

## PEER REVIEW HISTORY

BMJ Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

### ARTICLE DETAILS

<b>TITLE (PROVISIONAL)</b>	Association of Regular Plasmapheresis Donation with Serum Protein and Electrolyte Levels: A Multi-centre Cross-Sectional Study in China
<b>AUTHORS</b>	Xiao, Guanglin; Li, Changqing; Chen, Yongjun; Song, Wenfu; Yang, Hui; Yang, Yating; Zhang, Yu; Pu, Zhongping; Wang, Xiufang; Xie, Shina; Yang, Shouqiang; Zeng, Jun; Li, Wan; Wang, Ya

### VERSION 1 - REVIEW

<b>REVIEWER NAME</b>	Evers, Josef
<b>REVIEWER AFFILIATION</b>	Octapharma Plasma
<b>REVIEWER CONFLICT OF INTEREST</b>	No competing/conflict of interests
<b>DATE REVIEW RETURNED</b>	07-Mar-2024

<b>GENERAL COMMENTS</b>	Unfortunately, I cannot recommend the diligent work for the high-impact BMJ because in accordance with the Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work <a href="https://icmje.org/icmje-recommendations.pdf">chromeextension://efaidnbmnnnibpcajpcglclefindmkaj/https://icmje.org/icmje-recommendations.pdf</a> . it has not been previously entered a trial register.
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<b>REVIEWER NAME</b>	Mercier-ythier, Jean
<b>REVIEWER AFFILIATION</b>	Université Paris 2 Panthéon-Assas Collège Européen de Paris, Economics
<b>REVIEWER CONFLICT OF INTEREST</b>	No competing interests
<b>DATE REVIEW RETURNED</b>	26-Mar-2024

<b>GENERAL COMMENTS</b>	I noted a small number of typos: p. 5, line 68: continuous p. 5, lines 87-88: two redundant sentences : "Concerns grow ..." p. 10, line 262, end: delete "is" : "We surmise that this may ..."
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<b>REVIEWER NAME</b>	Nissen-Meyer, Lise Sofie H.
<b>REVIEWER AFFILIATION</b>	Oslo University Hospital
<b>REVIEWER CONFLICT OF INTEREST</b>	No competing interests
<b>DATE REVIEW RETURNED</b>	29-Apr-2024

<b>GENERAL COMMENTS</b>	<p>Congratulations on an important and well-conducted study, with interesting and important data suitable to support changes in practice for plasma donors, at least in China. Donor health in plasmapheresis donors is an important field of research, particularly in view of the increased demand for PDMPs and the work for sustainable national plasma programs.</p> <p>I recommend the publication of this manuscript after a few minor revisions. See attached pdf for single comments and questions.</p> <p>Abstract: last sentence can be misleading, I recommend to rewrite to clarify the meaning.</p> <p>Statistics: satisfactory as far as I am able to review. Should not need further review if performed as described. However, I am not an expert in this.</p> <p>Language issues: The authors should allow for a thorough revision of the English language as there are a number of small errors that altogether annoy the reader and reduce the quality of the paper. See also suggestions from me in the attached pdf.</p> <p>In table and figure labels, you consistently and repeatedly use the phrase "New donors were as the control group". This phrase is incomplete in English, and a proper verb should be inserted following "were"; either "treated" or "considered" or something similar.</p> <p>The reference list needs a little more attention.</p>
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<b>REVIEWER NAME</b>	Thorpe, Rachel
<b>REVIEWER AFFILIATION</b>	Clinical Services and Research, Australia Red Cross Lifeblood
<b>REVIEWER CONFLICT OF INTEREST</b>	None
<b>DATE REVIEW RETURNED</b>	10-May-2024

<b>GENERAL COMMENTS</b>	<p>Thank you for the opportunity to review this article analysing the impact of plasmapheresis donation on donor health in a cohort of Chinese donors. There is value in measuring the impact of plasmaphereis donation on long-term and repeat donors given the increasing demand for plasma-derived medicines. It is especially valuable to analyse the impact of country-specific policies for plasmapheresis donation, such as maximum frequency, on donors. While this paper has promise, the writing and clarity need to be improved before publication.</p> <p>Title: Should the running head be "Impact of plasma donation on protein and electrolytes"?</p> <p>Abstract: Objectives: This sentence is not clear – needs a full stop?</p> <p>Conclusions: This section is currently unclear. The conclusions seem to contradict the results that found IgG levels were significantly lower in the high donation frequency group and that Hb is lower in female donors with high donation frequency and a high total number of lifetime donations. Can you suggest any recommendations to offset the</p>
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reduction in IgG levels in male and female donors and in Hb in female donors?

