

Letter to Reviewers

Reviewer #1:

Scientific Quality: Grade D (Fair)

Language Quality: Grade B (Minor language polishing)

Conclusion: Major revision

Specific Comments to Authors: This study seems to be interesting, however, there are one important problem and several comments.

Authors: We thank the reviewer for his/her time and valuable suggestions.

Reviewer #1:

#1 This study was performed prospectively and this study needs the informed consent from patients and the ethics committee certificate of study protocol.

Authors: We thank the reviewer for the comment. The need for written informed consent was waived by the ethics committee due to the observational nature of the study. The ethics committee certificate of study protocol and the waiver of written informed consent have been submitted to the journal.

Reviewer #1:

#2 The authors should show the reasons of selecting patients of age > 65 years as the elderly patients.

Authors: We thank the reviewer for the comment. As stated in the manuscript "Elderly patients were selected due to the high prevalence of coexisting morbidities in this patient population" (*Page 5, Methods section*). Based on the traditional view patients with cardiovascular diseases are considered elderly if they are ≥ 65 years of age (J Geriatr Cardiol. 2016; 13(2): 115–117). Of note, the leading cause of death in those older than 65 years is heart disease, presenting challenges in diagnosis and

treatment (Circulation 2011; 123: e18-e209). The definition of older patients in the Hypertension guidelines (ACC/AHA 2017 and ESC/ESH 2018) was those aged above 65 years old (J Am Coll Cardiol 2018;71:2199-2269, J Hypertens 2018;36:1953-2041, <https://www.acc.org/latest-in-cardiology/articles/2020/02/26/06/24/older-adults-and-hypertension>). According to the most recent *Heart disease and stroke Statistics Report (2021)* the cut off value of 65 years has been used for several cardiovascular diseases such as kidney disease, sleep disorders, stroke, atrial fibrillation, valvulopathies, peripheral arterial disease, heart failure etc. (Circulation 2021;143(8):e254-e743). For these reasons the authors chose the 65 years as a cut off value for the definition of the elderly.

Reviewer #1:

#3 In the second paragraph of "Discussion" section, beginning with "In the current study RDW---", the meaning of this paragraph was unclear.

Authors: We thank the reviewer for the comment. The authors compared the RDW values ($RDW=12.6\pm0.8$) from a subgroup of elderly individuals (71-85 years old) of a historical cohort of 8089 individuals (Hoffmann JJ, et al. Clin Chem Lab Med. 2015 Nov;53(12):2015-2019) with the RDW found in the current study ($RDW=15.48\pm2.15$) **using summary data** and concluded that it was significantly higher in the examined population of the present study ($p < 0.0001$). The reason for this comparison can be found in the *limitations section* "Lack of a control group to compare RDW. However, a normal range of RDW value of 14.5% representing the upper normal limit is widely accepted (1, 6) and RDW was compared with an aged-matched historical control (6)."

This paragraph has been rewritten to avoid confusion.

“In the current study RDW was significantly higher than the RDW of a subgroup of elderly (i.e. 71-85 years old) (n=1479) of a historical cohort including a total of 8089 individuals (15.48 ± 2.15 vs. 12.6 ± 0.8 respectively, $p < 0.0001$) (6).”

Reviewer #1:

#4 Some values were expressed with the second decimal place, however, these expressions were meaningless to some extent. The authors should express values with proper decimal place.

Authors: We thank the reviewer for the comment. However, the presentation of the data and the statistical analysis results were provided by our professional statistician.

Reviewer #1:

#5 The whole of manuscript seems to be too lengthy. The authors should shorten the whole of manuscript, especially in the "Discussion" section.

Authors: Following the reviewer's thoughtful suggestion, the discussion section has been significantly shortened.

“DISCUSSION

In the present study including elderly patients hospitalized with CV disease RDW was significantly elevated. ADCHF was the most significant factor associated with RDW, whereas other important factors were AF and anemia.

In the current study RDW was significantly higher than the RDW of a subgroup of elderly (i.e. 71-85 years old) (n=1479) of a historical cohort including a total of 8089 individuals (15.48 ± 2.15 vs. 12.6 ± 0.8 respectively, $p < 0.0001$) (6).

