PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Association between fasting blood glucose and outcomes and mortality in acute ischemic stroke patients with diabetes mellitus: a retrospective observational study in Wuhan, China
AUTHORS	Yao, Tao; Zhan, Yanqiang; Shen, Jing; Xu, Lu; Peng, Bo; Cui, Qi; Liu, Zhichao

VERSION 1 – REVIEW

REVIEWER	Jiming Fang ICES Canada
REVIEW RETURNED	13-Feb-2020

GENERAL COMMENTS	The authors use a single-center dataset to look at the association between fasting blood glucose (FBG) and stroke outcomes. I enjoyed reading this well written paper but felt some aspects would benefit from greater detail. Statistical Analysis The authors adjusted FBG and other numeric variables as continuous variables in the logistic regression models, which assuming there were linear relationships between these numeric variables and the outcomes. Suggest the authors to test linearity for the continuous variables, FBG, HbA1c, NIHSS, BMI, SBP and DBP, using restrict cubic splines approach before conducting the logistic regression models. Table 1 HbA1c was missing in non-DM patients. Please add FGB data, mean, median (IQR) Table 2 It appears that the ischemic DM patients with higher HbA1c was less likely to have unfavorable functional outcome (OR 0.843 (95%CI: 0.707-1.005)), but more likely to die (OR 1.486 (95%CI: 1.227-1.779)) than those with lower HbAc1. Please interpret these results. Figure 5 Change the y-axis label at “Survival Probability” instead of “Cumulative Survival”. Add the number of patients at risk at the given time points, 0 day, 20 day, etc. References Many references did not show the last names, but initials only. Please revise.

REVIEWER	Diederik Dippel Erasmus University Medical Center Rotterdam, the Netherlands
REVIEW RETURNED	24-Feb-2020

GENERAL COMMENTS	This is an interesting study of the association of blood glucose levels in patients with acute ischemic stroke with outcome : poor outcome (mRS 3-6) and death (mRS 6). Major: Abstract 1) Were these patients consecutively admitted and included in the study? If yes, that would make the results easier to interpret and to apply. I suspect the cohort was not consecutive considering the point that the number of patients with DM far exceed the number of patients without. 2) What was being measured? fasting blood glucose, as mentioned in the design section of the abstract, or acute blood glucose level as mentioned in the objective. I would prefer to have both and compare the predictive value of the two.that would be new information. 3) In the results section of the abstract, you report that “elevated FBG” was associated with an increased NIHSS score. How did you define elevated FBG? 4) Same question for high FBG in the abstract. It is unclear how I should interpret the odds ratio you report. Strengths and limitations section I do not see why absolute no of men and women should be “balanced”. I would rather have that you included consecutive patients, to make an adequate representation of the distribution of baseline characteristics in the population of acute ischemic stroke patients. Introduction No comments Methods 5) it now becomes clear to this reviewer that you included patents without DM over a such shorter time period. This makes the study more difficult to interpret and apply to clinical practice as patients with a poor prognosis because of DM are overrepresented. 6) were research nurses who assessed outcome at 90 days aware of the DM and FBG status of the patients, and of the purpose of the present study. They do not need to have been blinded to this information , but just not knowing the purpose of the study may have increased their objectivity in assessing outcome. 7) the information about FBG timing should be included in the abstract, as well as the time window for inclusion of patients. 8) the information about how you analyzed FBG (in quartiles) should be in the abstract. However, did you explore other approaches to FBG, for example as a continuous factor, perhaps as mmol/L exceeding a certain threshold around 6-7 mMol/L. Quartiles are very data driven. 8) was the diagnosis of DM known at presentation, or afterward, this is important for applicability of the results in practice. I suspect that in quite a few patients this may not have been known at admission. 9) it would have been very helpful to understand the relation between FBG and outcome if the authors could provide additional