#### Introduction

This section should include a clear description of what plasmapheresis donation is, including what plasma-derived products are used for.

This section should also include a description of donor policies and procedures in China, such as whether donors are voluntary or remunerated, if remunerated what the rate is, whether donors can donate whole blood and plasma and platelets or just plasma and how donors come to donate plasma (eg is it because of their blood type, are they asked to, do they choose to).

The guidelines for plasmapheresis donation in Australia are very similar to China so perhaps this could be pointed out - the only difference is that the maximum number of donations per year is 26 compared with 24 in China. Data from Australian plasma donors may serve as a useful comparison.

Could the authors extrapolate upon the points that there are concerns about the health of plasma donors, such as explaining why this is a growing concern. The sentence “concerns about the health of plasma donors have risen” is a repeat of the previous sentence.

#### Study design and population

How were the participants approached for consent to participate in the research? Were participants provided with information about the study prior to consenting?

How were the definitions of low, medium and high donation frequencies and number of lifetime donations arrived at?

#### Inclusion and exclusion criteria

Second sentence – donors should be lower case

How was information about which supplements donors were taking obtained?

How did you establish levels of blood lipids, uric acid and cholesterol in donors?

#### Data collection:

How was the questionnaire data obtained – was this through paper surveys or online? Can you include a description of the process for obtaining questionnaire data? What was the completion rate for the questionnaire?

#### Quality control

The sentence “Upon completion of the collection process, serum samples were promptly chilled and avoid repeated freezing and thawing” seems to have a typo in it

#### Statistical analyses

Can you explain what is meant by “Literature-based covariates were identified by prior studies”?

#### Results

Can you include numbers for all results discussed in the text, such as mean age, BMI, household income and physical activity.

#### Discussion

The structure of the discussion could be improved. I would suggest starting out by discussing your findings, noting what is novel about them and then comparing them to other studies. Currently it is very unclear for the reader to understand how your results compare with other studies as, for example, the findings for TSP from your participants are not mentioned in the text either in results or discussion and the reader has to scroll down to Table 2 to understand how they compare with international findings.

	<p>The recommendations for changes to testing for donors could be placed into context more by including details of the testing currently conducted in the introduction. The recommendation for increasing the frequency of albumin screening for donors is unclear. Is this suggested because albumin is naturally lower in older people? As such, is increased frequency of screening suggested only for middle-aged to older donors?</p> <p>The discussion of findings for IgG is also unclear. Your results found significantly lower IgG levels in male and female donors compared with new donors but in the discussion it says "Although this study found no effect of plasma donation on low IgG rates, regular IgG monitoring of Chinese donors is still necessary given previous results that plasma donation affects IgG levels" – can this be explained more clearly.</p> <p>The findings of the impact of plasmapheresis on Hb in women also warrants more discussion. What is recommended to overcome the drop in Hb in women? Further testing? Reduced donation frequency?</p>
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### VERSION 1 – AUTHOR RESPONSE

Reviewer 1

Dr. Josef Evers, Octapharma Plasma:

Comment 1. Unfortunately, I cannot recommend the diligent work for the high-impact BMJ because in accordance with the Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work [chromeextension://efaidnbmnnnibpccjpcglclefindmkaj/https://icmje.org/icmje-recommendations.pdf](https://efaidnbmnnnibpccjpcglclefindmkaj/https://icmje.org/icmje-recommendations.pdf). it has not been previously entered a trial register.

Response 1: Thank you for your comments. Our study is an observational research, and while the ICMJE encourages registration of non-trial designs, it does not require it as the exposure or intervention is not dictated by the researchers. Additionally, our study was approved by The Ethics Committee of the Institute of Blood Transfusion, Chinese Academy of Medical Sciences (IBT) (No.2021042) prior to commencement.

Reviewer 2

Dr. Jean Mercier-ythier, Université Paris 2 Panthéon-Assas Collège Européen de Paris:

Comment 1. I noted a small number of typos:

- p. 5, line 68: continuous
- p. 5, lines 87-88: two redundant sentences : "Concerns grow ..."
- p. 10, line 262, end: delete "is" : "We surmise that this may ..."

