Dear Editor and Reviewers:

Thank you for giving us the opportunity to submit a revised draft of the manuscript "Serum ferritin levels in children with attention deficit hyperactivity disorder and tic disorder: A retrospective study based on a large Chinese sample" (Manuscript NO: 75511) for publication in the World Journal of Clinical Cases. We appreciate the time and effort that you and the reviewers dedicated to providing feedback on our manuscript. The comments were insightful and led to meaningful improvements to the paper.

- ***We have incorporated most of the suggestions made by the reviewers. Changes in the manuscript are in red font.***
- ***Below, we provide a point-by-point response to the reviewers' comments and concerns.***

Thank you for your consideration. I look forward to hearing from you.

Best regards and on behalf of my co-authors,

Reviewer 1:

please consider the following comments:

1.Introduction

- State specific objectives, including any prespecified hypotheses.

Response: Thank you very much for pointing out this issue. Following your suggestions, we have added specific objectives, including some prespecified hypotheses. For example, "It has been reported that iron deficiency in children is associated with neurodevelopmental diseases, leading to abnormalities in growth and development, learning behavior, motor function, social emotion, intellectual development, cognitive ability, language function, sleep cycle, etc. This study will explore the current status of iron deficiency in children with neurodevelopmental disorders and its influence on gender and age, providing an important reference for the correlation between neurodevelopmental disorders and ferritin and necessary iron supplementation." For more details, see the revised manuscript.

2、Method

- Clearly, define all outcomes, and potential confounders.

Response: Thank you very much for your comments. For the outcomes, we defined the standard of low serum ferritin as 15 μg/L in part 2.2 Methods for ferritin analysis.

For the potential confounding factors, it includes case selection bias, for which we strictly controlled the inclusion and exclusion criteria of the study subjects and used healthy children as controls. Another confounding factor is measurement bias. For this reason, we adopted the objective index of serum ferritin concentration and unified detection methods and strict quality control during blood collection. Finally, the mixed bias caused by sex and age on serum ferritin concentration was controlled by stratified analysis and regression analysis.

- Provide details of methods of assessment (measurement) in a separate part.

Response: Thank you very much for your suggestions. We have added details of the methods for ferritin analysis in a separate part. For example, "Serum ferritin was detected by a Beckman DXI800 automatic immunoelectrochemiluminescence analyzer and an original matching serum ferritin detection kit. After all children washed their hands with soap, 1 ml of venous blood was collected, and serum samples were separated for detection and tested by full-time staff in the biochemical room of the hospital testing center. The experimental process was strictly controlled by quality and tested in accordance with the operating procedures. The standard of low serum ferritin was 15 μg/L [23]."

- Explain who diagnosed the clients with ADHD, TD, and ASD.

Response: Thank you very much for your suggestions. We have added "The diagnosis procedures of these neurodevelopmental disorders(ADHD, TD and ASD) was carried out by professional psychiatrists."

- Describe any efforts to address potential sources of bias.

Response: Thank you very much for your kind suggestions. To avoid the bias of retrospective studies to the greatest extent, the inclusion criteria and exclusion criteria of research objects were restricted to narrow the differences between research objects. A case–control study was used to control for confounding factors. The age of confounding factors was stratified and then treated with corresponding statistical methods. Linear regression and logistic regression models were used to control for

confounding bias. We have added new section about these results. For example, "In the liner regression using the fitting least square method, ADHD (β=-0.110，P<0.001) and TD (β=-0.114，P<0.001) were both associated with lower level of SF even after age and sex (see Table3)."

- Explain how the study size was arrived at

Response: Thank you very much for your kind suggestion here. For the procedures to calculate the sample size. Our sampling method is convenience sampling, which is the limitation of our research method and will be explained in the limitations later.

- Describe statistical methods used to control for confounding.

Response: The age of confounding factors was stratified and then treated with corresponding statistical methods. To control for confounding factors of sex and age, we adopted linear regression and logistic regression models.

- Describe any methods used to examine subgroups and interactions.

Reply: We used analysis of variance to examine subgroups and interactions.

- Explain how missing data were addressed

Response: Thank you very much for your suggestion here. In this study, cases with missing data were eliminated. Studies indicate that a missing rate of 5% or less is inconsequential (Schafer et al., 1999), and statistical analysis is likely to be biased when more than 10% of data are missing (Bennett et al., 2001). That is why we do not include the missing data. In this study, the missing rate was less than 5%.

- Transfer the Data Extraction part information to the Results section.

Response: We have transferred the Data Extraction section to the Results section.

3、Results

- Give characteristics of study participants (e.g., demographic) in a Table and present number of participants with missing data for each variable.

Response: Thank you very much for your comments. In the revised version, we have given the characteristics of the study participants in a table and present the number of participants with missing data for each variable.