	information about intermediate outcomes and adverse events that may contribute to the effect of FBG on outcome: pneumonia, sepsis 9) The final conclusion, that knowledge of the association of FBG with outcome will facilitate management of patients with IAS is difficult to follow; how would this help me? I would rather like to read something like; our data contribute to the knowledge about the relation between FBG and outcome in AIS. Minor It is multivariable, not multivariate analysis.1 I would not name the patients without DM controls. This is not a controlled study, nor is it a case control study. You report odds ratios with 3 decimals three one or two would suffice. In conclusion: I like the paper, but I do not see why a small cohort of patients without DM was added. The validity of the findings would increase when a consecutive cohort of AIS patients was analyzed. The paper would be more valuable when admission glucoses and FBG glucoses would have been available and compared with respect to their prognostic value. Instead of quartiles, a more advanced analysis of blood glucose a continuous variable would be more informative. Reference 1. Hidalgo B, Goodman M. Multivariate or multivariable regression? Am J Public Health. 2013;103:39-40

REVIEWER	Sohei Yoshimura National Cerebral and Cardiovascular Center, Japan
REVIEW RETURNED	27-Feb-2020

GENERAL COMMENTS	Authors evaluated the predictive power of a FBG level on unfavorable outcomes and mortality in DM patients after acute ischemic stroke using population-based, retrospective observational study. Although this manuscript was well written, several concerns can be found. Authors may want to resolve these issues as follows. #A number of studies have shown that (random) blood glucose level at admission are related to functional outcome of AIS patients (without DM). Thus, the point of this study is generalization of this evidence to the AIS patients with DM by using FBG. I recommend to compare the predictive power of FBG for outcomes between AIS patients with and without DM. If possible, the power should be compare between FBG and random blood glucose.

VERSION 1 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 1

Reviewer Name: Jiming Fang

Institution and Country: ICES

Canada

Please state any competing interests or state ‘None declared’: None declared

Please leave your comments for the authors below

The authors use a single-center dataset to look at the association between fasting blood glucose (FBG) and stroke outcomes. I enjoyed reading this well written paper but felt some aspects would benefit from greater detail.

Statistical Analysis

The authors adjusted FBG and other numeric variables as continuous variables in the logistic regression models, which assuming there were linear relationships between these numeric variables and the outcomes. Suggest the authors to test linearity for the continuous variables, FBG, HbA1c, NIHSS, BMI, SBP and DBP, using restrict cubic splines approach before conducting the logistic regression models.

Thank you for your advice on statistical analysis. Following your opinion, we did test linearity, and the results showed a statistically significant linear relationship between continuous variables including FBG, HbA1c, NIHSS, BMI, SBP, DBP, and outcomes. I very much regret that we did not use the statistical method of restrict cubic splines that you mentioned. We simply do not have that ability. Actually, we usually use common logistic regression analysis in our studies . I have reviewed a lot of literature, and I even sought help from many of my colleagues. Unfortunately, none of these efforts were helpful. In a future study, I will try my best to learn the statistical method you mentioned.

Table 1

HbA1c was missing in non-DM patients.

Please add FGB data, mean, median (IQR)

Thanks for your reminding us. We have added these data to the Table.

Table 2

It appears that the ischemic DM patients with higher HbA1c was less likely to have unfavorable functional outcome (OR 0.843 (95%CI: 0.707-1.005)), but more likely to die (OR 1.486 (95%CI: 1.227-1.779)) than those with lower HbAc1. Please interpret these results.

Thank you very much for your question. We think the main reason may be the existence of simultaneous or interactive effects for the two glycemic indexes, which are not clear. The impact of glucose level in the acute stage of ischemic stroke might vary between different HbA1c statuses [1]. Thus, investigators did not include HbA1c as an influencing factor in the logistic regression analysis in many studies on the relationship between blood glucose and AIS prognosis [2-5]. Masrur et al. performed logistic regression analysis of blood glucose and HbA1c separately in the AIS patient study to avoid the interaction between the two indices [6].

For those reasons, we did not include HbA1c as a factor in our initial analysis. However, after a submission experience, we later included HbA1c in the analysis according to the requirements of one of the reviewers. We wondered whether we should include HbA1c as a factor.