Response 1: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comments regarding the typographical errors. The corrections made are as follows:

□Continuous plasma loss and anticoagulant use may impact donor physiology and protein levels. (Page 3, Lines 79-80)

□Donation serves the demand for plasma, but there are concerns among potential donors and the public about the impact of continuous blood loss and anticoagulant exposure during plasmapheresis process on physical health. (Page 4, Lines 98-100)

□We surmise that this may be due to the additive effect of the higher iron stores in men and menstruation in women. (Page 10, Lines 290-291)

We appreciate your time and effort in reviewing our work and believe that these revisions have improved the manuscript.

Reviewer 3

Dr. Lise Sofie H. Nissen-Meyer, Oslo University Hospital

Comment 1. I recommend the publication of this manuscript after a few minor revisions. See attached pdf for single comments and questions.

Response 1: Thank you for recommending the publication of our manuscript and for providing valuable feedback. We have carefully reviewed and addressed the minor revisions, typographical errors, and specific questions indicated in the attached PDF.

Comment 2. Abstract: last sentence can be misleading, I recommend to rewrite to clarify the meaning.

Response 2: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding the last sentence of the abstract. To ensure clarity, we have revised the sentence as follows:

Plasmapheresis donation is not associated with an increased risk of abnormalities in the analyzed parameters. However, the results provide preliminary evidence supporting the routine inclusion of IgG screening for donors, as plasmapheresis donation is associated with a decrease in IgG levels. Particular attention should be paid to the Hb levels of female donors, especially those who donate frequently. Testing of TSP at each donation may not be necessary. (Page 2, Lines 50-54)

Comment 3. Statistics: satisfactory as far as I am able to review. Should not need further review if performed as described. However, I am not an expert in this.

Response 3: Thank you for your feedback on our manuscript. We appreciate your assessment of the statistical methods used in our study. We welcome any further review or suggestions from statistical experts to confirm the robustness of our analyses.

Comment 4. Language issues: The authors should allow for a thorough revision of the English language as there are a number of small errors that altogether annoy the reader and reduce the quality of the paper. See also suggestions from me in the attached pdf.

Response 4: Thank you for your valuable feedback on our manuscript. We have thoroughly reviewed and corrected the language issues throughout the paper, addressing both the errors you highlighted and other minor errors to enhance the overall quality of the manuscript. We appreciate your attention to detail and believe that these revisions have significantly improved the clarity and readability of the paper.

Comment 5. In table and figure labels, you consistently and repeatedly use the phrase "New donors were as the control group". This phrase is incomplete in English, and a proper verb should be inserted following "were"; either "treated" or "considered" or something similar.

Response 5: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding the phrase used in the table and figure labels. The phrase "New donors were as the control group" has been revised to "New donors were considered as the control group" throughout the manuscript. We appreciate your attention to detail and believe that this revision has improved the clarity and accuracy of our labels.

Comment 6. The reference list needs a little more attention.

Response 6: Thank you for your valuable feedback on our manuscript. We have carefully reviewed the reference list and made the necessary corrections to ensure accuracy and consistency. All references have been checked and formatted according to the journal's guidelines.

Comment 7. "According to our previous study on iron deficiency, a higher donation frequency has been associated with reduced ferritin levels and an increased risk of iron deficiency in women." In my view you should elaborate more on the fact that reference 35 contains the ferritin and Hb measurements from this same donor population. It is not a previous study, but part of the same. I missed this point and was going to ask why you did not measure ferritin in your donors.

Response 7: Thank you for your insightful comments and for pointing out the need for further clarification regarding the ferritin and Hb measurements.

Reference 35 is indeed part of our team's previous work, which aimed to establish a predictive model for iron deficiency in plasmapheresis donors. As the ferritin measurement results were already published as part of our team's findings in that study, we did not measure ferritin levels again in the current study. Instead, we focused on building upon that foundation by exploring additional aspects related to donation frequency and its impacts.

Comment 8. "Second, the varying assay conditions between participating laboratories could be a confounding factor for Hb." As described in "methods" measures have been taken to reduce sample and analysis variation between laboratories. What variation in assay conditions may remain?

Response 8: Thank you for your valuable feedback and for pointing out the potential confounding factors related to varying assay conditions between participating laboratories.

As described in the "Methods" section, we have implemented several measures to reduce sample and analysis variation between laboratories. These measures include standardized protocols for sample collection, handling, and processing, as well as rigorous calibration and validation of equipment across all participating sites.

However, despite these efforts, some variation in assay conditions may still remain due to factors beyond our control. These factors could include slight differences in environmental conditions (such as temperature and humidity), minor variations in reagent batches, and human factors related to the operation of equipment. While we have minimized these potential sources of variation as much as possible, it is important to acknowledge that they can never be entirely eliminated in a multi-center study.

