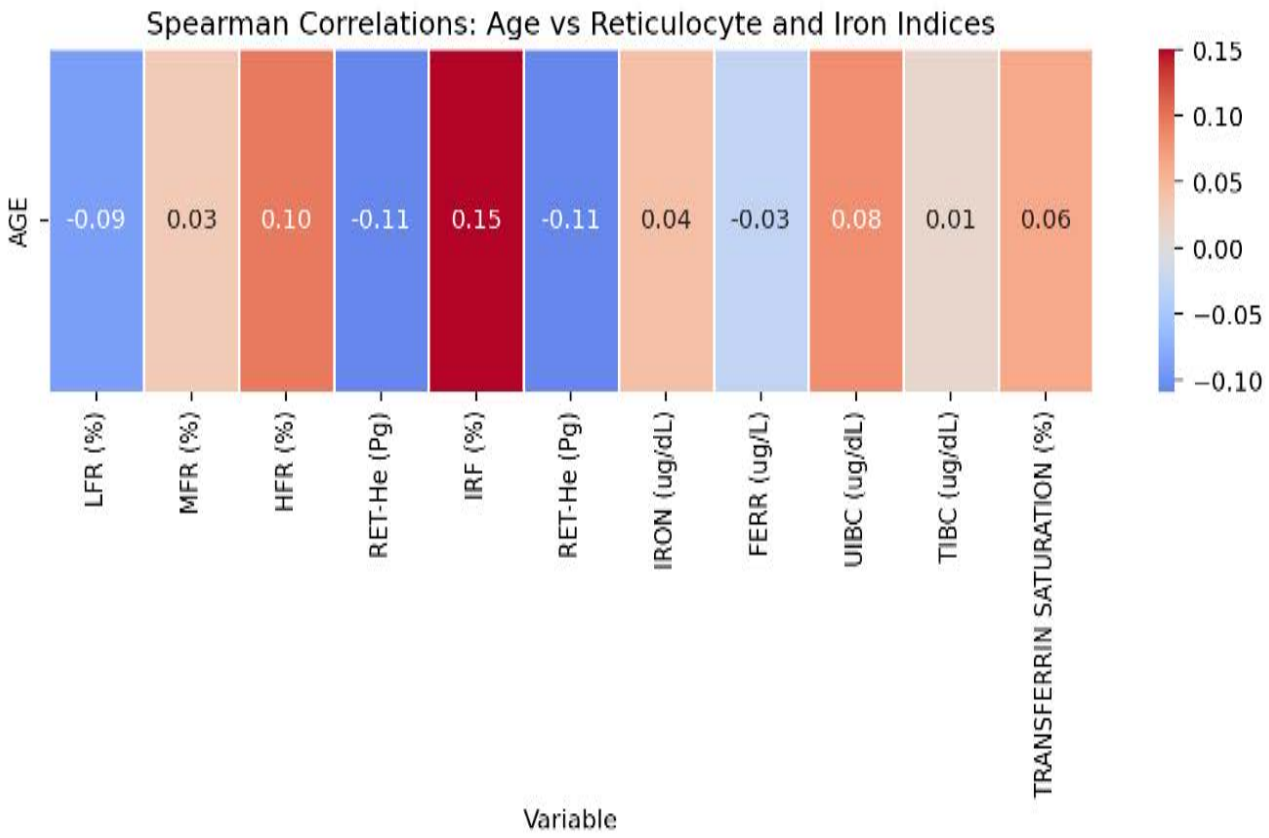

Figure 2. *Spearman correlations between age and reticulocyte/iron indices.*

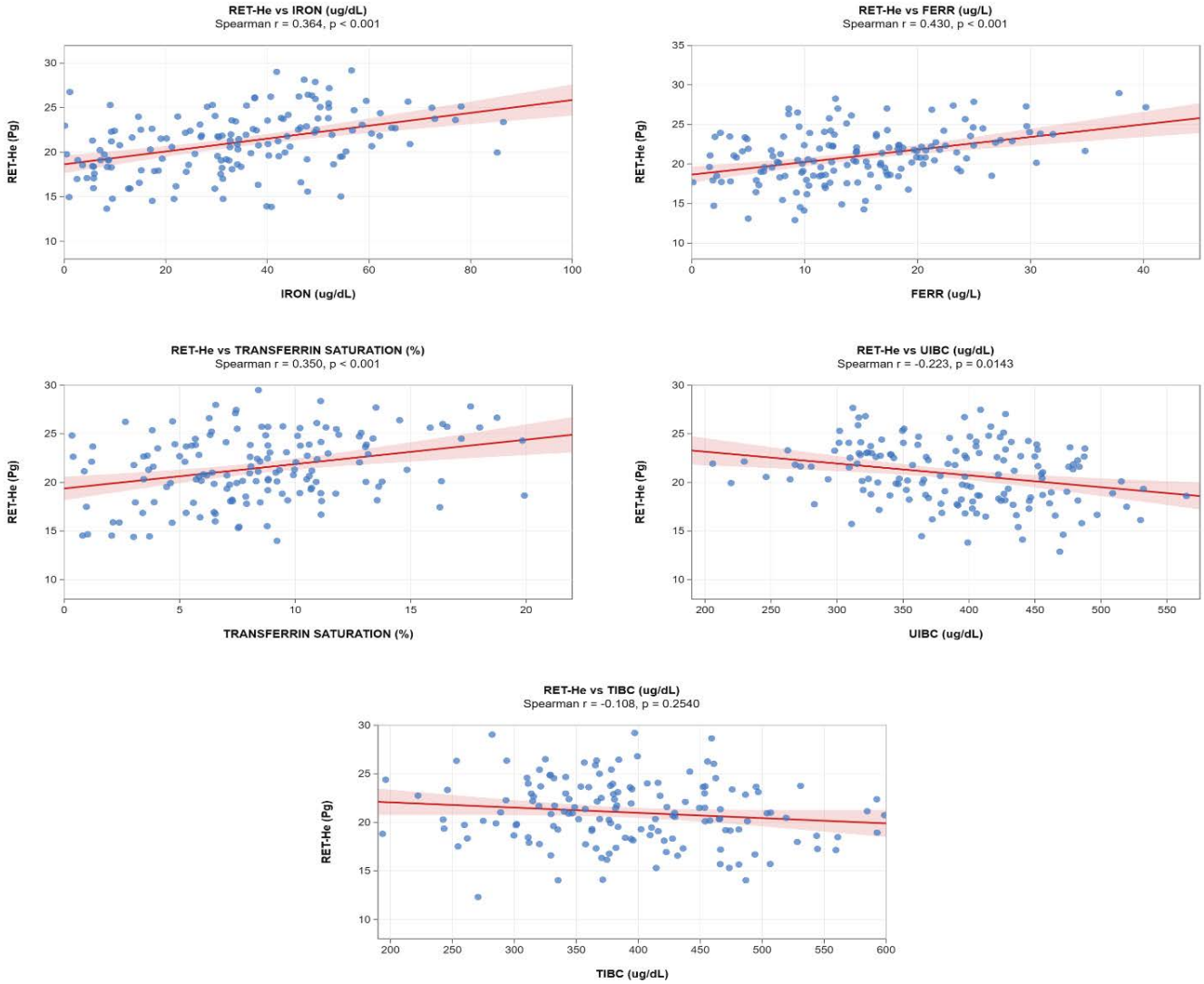

Figure 3. *Scatter plots of reticulocyte hemoglobin content (RET-He) versus iron status markers.*

reticulocyte phenotypes under erythropoietic stress, serving as a compensatory mechanism to enhance red cell release. The lack of notable age-related correlations with reticulocyte or iron indices (Table 4) indicates that reticulocyte parameters operate independently of chronological age, thereby increasing their relevance across various adult populations.

Treatment Response and Iron Supplementation Efficacy.

Participants administered iron supplementation (n=12) demonstrated reduced serum iron concentrations (p=0.0023) and transferrin saturation (TSAT) (p=0.0131) in comparison to untreated people, although ferritin levels were comparable between the groups (p=0.0727). Patients receiving iron treatment exhibited a marginally reduced RET-He value (p=0.0750, approaching the threshold of statistical significance), indicating potential incomplete iron repletion or ongoing functional iron insufficiency [11].

Nonetheless, these intergroup disparities must be treated with caution. A more plausible reason is severity bias, as individuals with greater iron deficiency are more inclined to obtain iron supplements. Consequently, the laboratory patterns seen may indicate the clinical state that necessitated the commencement of treatment rather than the biological impact of iron supplementation itself.

Due to the limited sample size of treated participants (n=12), the statistical power for between-group comparisons is constrained, precluding definitive conclusions about treatment response. From an epidemiological perspective, variations across cross-sectional groups should not be construed as proof of therapy efficacy. Longitudinal studies with higher sample numbers are essential for a more comprehensive assessment of alterations in blood iron parameters and RET-He subsequent to iron therapy.

Clinical and Diagnostic Implications.