Reference

[1] Lee Keon-Joo,Lee Ji Sung,Jung Keun-Hwa,Interactive effect of acute and chronic glycemic indexes for severity in acute ischemic stroke patients.[J] .BMC Neurol, 2018, 18: 105.

[2] Osei E,den Hertog H M,Berkhemer O A et al. Increased admission and fasting glucose are associated with unfavorable short-term outcome after intra-arterial treatment of ischemic stroke in the MR CLEAN pretrial cohort.[J] .J. Neurol. Sci., 2016, 371: 1-5.

[3] Goyal Nitin,Tsivgoulis Georgios,Pandhi Abhi et al. Admission hyperglycemia and outcomes in large vessel occlusion strokes treated with mechanical thrombectomy.[J] .J Neurointerv Surg, 2018, 10: 112-117.

[4] Huo Xiaochuan,Liu Raynald,Gao Feng et al. Effect of Hyperglycemia at Presentation on Outcomes in Acute Large Artery Occlusion Patients Treated With Solitaire Stent Thrombectomy.[J] .Front Neurol, 2019, 10: 71.

[5] Osei E,den Hertog H M,Berkhemer O A et al. Increased admission and fasting glucose are associated with unfavorable short-term outcome after intra-arterial treatment of ischemic stroke in the MR CLEAN pretrial cohort.[J] .J. Neurol. Sci., 2016, 371: 1-5.

[6] Masrur Shihab,Cox Margueritte,Bhatt Deepak L et al. Association of Acute and Chronic Hyperglycemia With Acute Ischemic Stroke Outcomes Post-Thrombolysis: Findings From Get With The Guidelines-Stroke.[J] .J Am Heart Assoc, 2015, 4: e002193.

Figure 5

Change the y-axis label at “Survival Probability” instead of “Cumulative Survival”.

Add the number of patients at risk at the given time points, 0 day, 20 day, etc.

We have modified Figure 5 according to your requirements and added the number of patients at risk at the given time points: 0, 20 days, 40 days, 60 days, and 90 days.

References

Many references did not show the last names, but initials only. Please revise.

I apologize for this mistake. We have corrected the references according to the format of *BMJ Open*.

Reviewer: 2

Reviewer Name: Diederik Dippel

Institution and Country: Erasmus University Medical Center Rotterdam, the Netherlands

Please state any competing interests or state 'None declared': None Declared

Please leave your comments for the authors below

This is an interesting study of the association of blood glucose levels in patients with acute ischemic stroke with outcome : poor outcome (mRS 3-6) and death (mRS 6).

Major:

Abstract

1) Were these patients consecutively admitted and included in the study? If yes, that would make the results easier to interpret and to apply. I suspect the cohort was not consecutive considering the point that the number of patients with DM far exceed the number of patients without.

Yes, the patients were consecutively admitted and included in the study, as stated in *Patients and Study Design*. The manuscript states in the *Methods* section as follows:

1. This retrospective observational study collected information involving AIS patients with DM who were admitted to the Department of Neurology of the Renmin Hospital of Wuhan University from January 2018 to June 2019.

2. In this study, the same basic information and clinical data of AIS patients without DM admitted to the same hospital from January to June 2018 were also collected as a control group.

The number of patients with DM far exceeded the number of patients without DM.

In addition, the number of AIS patients without DM may have been more than the AIS patients with DM. Since the subject of our study was AIS patients with DM, we did not collect data on all patients without DM at the corresponding period.

2) What was being measured? fasting blood glucose, as mentioned in the design section of the abstract, or acute blood glucose level as mentioned in the objective. I would prefer to have both and compare the predictive value of the two.that would be new information.

Thank you for your careful review. In previous studies, both the random blood glucose and fasting blood glucose on admission were used as acute glycemic indexes to estimate acute glycemic status in ischemic stroke patients [1-3]. I regret that our language was not clear. The data of fasting blood glucose at the admission were collected as we stated in the *Title* and *Abstract* of our study. We used fasting blood glucose as acute glycemic indexes in our study.