Reviewer 4

Dr. Rachel Thorpe, Clinical Services and Research, Australia Red Cross Lifeblood

Comment 1. Title:Should the running head be "Impact of plasma donation on protein and electrolytes"?

Response 1: Thank you for your valuable feedback on our manuscript. We have revised the running. The new running head is now: "Association of plasma donation and protein and electrolytes." We appreciate your attention to detail and believe that this revision has improved the clarity and appropriateness of our manuscript's running head.

Comment 2. Abstract:Objectives:This sentence is not clear – needs a full stop?

Conclusions:This section is currently unclear. The conclusions seem to contradict the results that found IgG levels were significantly lower in the high donation frequency group and that Hb is lower in female donors with high donation frequency and a high total number of lifetime donations. Can you suggest any recommendations to offset the reduction in IgG levels in male and female donors and in Hb in female donors?

Response 2: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comments regarding the clarity of the Objectives and Conclusions sections of the abstract.

For the Objectives section, we have revised the text for clarity as follows:

□Background: China's plasmapheresis donation policy differs from that of Western countries. The association of regular plasmapheresis donation and donor health in China is still unknown.

□Objectives: To investigate the association of regular plasmapheresis donation with serum protein and electrolyte levels and provide scientific evidence for policy improvement. (Page 2, Lines 32-35)

For the Conclusions section, we have revised the text to better align with the results and provide clear recommendations:

□Plasmapheresis donation is not associated with an increased risk of abnormalities in the analyzed parameters. However, the results provide preliminary evidence supporting the routine inclusion of IgG screening for donors, as plasmapheresis donation is associated with a decrease in IgG levels. Particular attention should be paid to the Hb levels of female donors, especially those who donate frequently. Testing of TSP at each donation may not be necessary. (Page 2, Lines 50-54)

We appreciate your attention to detail and believe that these revisions have improved the clarity and consistency of the abstract.

Comment 3. Introduction: This section should include a clear description of what plasmapheresis donation is, including what plasma-derived products are used for.

This section should also include a description of donor policies and procedures in China, such as whether donors are voluntary or remunerated, if remunerated what the rate is, whether donors can donate whole blood and plasma and platelets or just plasma and how donors come to donate plasma (eg is it because of their blood type, are they asked to, do they choose to).The guidelines for plasmapheresis donation in Australia are very similar to China so perhaps this could be pointed out - the only difference is that the maximum number of donations per year is 26 compared with 24 in China. Data from Australian plasma donors may serve as a useful comparison.

Could the authors extrapolate upon the points that there are concerns about the health of plasma donors, such as explaining why this is a growing concern.

The sentence "concerns about the health of plasma donors have risen" is a repeat of the previous sentence.

Response 3: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comments regarding the Introduction section. The specific changes made are as follows:

□Description of Plasmapheresis Donation and Donor Policies:

We have added a clear description of plasmapheresis donation, including the use of plasma-derived products and the donor policies and procedures in China. The revised text is as follows:

Blood and blood products are essential medicines for clinical use, saving millions of lives annually and being included in the World Health Organization's Model List of Essential Medicines. Plasma-derived



medicinal products (PDMPs), such as albumin, coagulation factors, and immunoglobulins, are prepared from human plasma and are crucial in preventing and treating a variety of life-threatening diseases. Source plasma (SP) is a vital raw material for PDMP production and is exclusively used for further manufacturing into final therapies through fractionation. In China, all SP is obtained through apheresis plasma donation. Plasma donors are required to undergo a health assessment and blood tests, and only those who meet the criteria are eligible to donate (Supplementary Table 1). (Page 3, Lines 69-76)

Supplementary Table 1. Plasmapheresis donor selection criteria in China.

Parameters Values

Age 18-60 years

Weight  $\geq 50$  Kg for males and  $\geq 45$  Kg for females

Blood pressure Systolic between 90 and 140 mm Hg vs. diastolic between 60 and 90 mm Hg vs. pulse pressure difference  $\geq 30$  mm Hg

Pulse Regular pulse between 60-100 beats per minute

Temperature Normal

Hb  $\geq 120$  g/L for males and  $\geq 110$  g/L for females

Serum/plasma protein Serum protein  $\geq 60$  g/L; plasma protein  $\geq 50$  g/L

Alanine aminotransferase (ALT)  $\leq 50$   $\mu$ /L for both males and females

Hepatitis B virus surface antigen (HBsAg) Negative

Hepatitis C virus antibody (HCVAb) Negative

HIV-1 and HIV-2 antibody Negative

Syphilis Negative

Donation interval 2 weeks

Serum/plasma electrophoresis Compared with the normal plasma electrophoretogram, the electrophoretogram bands of donors did not increase or decrease. Albumin content not less than 50%.