An increased RDW mirrors a profound deregulation of erythrocyte homeostasis involving both impaired erythropoiesis and abnormal RBC survival and is used along with other RBC indices to help determine the causes of anemia (7). A

high RDW provides a clue for anisocytosis and/or the presence of two red cell populations, since other RBC indices (e.g., MCV or mean corpuscular hemoglobin concentration [MCH]) reflect average values and may not adequately reflect RBC changes where mixed RBC populations are present (e.g., dimorphic RBC populations in sideroblastic anemia or combined iron deficiency anemia [decreased MCV and MCH] and megaloblastic anemia [increased MCV]).

RDW has additionally been used as a prognosticator in diverse pathologies including CV disease (8). ~~In The Third National Health and Nutrition Examination Survey including 15852 adult participants, RDW was associated with risk of death due to CVD (HR 1.22, 95% CI 1.14–1.31), cancer (HR 1.28, 95% CI 1.21–1.36), and chronic lower respiratory disease (HR 1.32, 95% CI 1.17–1.49) (9).~~ Many of the conditions for which an increase in RDW was observed are associated with systemic inflammation and critical illness, but the exact pathophysiologic mechanisms underlying the association of increase in RDW with morbidity and mortality remains unclear (10). Given that erythropoietin is a key determinant of the RDW, it has been postulated that any condition affecting erythropoietin activity (eg, inflammation, primary renal disease, HF, bone marrow failure) may potentially lead to increased RDW values (11-13). Another consideration could be poor nutritional status, often present in patients with chronic diseases or critical illness, or nutrient deficiencies (eg, iron, vitamin B12, or folate deficiency) that are associated with anisocytosis (14). Other physiologic determinants that have been found to be associated with RDW changes include aging, black ethnicity, and physical exercise (6, 15).

The findings of the present study regarding the inflammatory nature of RDW elevation are contradictory. Inflammatory diseases (16) were both included in (e.g. AF and ADCHF) and excluded from (e.g. CKD, and COPD) the final model. Moreover, biomarkers of inflammation were unrelated (white blood cells) or weakly related (CRP) to RDW in univariable analysis and both were not included in the final model. The results of the studies on the relationship between RDW and inflammatory biomarkers have been conflicting. In the study of Lippi and colleagues including 3845 outpatients, when participants were grouped according to RDW quartiles, there were strong, graded increases in erythrocyte sedimentation rate (ESR) and hsCRP ($P < 0.001$), both parameters being up to 3-fold higher in the fourth versus the first quartile (17). In contrast, Lappe and colleagues observed a significant

but meaningless correlation between RDW and high-sensitivity CRP ($r = 0.181$; $p < 0.001$) in 1489 patients with CAD (18).

Inflammatory processes are present during the development and complications of CV disease. However, although there is a wealth of information about the role of inflammatory cells and pathways during acute injury and the reparative processes that are subsequently activated, little is known about the contribution of the immune system once the trajectory has been set, and chronic CV disease has been established – which clinically represents the majority of patients (19). The causative role inflammation plays in disease progression is not well defined, and the majority of clinical trials that target aspects of inflammation in patients with chronic CV disease have largely been negative (20, 21). This may be partly due to the fact that the tools currently used to measure ‘inflammation’ are insufficiently precise and do not provide information about disease site, activity, or discrimination between functionally important activation pathways (20, 22). ~~To address the limitations associated with assessing only a few select inflammatory biomarkers, some researchers have employed a multi-dimensional and multi-omics approaches (23).~~

Anisocytosis can be produced by any factor that increases erythropoiesis. In the present study, in contrast to anemia the two most important non-CV factors inducing anisocytosis, ADCHF and AF were associated with increased heart rate confirming that both are hyper-catecholaminergic states (24, 25). The nervous system emerges as a critical regulatory player of the bone marrow, the primary site of postnatal hematopoiesis and hematopoietic stem cell (HSC) maintenance, both under homeostatic and pathologic settings, with essential roles in cellular anchorage and egress, stem cell differentiation, and endothelial cell permeability (26, 27). Factors involved in erythropoiesis appear to revolve around the nervous system, and catecholamines seem to be the centerpiece. Several studies support the central role of the sympathetic nervous system (SNS) in the regulation of hematopoiesis (28, 29). Norepinephrine (NE) is delivered to the bone marrow (BM) by the sympathetic nerve in a circadian (diurnal) manner. ~~Circadian-linked changes in sympathetic nerve activity regulate the expression of core genes in the BM, leading to fluctuation of immune cell egress in both humans and rodents (30). Studies have also found a correlation between the proliferation of many HSCs and the circadian oscillation in~~