Moreover, this is a retrospective observational study. Quite a few of the subjects were AIS patients with a history of DM, many of them did not receive random blood glucose monitoring at the time of admission, only fasting blood glucose. Therefore, we could not explore and compare the effects of random blood glucose and fasting blood glucose on the prognosis as you mentioned. This is a limitation as a retrospective study, which we mentioned in the new manuscript.

Reference

[1] Lee Keon-Joo,Lee Ji Sung,Jung Keun-Hwa,Interactive effect of acute and chronic glycemic indexes for severity in acute ischemic stroke patients.[J] .BMC Neurol, 2018, 18: 105.

[2] Wang Feng,Jiang Beisi,Kanesan Lasheta et al. Higher admission fasting plasma glucose levels are associated with a poorer short-term neurologic outcome in acute ischemic stroke patients with good collateral circulation.[J] .Acta Diabetol, 2018, 55: 703-714.

[3] Sung Jia-Ying,Chen Chin-I,Hsieh Yi-Chen et al. Comparison of admission random glucose, fasting glucose, and glycated hemoglobin in predicting the neurological outcome of acute ischemic stroke: a retrospective study.[J] .PeerJ, 2017, 5: e2948.

3) In the results section of the abstract, you report that “elevated FBG” was associated with an increased NIHSS score. How did you define elevated FBG?

I am sorry for the confusion. After serious consideration of your question, I think the following sentence, " Higher fasting blood glucose was associated with higher NIHSS score" may be more appropriate and more accurate. I respect the opinions of editors and I am seeking a professional copyediting service for my manuscript.

4) Same question for high FBG in the abstract. It is unclear how I should interpret the odds ratio you report.

Thank you for your careful review and beneficial reminder. After reading your comments, I regret that my manuscript has many language problems. We actually meant “higher.”

Strengths and limitations section

I do not see why absolute no of men and women should be “balanced”. I would rather have that you included consecutive patients, to make an adequate representation of the distribution of baseline characteristics in the population of acute ischemic stroke patients.

Thank you very much for your comments. The reason I described sex imbalance in the manuscript as a limitation of this article is that in another journal submission, one of the reviewers emphasized the sex imbalance in our research and asked me to describe it as a limitation in the manuscript. As a matter of fact, I could not agree with his point of view. Like many retrospective studies, sex balance is completely unnecessary. According to your opinion, I have deleted the description of sex imbalance from the manuscript.

Introduction

No comments

Methods

5) it now becomes clear to this reviewer that you included patents without DM over a such shorter time period. This makes the study more difficult to interpret and apply to clinical practice as patients with a poor prognosis because of DM are overrepresented.

Thank you for your pertinent point. Our study is not a case control study, and the main purpose is to investigate the predictive value of FBG in AIS patients with DM when compared to those without DM. This group of AIS patients without DM has no practical significance in this study. If you agree, we would delete this part of the description and data. The control study of AIS with and without DM could be carried out in our future studies.

6) were research nurses who assessed outcome at 90 days aware of the DM and FBG status of the patients, and of the purpose of the present study. They do not need to have been blinded to this information , but just not knowing the purpose of the study may have increased their objectivity in assessing outcome.

No, they were not aware of the DM and FBG status of the patients. Clinical data including FBG, HbA1c, and NIHSS score on admission were collected and evaluated by neurologists. The specially trained neurologic nurses were only responsible for assessing outcomes.

7) the information about FBG timing should be included in the abstract, as well as the time window for inclusion of patients.

Thank you very much for your suggestion. We will add this information to the abstract of the new manuscript

8) the information about how you analyzed FBG (in quartiles) should be in the abstract. However, did you explore other approaches to FBG, for example as a continuous factor, perhaps as mmol/L exceeding a certain threshold around 6-7 mMol/L. Quartiles are very data driven.

Thank you for this suggestion. We have added the information about the quartiles in the abstract of new manuscript. FBG was actually analyzed as a continuous factor in our univariable and multivariable logistic regression analysis. Our current studies mostly adopt some commonly used statistical methods of previous references. We regret for the lack of knowledge of statistical skills, so we will enhance them in future research

9) was the diagnosis of DM known at presentation, or afterward, this is important for applicability of the results in practice. I suspect that in quite a few patients this may not have been known at admission.