Temporary deferral Prospective plasmapheresis donor ineligible in donation for a time-limited period and can return for further donation

Permanent deferral Prospective plasmapheresis donor unable to donate forever for one or a variety of reasons

Concerns About Donor Health:

We have elaborated on the concerns about the health of plasma donors, particularly the impact of blood loss on physical health. The revised text is as follows:

Donation serves the demand for plasma, but there are concerns among potential donors and the public about the impact of continuous blood loss and anticoagulant exposure during plasmapheresis process on physical health. (Page 4, Lines 99-100)

□Removal of Repetitive Sentences:

We have removed the repetitive expression regarding the concerns about the health of plasma donors to improve clarity and avoid redundancy.

Comment 4. Study design and population: How were the participants approached for consent to participate in the research? Were participants provided with information about the study prior to consenting?

Response 4: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding how participants were approached for consent to participate in the research. The following information has been added to the Study design and population section:

Participants were approached for consent to participate in the research through direct communication during their donation visits. They were provided with comprehensive information about the study, including its purpose, procedures, potential risks, and benefits, prior to consenting. Written informed consent was obtained from all participants before their inclusion in the study. (Page 4, Lines 109-112)

Comment 5. How were the definitions of low, medium and high donation frequencies and number of lifetime donations arrived at?

Response 5: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding the definitions of low, medium, and high donation frequencies and the number of lifetime donations. The following information has been added to the Study design and population section:

Due to the allowance of up to 24 donations per year in China, we categorized regular donors based on their donation frequency in the previous 12 months into three groups: low (1 to 8 donations), medium (9 to 16 donations), and high (17 to 24 donations). Additionally, we divided regular donors into low (1 to 50 donations), medium (51 to 100 donations), and high (more than 100 donations) total number of lifetime donations groups based on a combination of the distribution within our study cohort and expert consensus to ensure a balanced distribution of participants across categories. (Page 4 and Page 5, Lines 114-119)

Comment 6. Inclusion and exclusion criteria: Second sentence – donors should be lower case. How was information about which supplements donors were taking obtained? How did you establish levels of blood lipids, uric acid and cholesterol in donors?

Response 6: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comments regarding the inclusion and exclusion criteria. The following clarifications have been added to the manuscript:

Participants were asked whether they had taken protein and/or electrolyte supplements in the past year prior to enrollment. (Page 5, Lines 129-130)

Additionally, donors with a self-reported chronic inflammatory syndrome or a history of metabolic diseases, including abnormal blood lipids, uric acid, and cholesterol, were also excluded. (Page 5, Lines 132-133)

Comment 7. Data collection: How was the questionnaire data obtained – was this through paper surveys or online? Can you include a description of the process for obtaining questionnaire data? What was the completion rate for the questionnaire?

Response 7: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comments regarding the data collection process. The following clarifications have been added to the manuscript:

Demographic information (living place, sex, age, female menstrual history), socioeconomic information (education, annual household income), and lifestyle variables (smoking, drinking, meat intake, physical activity) were collected through face-to-face interviews conducted by staff at each plasma donation centre using paper questionnaires. (Page 6, Lines 150-153)

Comment 8. Quality control: The sentence “Upon completion of the collection process, serum samples were promptly chilled and avoid repeated freezing and thawing” seems to have a typo in it.

Response 8: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding the sentence on the handling of serum samples. The revised sentence now reads:

Upon completion of the collection process, serum samples were promptly chilled and repeated freeze-thaw cycles were avoided. (Page 6, Lines 165-166)

Comment 9. Statistical analyses: Can you explain what is meant by “Literature-based covariates were identified by prior studies”?

Response 9: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding the explanation of “Literature-based covariates were identified by prior studies.” The intention was to convey that the selection of covariates was based on factors reported in previous literature as potentially related to protein and electrolyte metabolism. We have revised the sentence for clarity:

Covariates were selected based on factors reported in previous literature as potentially related to protein and electrolyte metabolism. (Page 7, Lines 180-181)

Comment 10. Results: Can you include numbers for all results discussed in the text, such as mean age, BMI, household income and physical activity.