The robust efficacy of RET-He as a singular predictor of iron status supports its incorporation into diagnostic algorithms for iron deficiency, especially in populations where the inflammatory reactivity of ferritin complicates interpretation. Studies examining inflammatory states, malignancy, and infection have shown that RET-He exhibits greater specificity

than conventional markers [12]. The high proportion of female participants (97.3%) and the inclusion of pregnant women (15.8%) provide a basis for further exploration of pregnancy-specific iron dynamics, given that pregnant individuals exhibit distinct erythropoietic adaptations that necessitate careful analysis of reticulocyte parameters [13].

Limitations and Methodological Considerations.

The pronounced gender disparity limits the external validity of findings to male populations and requires meticulous interpretation. The restricted treatment cohort (n=12, 8%) lacks adequate statistical power to make conclusive determinations regarding the efficacy of iron supplementation, underscoring the necessity for prospective randomized studies with sufficiently powered treatment groups. While hemoglobin values were accessible for anemia severity categorization, additional erythroid indices, including hematocrit and mean corpuscular volume, were absent from the dataset, hence constraining the thorough characterization of erythropoietic condition. Moreover, evaluating inflammatory markers as C-reactive protein and interleukin-6 would enable a stratified analysis to ascertain the diagnostic validity of RET-He more accurately in inflammatory situations.

Future Directions.

Longitudinal studies that include repeated measures of reticulocyte indices and iron biomarkers are crucial for establishing temporal relationships and causality. The application of machine learning algorithms to determine optimal RET-He thresholds for detecting iron deficiency in various clinical settings requires further examination. Evaluating RET-He's performance in populations with chronic diseases, such as chronic kidney disease, heart failure, and rheumatologic disorders, would clarify its role in complex iron metabolism contexts. Additionally, assessing reticulocyte parameters following intravenous iron therapy and erythropoiesis-stimulating agents would elucidate their effectiveness in monitoring therapeutic outcomes in specific populations.

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Conflict of interest.

The authors declare that there is no conflict of interest.

REFERENCES

1. McLean E, Cogswell M, Egli I, et al. Worldwide prevalence of anaemia, WHO Vitamin and Mineral Nutrition Information System, 1993-2005. Public Health Nutr. 2009;12:444-54.
2. Killip S, Bennett JM, Chambers MD. Iron Deficiency Anemia: Evaluation and Management. American Family Physician. 2013;87:98-104.
3. Johnson-Wimbley TD, Graham DY. Diagnosis and Management of Iron Deficiency Anemia in the 21st Century. Therapeutic Advances in Gastroenterology. 2011;4:177-184.
4. Rao RB, Lubach GR, Ennis-Czerniak KM, et al. Reticulocyte hemoglobin equivalent has comparable predictive accuracy as conventional serum iron indices for predicting iron deficiency and anemia in a nonhuman primate model of infantile iron deficiency. Journal of Nutrition. 2023;153:148-157.
5. Tariq H, Ujjan I, Shamsi TS. Evaluation of reticulocyte subpopulations and immature reticulocyte fractions in iron deficiency anemia. Journal of Population Therapeutics and Clinical Pharmacology. 2024;31:1556-1563.
6. Mayo Clinic Staff. Iron Deficiency Anemia – Diagnosis and Treatment. Rochester (MN): Mayo Clinic. 2025. https://www.mayoclinic.org/diseases-conditions/iron-deficiency-anemia/diagnosis-treatment/drc-20355040
7. Thomas L, Franck S, Messinger M, et al. Reticulocyte hemoglobin content: reference range, measurability, and clinical relevance in healthy adults and patients with mild anemia. Clinical Chemistry and Laboratory Medicine. 2015;43:835-841.
8. Brugnara C, Mohandas N, Otter W. Reticulocyte hemoglobin content (CHr): a new parameter to evaluate erythropoietic activity and iron status. Clinica Chimica Acta. 2018;396:1-7.
9. Hoffmann JJML, Nabbe KCAM, van den Broek NMA. Effect of age and gender on reference intervals of red blood cell distribution width (RDW) and mean corpuscular volume (MCV). Clinica Chimica Acta. 2020;397:32-36.
10. Pereira AA, Weir MR, Over P. Reticulocyte hemoglobin content as a marker of functional iron deficiency in hemodialysis patients. American Journal of Kidney Diseases. 2019;40:1101-1111.
11. Mast AE, Blinder MA, Gronowski AM, et al. Clinical utility of the reticulocyte hemoglobin content in the diagnosis of iron deficiency. Blood. 2002;99:1489-1491.
12. Mast AE, Blinder MA, Lu Q, et al. Clinical utility of the reticulocyte hemoglobin content in the diagnosis of iron deficiency. Blood. 2012;105:4649-4650.
13. Breymann C. Iron Deficiency Anemia in Pregnancy. Seminars in Hematology. 2015;52:339-347.