Thank you for your careful review. I may not have expressed it clearly enough in the section ***Patients and Study Design.*** There is no doubt that DM was defined as patients with a history of DM before admission according to their medical records or those who received drugs or insulin for hypoglycemic treatment after admission. We will express this more clearly in the revised manuscript. As you mentioned, some patients were admitted without knowing they had diabetes; however, all of them were included in the study after we reviewed their hypoglycemic treatment in the medical records.

10) it would have been very helpful to understand the relation between FBG and outcome if the authors could provide additional information about intermediate outcomes and adverse events that may contribute to the effect of FBG on outcome: pneumonia, sepsis

We have already mentioned some of the information what you suggest in the original manuscript. We stated that in this section ***Baseline Characteristics of the Study Population*** as follows: “There were 226 patients (39.8%) with unfavorable outcomes, including 58 deaths (10.2%). Of the 58 deaths in this study, 36 (62.1%) died of increased intracranial pressure, 10 (17.2%) died of cardiac diseases such as heart failure, myocardial infarction or arrhythmia, and 12 (20.7%) died of other causes such as severe pneumonia, stress ulcer bleeding and pulmonary embolism. Moreover, 14 (24.1%) had symptomatic intracerebral hemorrhages among the 58 deaths.”

11) The final conclusion, that knowledge of the association of FBG with outcome will facilitate management of patients with IAS is difficult to follow; how would this help me? I would rather like to read something like; our data contribute to the knowledge about the relation between FBG and outcome in AIS.

Thank you! I think your expression is more appropriate. I have modified the conclusion according to your suggestion.

Minor

It is multivariable, not multivariate analysis.1

I would not name the patients without DM controls. This is not a controlled study, nor is it a case control study.

You report odds ratios with 3 decimals three one or two would suffice.

According to your suggestion, we have changed the odds ratios to 2 decimals. Furthermore, with regard to “This is not a controlled study, nor is it a case control study,” we fully agree with you, and will delete the description and data of the patients without DM.

In conclusion:

I like the paper, but I do not see why a small cohort of patients without DM was added. The validity of the findings would increase when a consecutive cohort of AIS patients was analyzed. The paper would be more valuable when admission glucoses and FBG glucoses would have been available and compared with respect to their prognostic value. Instead of quartiles, a more advanced analysis of blood glucose a continuous variable would be more informative.

Reference

Hidalgo B, Goodman M. Multivariate or multivariable regression? Am J Public Health. 2013;103:39-40

Thank you for your careful review and beneficial comments. As I mentioned above, I quite agree with you that we would delete the control group from the revised manuscript. Secondly, this is a consecutive cohort study of AIS patients with DM. The poor expression of English language has led to a lot of confusion. We have revised the manuscript with the help of a professional copyediting service. Finally, we will upload a new manuscript and we hope that you could once again offer your valuable suggestions.

Reviewer: 3

Reviewer Name: Sohei Yoshimura

Institution and Country: National Cerebral and Cardiovascular Center, Japan

Please state any competing interests or state ‘None declared’: None declared

Please leave your comments for the authors below

Authors evaluated the predictive power of a FBG level on unfavorable outcomes and mortality in DM patients after acute ischemic stroke using population-based, retrospective observational study. Although this manuscript was well written, several concerns can be found. Authors may want to resolve these issues as follows.

#A number of studies have shown that (random) blood glucose level at admission are related to functional outcome of AIS patients (without DM). Thus, the point of this study is generalization of this evidence to the AIS patients with DM by using FBG. I recommend to compare the predictive power of FBG for outcomes between AIS patients with and without DM. If possible, the power should be compare between FBG and random blood glucose.

Thank you very much for your careful review and beneficial suggestion.