~~concentrations of NE in the BM (31). Thus, a~~ A close communication exists between the SNS and the BM and dysregulation in this communication may lead to aberrant hematopoietic and immune system responses (32)."

Reviewer #2:

Scientific Quality: Grade B (Very good)

Language Quality: Grade A (Priority publishing)

Conclusion: Minor revision

Specific Comments to Authors: The paper investigated the red blood cell distribution width (RDW) in elderly patients with cardiovascular disease, and the results show that RDW was significantly elevated in these patients. The findings in this paper are interesting and worth to be published.

Authors: We thank the reviewer for his/her time and valuable suggestions.

Reviewer #2:

Yet, the discussion part on immunity and inflammation has room for improvement. As what was pointed out by the authors, hypertension, coronary artery disease, diabetes mellitus and chronic obstructive pulmonary disease are among the most common morbidities in patients with elevated RDW. These morbidities all belong to metabolic syndromes. So, instead of considering poor nutritional status as mentioned in the manuscript, one may suspect the contribution of overnutrition to chronic diseases or critical illness. Human immunity is an essential part of the integrated nutrition acquisition and supply mechanism of the human body [1], especially when the person falls ill and inflammation is initiated in certain tissues [2, 3]. The human immune system initiates inflammation in response to tissue damage [4].

Inflammation is a protective immunological reaction to remove the injurious stimuli, and remove the damaged tissue as well as initiate the tissue healing process [4]. Yet, overnutrition will prevent the tissue healing process from happening. This is because the nutrition from the breakdown of the damaged tissue by inflammation together with the excessive nutrition already inside the body will be mostly turned into lipid

intermediates, causes lipotoxicity (the deposition of lipid intermediates in non-adipose tissue, leading to cellular dysfunction and death) [5] in healthy non-adipose tissues, and induces further tissue damage. The breakdown of non-adipose tissues and formation of lipid intermediates results in a vicious cycle. Thus, the overnutrition situation is worsened by the loss of lean body mass, coupled with perpetual chronic inflammation. The following reference may be included in the manuscript to provide a more broad picture of the topic discussed in this manuscript:

References: [1] Yu, LG (2021) Restoring Good Health in Elderly with Diverse Gut Microbiome and Food Intake Restriction to Combat COVID-19. *Indian Journal of Microbiology*, 61, 104–107. <https://doi.org/10.1007/s12088-020-00913-3> [2] van Niekerk G, Isaacs AW, Nell T, Engelbrecht AM (2016) Sickness-Associated Anorexia: Mother Nature's Idea of Immunonutrition? *Mediators of Inflammation* 2016 Article Number: 8071539. DOI: 10.1155/2016/8071539. [3] Arabi YM, Reintam BA, Preiser JC (2019) Less is more in nutrition: critically ill patients are starving but not hungry. *Intensive Care Medicine*, 45(11), pp. 1629-1631. DOI: 10.1007/s00134-019-05765-0 [4] Costantini S, Sharma A and Colonna G (2011). *The Value of the Cytokinome Profile, Inflammatory Diseases - A Modern Perspective*, Dr. Amit Nagal (Ed.), ISBN: 978-953-307-444-3, InTech, Available from: <http://www.intechopen.com/books/inflammatory-diseases-a-modern-perspective/the-value-of-the-cytokinome-profile> [5] Garbarino J, Sturley SL (2009) Saturated with fat: new perspectives on lipotoxicity. *Curr Opin Clin Nutr Metab Care* 12:110–116. DOI: 10.1097/mco.0b013e32832182ee

Authors: We would like to thank the reviewer for this very important comment. Accordingly, the discussion section has been edited as follows (Page 11, 1st paragraph):

“On the other hand, higher RDW has been associated with the Metabolic Syndrome (MS) [15, 16]. Proinflammatory cytokines have been found to inhibit erythropoietin-induced erythrocyte maturation, which may lead to increased RDW [16]. Overnutrition causes lipotoxicity in healthy non-adipose tissues, and induces tissue damage [17].”