Response 10: Thank you for your valuable feedback on our manuscript. We have carefully reviewed and addressed your comment regarding the inclusion of numbers for all results discussed in the text. We have added specific data for age and BMI, as they are important variables reflecting the characteristics of the population. Due to space constraints, we did not include specific results for household income and physical activity but noted the significant differences. The revised text is as follows:

The regular donors were older than new donors among males, with the median age of male new donors being 36 years (IQR 25-46) and that of regular donors being 41 years (IQR 31-50) ( $p < 0.001$ ). However, there was no significant difference in age between the two groups among females, with the median age of female new donors being 41 years (IQR 32-49) and that of regular donors being 42 years (IQR 32-50) ( $p = 0.102$ ). The BMI of regular donors was higher than that of new donors in both males and females (male:  $p = 0.002$ ; female:  $p = 0.018$ ), with the median BMI of male new donors being 24.48 (IQR 21.82-27.10) compared to 25.57 (IQR 22.65-28.39) for regular donors, and the median BMI of female new donors being 23.76 (IQR 21.48-26.59) compared to 24.60 (IQR 22.06-27.25) for regular donors. Significant differences were observed in annual household income and physical activity between new donors and regular donors ( $p < 0.05$ ). (Page 7, Lines 200-209)

Comment 11. Discussion: The structure of the discussion could be improved. I would suggest starting out by discussing your findings, noting what is novel about them and then comparing them to other studies. Currently it is very unclear for the reader to understand how your results compare with other studies as, for example, the findings for TSP from your participants are not mentioned in the text either in results or discussion and the reader has to scroll down to Table 2 to understand how they compare with international findings.

Response 11: Thank you for your valuable feedback on our manuscript. We have restructured the discussion to improve clarity and flow, starting with our findings, noting their novelty, and then comparing them to other studies. Specifically, we included our findings on TSP levels in both the results and discussion sections for better clarity.

The study found that mean TSP levels were not significantly associated with either the frequency of plasmapheresis donations or the total number of lifetime donations. (Page 9, Lines 254-255)

Comment 12. The recommendations for changes to testing for donors could be placed into context more by including details of the testing currently conducted in the introduction. The recommendation for increasing the frequency of albumin screening for donors is unclear. Is this suggested because albumin is naturally lower in older people? As such, is increased frequency of screening suggested only for middle-aged to older donors?

Response 12: Thank you for your valuable feedback on our manuscript. We have clearly states the current albumin (Alb) monitoring policy. Additionally, we clarified the recommendation for increasing

the frequency of Alb screening, particularly for older donors, in the discussion section. The revised text now reads:

According to Chinese guidelines, Alb should be measured every 12 months, and serum/plasma electrophoresis should be  $\geq 50\%$ , with no significant change from the previous time. Given the negative correlation between Alb levels and age, Alb monitoring before plasma donation should be adjusted based on the donor's age. For older donors, it is recommended to increase the frequency of Alb testing. (Page 10, Lines 270-274)

Comment 13. The discussion of findings for IgG is also unclear. Your results found significantly lower IgG levels in male and female donors compared with new donors but in the discussion it says "Although this study found no effect of plasma donation on low IgG rates, regular IgG monitoring of Chinese donors is still necessary given previous results that plasma donation affects IgG levels" – can this be explained more clearly.

Response 13: Thank you for your valuable feedback on our manuscript. We have addressed your comment regarding the discussion of findings for IgG by further clarifying our results. The revised text now reads:

Although this study found no significant association between plasmapheresis donation and the risk of IgG abnormality, it did find that IgG levels significantly decreased with increased donation frequency and the total number of donations. Therefore, regular monitoring of IgG levels in donors is still necessary. (Page 10, Lines 283-285)

Comment 14. The findings of the impact of plasmapheresis on Hb in women also warrants more discussion. What is recommended to overcome the drop in Hb in women? Further testing? Reduced donation frequency?

Response 14: Thank you for your valuable feedback on our manuscript. We have addressed your comment regarding the impact of plasmapheresis on Hb levels in women by providing further discussion and recommendations in the manuscript. The revised text now includes:

High-frequency female donors are advised to regularly take iron supplements to prevent and treat iron deficiency anemia. A diet rich in iron and vitamin C, including red meat, leafy green vegetables, and citrus fruits, is also encouraged to enhance iron absorption. Extending the interval between donations in female plasma donors with low Hb values is recommended. Additionally, regular monitoring of Hb levels is essential for timely interventions. (Page 11, Lines 298-302)