- Use Regression analysis according to gender and age.

Response: Thanks very much. In the revised version, we have added new analysis of regression analyses based on sex and age.

- Give unadjusted estimates and their precision (e.g., 95% confidence interval).

Response: Thank you very much for your comments. In the regression analysis, we have added the 95% confidence interval.

Make clear which confounders were adjusted for and why they were included.

Response: Thank you very much for your kind suggestion. Gender, age and disease type were confounders of ferritin concentration, so linear regression and logistic regression were used for adjustment.

- Enter data of clients with ASD in all Tables and text.

Response: Thank you very much for your kind suggestion. We have added the ASD data into all some tables and text as well as the related statements. However, due to the small sample content and age dispersion of THE ASD group, it is not suitable for an OVA and regression analysis of subgroups, so ASD data are still not included in the above two statistical methods

- Use Variance analysis to compare groups.

Response: We have added variance analysis to compare these groups. The new results showed that ferritin levels in different age groups of different disease groups were statistically significant.

4、Discussion

- Update the Discussion section based on using new analysis.

Response: We have updated the discussion section with new statistical analysis results. For example, "Analysis of variance was used for comparisons between subgroups, and regression analysis was used for confounding factor control."

- Discuss both direction and magnitude of any potential bias.

Response: Thank you very much for your comments. We have added new statements about these contents. For example, "Some limitations of our study need to be pointed out here. First, neurodevelopmental disorders were studied from outpatient cases, and only individuals who used medical resources to seek psychiatric care were identified. There may be some selection bias in the sample, however, the patients included in our study were diagnosed by professional psychiatrists, and the diagnoses were more reliable than self-reported diagnoses. Second, serum ferritin concentrations may be due to inappropriate dietary habits, and the link between neurodevelopmental disorders and altered dietary patterns remains unclear. Third, we did not have personal information that would help us understand patients' risk of mental disorders, such as environmental factors (such as long-term life stress, traumatic experiences) and a family history of mental disorders. Fourth, our study cannot prove a causal relationship between serum ferritin low and neurodevelopmental disease, although our results suggest a significant association between neurodevelopmental disease and serum ferritin.".

- Discuss the generalizability (external validity) of the study results.

Response: Thank you very much for your kind suggestion here. We have added new statements about this point. For example, "Some limitations of our study need to be pointed out here. First, neurodevelopmental disorders were studied from outpatient cases, and only individuals who used medical resources to seek psychiatric care were identified. There may be some selection bias in the sample, however, the patients included in our study were diagnosed by professional psychiatrists, and the diagnoses were more reliable than self-reported diagnoses. Second, serum ferritin concentrations may be due to inappropriate dietary habits, and the link between neurodevelopmental disorders and altered dietary patterns remains unclear. Third, we did not have personal information that would help us understand patients' risk of mental disorders, such as environmental factors (such as long-term life stress, traumatic experiences) and a family history of mental disorders. Fourth, our study cannot prove a causal relationship between serum ferritin low and

neurodevelopmental disease, although our results suggest a significant association between neurodevelopmental disease and serum ferritin."

- Overall, the analysis of the Result section should be modified, please consult with an expert in analysis of data and perform it again.

Response: In general, we have consulted data analysis experts for reanalysis and modification of the Results section. We also added further analysis of the data. For more details, see the revised manuscript.

Reviewer #2:

some more references should be added.

Response: Thank you very much for your suggestions. We have added 4 new references. For example,

25. ***Topal Z, Tufan AE, Karadag M, Gokcen C, Akkaya C, Sarp AS, Bahsi I, Kilinc M: Evaluation of peripheral inflammatory markers, serum B12, folate, ferritin levels and clinical correlations in children with autism spectrum disorder (ASD) and attention deficit hyperactivity disorder (ADHD). Nord J Psychiatry 2022, 76(2):150-157.***

26. ***Daniel A. Gorman MD, Hongtu Zhu, Ph.D., George M. Anderson, Ph.D., Mark Davies, M.P.H., and Bradley S. Peterson, M.D.: Ferritin Levels and Their Association With Regional Brain Volumes in Tourette's Syndrome. Am J Psychiatry 2006, 163(7):1264–1272.***

27. ***De Giacomo A, Medicamento S, Pedaci C, Giambersio D, Giannico OV, Petruzzelli MG, Simone M, Corsalini M, Marzulli L, Matera E: Peripheral Iron Levels in Autism Spectrum Disorders vs. Other Neurodevelopmental Disorders: Preliminary Data. Int J Environ Res Public Health 2022, 19(7).***

32. ***Degremont A, Jain R, Philippou E, Latunde-Dada GO: Brain iron concentrations in the pathophysiology of children with attention deficit/hyperactivity disorder: a systematic review. Nutr Rev 2021, 79(5):615-626.***