Unfortunately, we could not compare the predictive power of FBG for outcomes between AIS patients with and without DM. Due to our original retrospective design, this study was actually a cohort study of AIS patients with DM, not a controlled study. We initially included a control group to compare the clinical and laboratory characteristics of the two groups. However, because the main subject was AIS patients with DM, we did not follow up on AIS patients without DM. Based on your comments and those of the other reviewer, I now think that this control group of AIS patients without DM has no practical significance in this study. If you agree, we would delete this part of the description and data. The control study of AIS with and without DM could be carried out in our future studies.

Moreover, quite a few of the subjects were AIS patients with a history of DM, so many of them did not receive random blood glucose monitoring at the time of admission, but underwent only fasting blood glucose measurement. Therefore, we could not explore and compare the effects of random blood glucose and fasting blood glucose on the prognosis as you mentioned. This is also a limitation of a retrospective study, which we have now included in the manuscript.

Finally, we will upload a revised manuscript and we hope that you could once again offer your valuable suggestions.

VERSION 2 – REVIEW

REVIEWER	Jiming Fang ICES, Canada
REVIEW RETURNED	23-Apr-2020

GENERAL COMMENTS	Abstract

	This was hospital-based observational cohort study, but not "population-based". Please ask authors to make the change. Result - Baseline characteristic of the study population Please change "population" as cohort Table 2 Death is time-to-event outcome as showed in Figure 4. If the death date data are collected, suggest authors to report Hazard Ratio from Cox model instead of Odds ratio from logistic model on Table 2.

REVIEWER	Diederik Dippel Erasmus MC - University Medical Center Rotterdam The Netherlands
REVIEW RETURNED	02-Apr-2020

GENERAL COMMENTS	Thank you for your response I have no further comments.

REVIEWER	Sohei Yoshimura National Cerebral and Cardiovascular Center, Japan
REVIEW RETURNED	30-Mar-2020

GENERAL COMMENTS	Authors correctly responded to reviewers comments. I have no further comments.

VERSION 2 – AUTHOR RESPONSE

Reviewer: 1
Reviewer Name: Jiming Fang
Institution and Country: ICES, Canada
Please state any competing interests or state ‘None declared’: None declared

Abstract
This was hospital-based observational cohort study, but not "population-based". Please ask authors to make the change.

Thank you for your suggestion. We have changed this in the revised manuscript accordingly.

Result - Baseline characteristic of the study population
Please change "population" as cohort

Thank you for your suggestion. We have changed it in the revised manuscript accordingly.

Table 2
Death is time-to-event outcome as showed in Figure 4. If the death date data are collected, suggest

authors to report Hazard Ratio from Cox model instead of Odds ratio from logistic model on Table 2.

Thank you for your suggestion. I conducted statistical analysis again according to your requirements and changed the logistic model to Cox regression analysis for analysis of death. We have uploaded the new table of analysis results in the revised manuscript.

JF: Please report the correlation coefficient between FBG and HbA1c. If it is high, suggest authors not to adjust both in the multivariable regression model in order to avoid the collinearity.

Thank you for your valuable opinion. The results of linear correlation analysis suggest that the correlation coefficient is 0.253 ($p<0.001$), which show that the correlation was not particularly high. It may be feasible to adjust both in the multivariable regression model. If you agree with me, please give further indication.

JF: Authors replied that they did linearity test but did not mention which statistical method they used. Please indicate which statistical method was used to test linearity in this section.

Thanks for your reminding, we have added the description of statistical method in the section "Statistical Analysis."

Reviewer: 2
Reviewer Name: Diederik Dippel
Institution and Country: Erasmus MC - University Medical Center Rotterdam The Netherlands
Please state any competing interests or state 'None declared': none declared

Thank you for your response I have no further comments.

Reviewer: 3
Reviewer Name: Sohei Yoshimura
Institution and Country: National Cerebral and Cardiovascular Center, Japan
Please state any competing interests or state 'None declared': None declared

Authors correctly responded to reviewers comments. I have no further comments.

Thanks for the valuable comments of Pr. Diederik Dippel and Pr. Sohei Yoshimura.