EDITORIAL OFFICE'S COMMENTS

(1) *Science editor:* 1 Scientific quality: The manuscript describes a Prospective Study of the red blood cell distribution in elderly hospitalized patients. The topic is within the scope of the WJC. (1) Classification: Grade B and Grade D; (2) Summary of the Peer-Review Report: The findings in this paper are interesting and worth to be published. Authors may discuss the contribution of overnutrition to chronic diseases or critical illness. The authors should shorten the whole of manuscript. The questions raised by the reviewers should be answered; (3) Format: There are 4 tables and 1 figure; (4) References: A total of 35 references are cited, including 6 references published in the last 3 years; (5) Self-cited references: There are 2 self-cited references ; and (6) References recommendations (kindly remind): The authors have the right to refuse to cite improper references recommended by the peer reviewer(s), especially references published by the peer reviewer(s) him/herself (themselves). If the authors find the peer reviewer(s) request for the authors to cite improper references published by him/herself (themselves), please send the peer reviewer's ID number to editorialoffice@wjgnet.com. The Editorial Office will close and remove the peer reviewer from the F6Publishing system immediately. 2 Language evaluation: Classification: Grade A and Grade B. The manuscript is reviewed by a native English speaker. 3 Academic norms and rules: The authors should provided the the Institutional Review Board Approval Form, CONSORT 2010 Statement. Written informed consent was waived. No academic misconduct was found in the Bing search. 4 Supplementary comments: This is an invited manuscript. No financial support was obtained for the study. The topic has not previously been published in the WJC. 5 Issues raised: (1) The authors did not provide original pictures. Please provide the original figure documents. Please prepare and arrange the figures using PowerPoint to ensure that all graphs or arrows or text portions can be reprocessed by the editor; (2) PMID and DOI numbers are missing in the reference list. Please provide the PubMed numbers and DOI citation numbers to the reference list and list all authors of the references. Please revise throughout; and (3) The "Article Highlights" section is missing. Please add the "Article Highlights" section at the end of the main text. 6 Recommendation: Conditional acceptance.

(2) *Editorial office director:*

(3) *Company editor-in-chief:* I have reviewed the Peer-Review Report, full text of the manuscript, and the relevant ethics documents, all of which have met the basic publishing requirements of the World Journal of Cardiology, and the manuscript is conditionally accepted. I have sent the manuscript to the author(s) for its revision according to the Peer-Review Report, Editorial Office's comments and the Criteria for Manuscript Revision by Authors.

Authors: We thank the editors for the comments. The manuscript has been edited based on their valuable suggestions.

Response letter

Manuscript ID: 65348

“Red Blood Cell Distribution Width in Elderly Hospitalized Patients with Cardiovascular Disease”

Reviewer: As the authors have addressed all my concerns in my previous review report, I have no further comments except the following minor revision suggestions:

Authors: We are grateful to the reviewer for the encouraging comments

Reviewer: 1. Page 10, last line, “poor nutritional status” may be replaced by “nutritional imbalance”;

Authors: Per reviewer’s suggestion, “poor nutritional status” was replaced by “nutritional imbalance”.

Reviewer: 2. Page 11, line 1, “or nutrient deficiencies (eg, iron, vitamin B12, or folate deficiency) that are associated with anisocytosis [14].” may be replaced by “expressed by micronutrient deficiencies (eg, iron, vitamin B12, or folate deficiency) that are associated with anisocytosis [14], and excess of macronutrients.”

Authors: Based on the reviewer’s comment, the sentence was rewritten.

Reviewer: 3. Page 11, line 5, “Overnutrition” may be replaced by “Macronutrient surplus”

Authors: We agree with the reviewer. The word “Overnutrition” was replaced by “Macronutrient surplus”.

Once again we would like to thank the Reviewer and the Editor for their valuable comments